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GRADUATE COLLEGE

DEVELOPMENT, ANALYSIS AND USE OF AN EXPRESSED SEQUENCE TAG
DATABASE FROM THE MULTICELLULAR ASCOMYCETE, *ASPERGILLUS*
NIDULANS

A Dissertation

SUBMITTED TO THE GRADUATE FACULTY

in partial fulfillment of the requirements for the

degree of

Doctor of Philosophy

By

DORIS MARIE KUPFER

Norman, Oklahoma

1999

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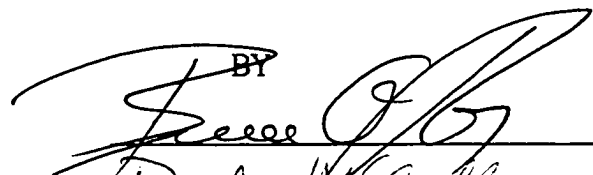
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DATABASE FROM THE MULTICELLULAR ASCOMYCETE, *ASPERGILLUS*
NIDULANS

A Dissertation APPROVED FOR THE
DEPARTMENT OF BOTANY AND MICROBIOLOGY

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Eunice Bird Kupfer 1918-1999

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Abstract

A. nidulans is a well-studied model multicellular ascomycete closely related to both human and plant fungal pathogens. To investigate the expressed genes in this model fungus, a cDNA library containing clones from vegetatively and asexually expressed genes was sequenced from both the 3' and 5' ends. These cDNA end sequences called expressed sequence tags or ESTs reveal genes which were expressed at the time of mRNA harvest. The 3' ESTs were aligned using the multiple sequence assembly program Phrap to follow the level of sequence redundancy and to measure the percent of new genes detected during the sampling of the cDNA library. A Unigene database representing approximately 3200 genes was generated after assembly of the 3' and 5' ESTs using Phrap to cluster the EST sequences which were generated from the same gene. The Unigene sequences were examined for homologs in GenBank using the Blast alignment program and organized into groups based on a Biological Function Classification schema. Almost 56% of the Unigene database members had no homologs in GenBank and thus represented newly discovered genes. The Unigene database also was useful in annotating three *A. nidulans* chromosome VIII cosmids and a 60 Kbp region from chromosome IV containing the sterigmatocystin gene cluster. Through these studies, an average gene density of 1 gene/ 2.6 Kbp was obtained and a total number of 12,000 genes was predicted for the *A. nidulans* genome.

Since these ESTs are a rich resource of expressed gene sequences useful to the large community of fungal researchers, the resulting EST database was made publicly available by submission to GenBank and placement on the ACGT website. In addition, the cDNA clones were submitted to the Fungal Genetics Stock Center, University of Kansas Medical Center, Kansas City. These ESTs will provide a sequence and clone source for hybridization probes for *A. nidulans* expressed genes as well as for related fungi with industrial, agricultural and health application.

Chapter 1

Introduction

1.1 Overview

During the course of this study an *Aspergillus nidulans* expressed sequence tag database (EST) of 4.268 Mbp and a total of 12,485 EST sequences was created. Improved protocols for optimizing template production and data handling and analysis also were developed during this work. A Unigene database, which uses multiple sequence alignments to take advantage of clone redundancy, extended the quality and length of the single-pass EST sequences to produce a series of consensus sequences. This resulting Unigene database was useful for finding gene families, determining expression levels of detected genes, creating a biological function overview of the organism, and for annotating sequenced genomic DNA.

1.2 EST Background

Messenger RNA is the transcribed intermediate in the protein biosynthetic pathway of DNA to RNA to proteins. The discovery of reverse transcriptase and the ability to isolate mRNA has allowed the generation of a stable DNA copy of the less stable mRNA called a copy-DNA or cDNA. These cDNAs are a copy of processed expressed gene sequences since they lack introns. The mature mRNA from which they are produced provides the open reading frame for the translation product that includes both the 5' start and 3' stop codons. There is additional information such as the position of the ribosome binding site in the 5' untranslated region and the polyadenylation signal in the 3' untranslated region.

The value of cDNA sequences for annotation of genomic regions was realized early in the human genome project (Adams, 1991). The presence of variable-sized introns as well as repeat sequences has made annotation of human genomic sequences

difficult. To aid in annotating the human genome, a large number of tissue specific cDNA libraries was prepared by The Institute for Genome Research (Adams, 1991) and in conjunction with Merck and Company, at Washington University, St. Louis (Hillier, 1996). Since partial cDNA sequences could identify and tag coding regions of genomic sequence, high quality, full-length cDNA sequences were found to be unnecessary. This realization led to an end-sequencing only approach and the generation of single-pass sequences, termed Expressed Sequence Tags (ESTs), from the ends of the cDNA templates (Adams, 1995). These sequences provide 1x or less coverage of a cDNA and are useful even though considered error prone (Marra, 1999).

Presented below is an overview of the large scale sequencing methods which have been used during this dissertation research to generate an EST database for the multicellular ascomycete fungus, *Aspergillus nidulans*. The computer-based methods developed to efficiently handle the large number of sequences generated will be outlined, as will the approaches taken for optimal use of the ESTs as a gene rich information source for annotation of genomic regions. These studies have provided a window into the classes of genes and the level of their expression in *A.nidulans*.

1.3 EST Database

ESTs represent sequences of expressed genes that are generated from both the 3' and 5' ends of randomly selected cDNAs (Adams, 1991). An interest in genome studies led to the realization that these single-pass sequences could be rapidly generated, and then used for gene discovery, constructing physical maps, studying expression patterns, and as an aid in sequence interpretation in large-scale genome projects (Hillier, 1996). There are growing numbers of human and other eucaryotic ESTs in a separate database at GenBank, dbEST (Benson, 1996).

Based on the intrinsic and extrinsic approaches developed by Borodovsky et al. (Borodovsky, 1994), which incorporate EST information into the analysis of completed

genomic sequences as a means of improving gene identification, potential exons can be predicted with great certainty using tools such as GeneMark (Borodovsky, 1994) since EST matches then can be used to verify the predictions. Thus, an EST database for a target organism is extremely useful as it complements the gene prediction algorithms based on ORFs with conserved characteristics such as codon usage and intron-exon borders. Those predicted exons which have an EST match are verified as such. Even though an EST is only a partial sequence of a cDNA, it is extremely useful for indicating the correct position of sequence landmarks such as transcription start, translation start and stop, intron-exon borders, and polyadenylation signals of the predicted genes. This information helps in predicting correct reading frames and eventually aids the construction of accurate physical maps of the genome. 3' ESTs have a uniform start site since they are anchored at the polyA tail by employing a polyT primer in first strand synthesis (Short, 1988 and see cDNA construction, below). The EST sequence then predictably will contain the 3' untranslated region (UTR). Even though this sequence may or may not be long enough to extend into the coding region, 3' ESTs represent gene specific sequences that have been used to provide genomic markers (Adams, 1991). The 5' ESTs are more variable in their start point because the length of first strand synthesis may not extend to the mRNA 5' end. Although the 5' EST does not necessarily contain the 5' UTR or the AUG start, it will contain coding sequence. The 5' ESTs therefore are considered gene family specific sequences rather than individual gene specific sequences (Hillier, 1996; Khan, 1992). The abundance of EST classes, which is proportional to their relative transcription levels can be determined from an EST database after assembly with multiple sequence alignment tools such as Phrap (Green, copyright 1994-1996). The EST database and the corresponding cDNA clones then serve as a source of expressed gene sequences for investigating specific genes. Therefore, the *A.nidulans* EST database created during the work described here will allow gene prediction, identification, annotation and suggest expression patterns as well as serve as a reservoir of vegetative

and asexually expressed gene sequences for immediate study.

1.4 cDNA Construction

In eucaryotic organisms the processes of transcription and translation are said to be uncoupled because the gene is transcribed in the nucleus and the mRNA passes through the nuclear pores into the cytoplasm before translation occurs. The coding regions in eucaryotic genes typically are not continuous but contain exons, regions of coding sequence interrupted by introns or noncoding sequences. The primary transcript, the heterogeneous nuclear RNA, found in the nucleus, contains both the intron and exon sequences (Abelson, 1979). Processing of the primary transcript to a messenger RNA requires the removal of the introns as well as the addition of a 5' cap (Furuichi, 1975) and a 3' poly A tail (Sachs, 1993) (Figure1).

Many methods of isolating RNA from sources as diverse as plant and mouse tissue have been developed but those including an isothiocyanate treatment step are commonly used (Chomczynski, 1987). Subsequent passage of the cellular RNA fraction over poly dT Sephadex enriches for the poly A containing mRNA (Chomczynski, 1987). The key discovery in cDNA synthesis was of an RNA-dependent DNA polymerase, reverse transcriptase (Baltimore, 1970) used for first-strand synthesis to produce a DNA copy of an mRNA (for example Houts, 1979) and RNase H (Krug, 1989), an RNA endonuclease associated with viral reverse transcriptase specific for DNA/RNA hybrids. The availability of these enzymes led to the development of protocols for synthesis of a double-stranded DNA copy (cDNA) and its subsequent insertion into a vector for maintenance and amplification. For example, the Lambda ZAP system was developed by Short et al. (Short, 1988), by adapting the methods of Gubler and Hoffman (Gubler, 1983), which uses poly dT primers containing a specific restriction enzyme sequence in first-strand synthesis to anchor the first strand synthesis start to the polyA tail. Second-

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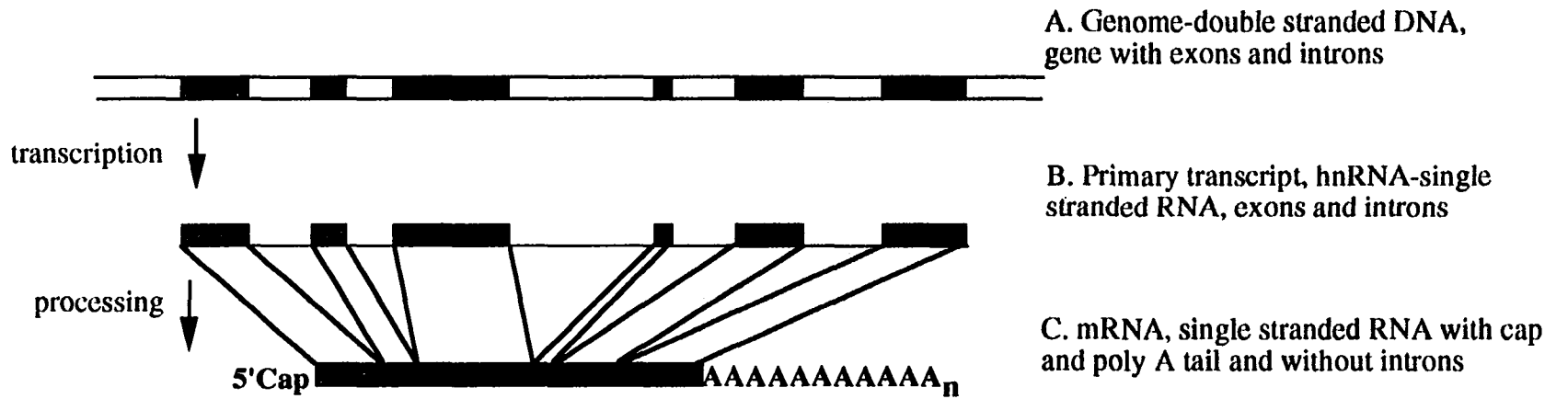


Figure 1. Steps required for generation of a processed mRNA ready for export to the cytoplasm and translation.

strand synthesis is performed by incubation with *E. coli* polymerase I in the presence of 5'-methyl dCTP. After the double-stranded cDNA is ligated to a second restriction enzyme cassette, it is digested with both enzymes. Since the methylated C residues prevent internal cleavage, only the sites flanking the cDNA are cleaved to create compatible ends on a double-stranded fragment which then can be directionally cloned into a suitable vector such as the *E. coli* bacteriophage lambda (Figure 2).

The lambda phage has been well characterized (Ptashne, 1992). A large region of this genome can be removed and replaced by ligation with foreign DNA inserts. The lambda Zap system, which is now a commercially available product, has been used for construction of a large number of cDNA libraries (Adams, 1995; Aramayo, 1990; Hillier, 1996; Nelson, 1997) and allows insertion of inserts up to 10 Kbp into the vector multicloning region (Short, 1988). The resulting molecule can be packaged in the presence of a "packaging mix" containing capsid proteins. This cell-free process prevents selection against any of the hybrid phage genomes. The lambda Zap vector also contains f1 and colE1 origins of replication and the beta lactamase gene for ampicillin resistance which allows conversion of a hybrid phage genome to a plasmid, and the lacZ gene containing the multicloning site allows selection for insert-containing plasmids.

Coinfection of the primary lambda library with an f1 helper phage, a filamentous single-stranded phage containing an amber mutation, allows excision of the cDNA from the lambda portion of the vector. A nonsuppressing host then is infected with the f1 lysate which prevents replication of the f1 phage genome (Dotto, 1984). Since f1 replication is not supported and the single-stranded form of the cDNA carries not only the f1 origin but also the colE1 origin of replication, a double-stranded form of the cDNA contained in a plasmid vector containing an ampR gene and lacZ interrupted by the cDNA insert results (Short, 1988). This plasmid form of the cDNA easily is maintained in the laboratory. It can be stably grown and double-stranded sequencing template can be prepared from it.

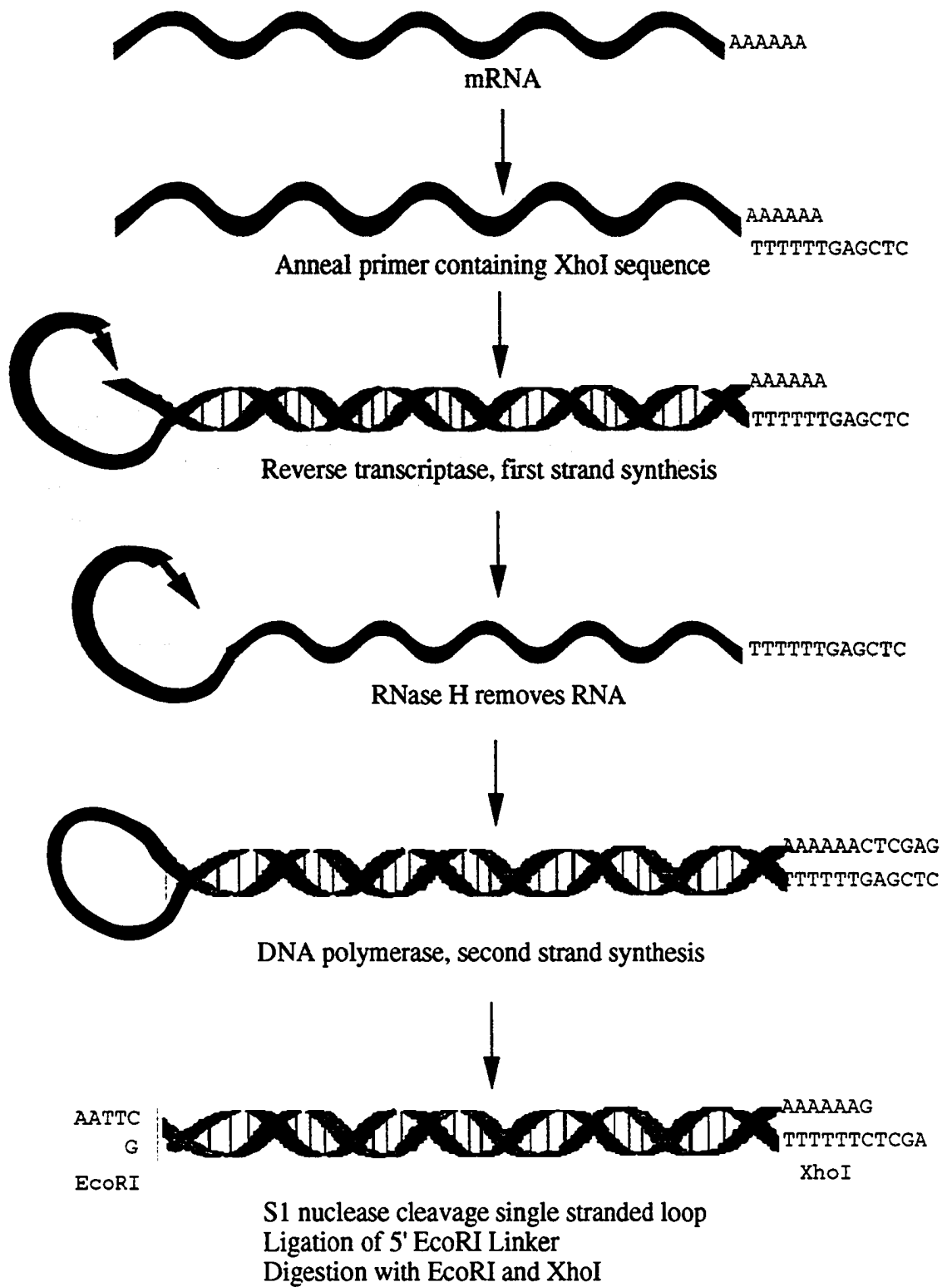


Figure 2. In vitro synthesis of double stranded cDNA from single stranded mRNA.

Figure 3 depicts a full length cloned cDNA with the position and direction of sequence for a pair of ESTs. Ideally, a full length cDNA will yield two ESTs where the 5' EST covers the 5' UTR, translation start, gene coding region and the 3' EST starts in the poly A tail, includes the 3' UTR region, translation stop and coding region which may or may not overlap with the 5' EST. If the two ESTs do not overlap then custom synthetic primers can be synthesized and used for another round of DNA synthesis to obtain the full length cDNA sequence.

1.5 DNA Sequencing

Since the original Sanger dideoxynucleotide DNA sequencing method was reported (Sanger, 1977), modifications have been introduced to improve both its efficiency and ease of use, making it the primary sequencing technique employed today. Two recent key modifications will be mentioned here.

The Sanger method is an enzyme based synthesis method and the original enzyme used was the Klenow fragment of DNA polymerase I which contains both 5'-3' polymerizing activity and 3'-5' exonuclease activity but has the 5'-3' exonuclease domain removed (Klenow, 1971). The discovery of thermostable DNA polymerases, including Taq from *Thermus aquaticus* (Innis, 1988b), allowed the polymerization reactions to be incubated at high temperatures (72⁰C for Taq) which inactivate Klenow enzyme but are required to reduce DNA secondary structure and GC compressions (Innis, 1988a). The original Taq polymerase has been modified to improve its efficiency by the removal of the 5' to 3' exonuclease domain and by a single amino acid change at position 667 from phenylalanine to tyrosine (Barnes, 1992; Tabor, 1995). This latter change reduced the discrimination against ddNTPs several thousand fold, a phenomenon first described by Tabor and Richardson (Tabor, 1995) who recognized that this discrimination could be altered by the presence of a tyrosine hydroxyl group at this position. The decreased discrimination against ddNTPs not only reduces the cost of DNA sequencing since much

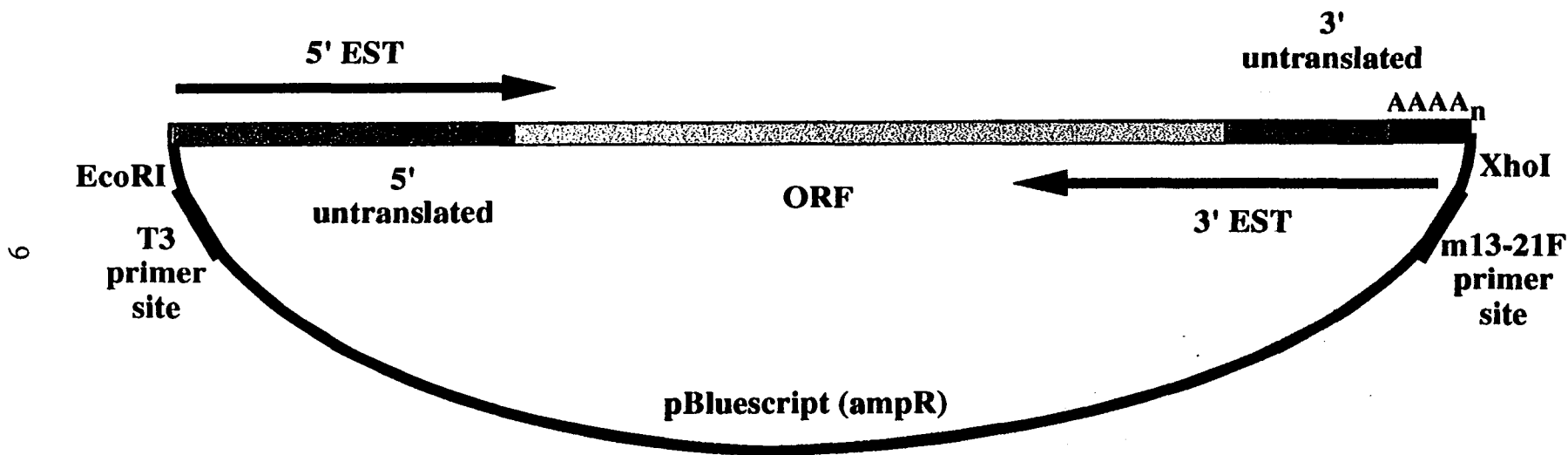


Figure 3. Position of an EST sequence pair on a cDNA clone. Blocks represent the cDNA insert. Red is the untranslated region, gray the coding region and green the polyA tail. The black line represents the cloning vector used in the *A. nidulans* library, pBluescript which contains the cloning sites, EcoRI and XhoI, and the sequencing primer sites (blue bars). The arrows represent the position and direction of the EST sequences.

less of the dideoxynucleotides need to be used in the cycle sequencing reactions but also improves the DNA sequence data quality and read length by producing more even signals and less background. Amplitaq-FS used in the work presented here is marketed by Applied Biosystems and contains both the Phe667Tyr mutation and an undisclosed N-terminal mutation to remove the 5' to 3' exonuclease activity (P. E. Applied Biosystems, 1998).

The introduction of fluorescent labeling and detection replaced the earlier radioisotope methods. Both fluorescent labeled primers and terminators now are available (Prober, 1987; Smith, 1986). For dye-labeled primers, the dyes are attached at the 5' most nucleotide base and for dye-terminator reactions, the fluorescent dyes are attached to the dideoxynucleotide base. Dye-labeled terminators have the advantage over labeled primers in that all four labeled dideoxynucleotides can be present in the same extension reaction, unlike labeled primer reactions where four separate reactions are required for each template. In addition, dye labeled dideoxynucleotides often eliminate any fold-back compressions because the large fluorescent group attached to the base of the ddNTP prevents base stacking on the 3' end of the newly synthesized chain. The background also is reduced because abortive stops which are not terminated with a labeled ddNTP are undetected (Lee, 1986). The fluorescent dyes typically used are fluorescein or rhodamine derivatives, each with a slightly different maximum emission wavelength. Applied Biosystems recently developed a dye set for terminator DNA sequencing which consists of energy-transfer dyes termed Big-Dye terminators, that use the 5-carboxy-d-rhodamine dyes as acceptor dyes coupled to 5- or 6-carboxy isomers of 4'-aminomethylfluorescein as the donor dye which results in a more even and intense signal (Rosenblum, 1997).

1.6 Instrumentation

Fluorescence-based DNA sequencers were first commercially developed by

Applied Biosystems (ABI 377 User manual, 1996) and all data reported in this dissertation was collected on the Applied Biosystems model ABI377. This instrument detects four different fluorescent dyes simultaneously and thus the four base-specific reactions can be pooled and electrophoresed in a single lane of the sequencing polyacrylamide gel. The ABI377 has a scanning argon laser and uses a CCD (charged coupled device) camera for detection. The argon laser excites the fluorophores and a series of lenses collect and focus the emitted light onto a spectrograph diffraction grating which separates the light based on wavelength into a predictably spaced pattern across the CCD camera. The collection software records the amplified emission signals and stores it in the Macintosh associated computer.

1.7 DNA sequence analysis

Several techniques were used to evaluate the sequence data in the EST and cosmid databases for quality and similarity with other sequences in the public databases.

Phred (Ewing, 1998; Ewing, 1998) and Phrap (Green, copyright 1994-1996) are companion programs developed in P. Green's group at the University of Washington. Phred examines the trace files obtained from the ABI377 by reading the raw DNA sequence trace data and then calls the bases. Phred also assigns sequence quality values to the bases by examining the four base traces in the region surrounding each point in the data set and predicting a series of evenly spaced locations. Phred then finds the center of the observed peaks in the trace files and the areas of these peaks relative to their neighbors. The observed peaks are compared to the predicted peaks to yield a calculated quality score (Green, copyright 1993-1996). Phrap, the assembly program, constructs contiguous sequences as a mosaic of the highest quality parts of reads using Phred quality values that are based on :

$q = -10 \log_{10} p$, where a quality value of 10, 20, 30 and 40 correspond to an error rate of 1/10, 1/100, 1/1000, and 1/10,000 respectively. Phrap currently is the program of choice

for assembly of the human genome project sequences because it is the only public domain program which calculates individual quality scores.

Consed, a program for viewing and editing Phrap sequence assemblies, uses a color system to indicate the quality of the bases in the reads that are assembled as well as those of the resulting contiguous sequence (Gordon, 1998). Consed also contains an algorithm for determining quality of the contiguous sequence expressed in number of errors/10,000 bases. An error rate of less than 1 error /10,000 bases which generally corresponds to a coverage of each base once in both directions (B. A. Roe, personal communication), is considered acceptable for the human genome project and is the level to which the cosmids discussed in this work were finished.

A comparison of any DNA query sequence to the National Institutes of Health (NIH) database of all known nucleotide and protein sequences, GenBank (Benson, 1996), reveals if any previously sequenced nucleic acids have homology to the query sequence. GenBank is based at the National Center for Biotechnology Information (NCBI), a division of the National Institutes of Health (NIH) and is accessible through the web at URL ncbi.nlm.nih.gov. NCBI builds GenBank both from direct submissions of sequence data from authors and from scanning the journal literature. The GenBank database also includes the data deposited in the EMBL Data Library in the United Kingdom and the DNA Databank of Japan. Data is exchanged daily between the three international databases thus maintaining a comprehensive set of public sequence information. ESTs are the most rapidly-expanding source of new genes at GenBank and the dbEST division of GenBank has been established specifically for these sequences.

For sequence similarity searches, Blast (Basic Local Alignment Search Tool, developed at the NCBI) is a heuristic search used by the programs BlastP, which compares an amino acid query sequence against a protein sequence database; BlastN, which compares a nucleotide query sequence against a nucleotide sequence database; BlastX which compares the six-frame translation products of a nucleotide query sequence

against a protein sequence database; tBlastN which compares a protein query sequence against a nucleotide sequence database translated in all six reading frames; and tBlastX which compares the six-frame translations of a nucleotide query sequence against the six-frame translations of a nucleotide sequence database. These programs use the statistical methods of Karlin and Altschul (Altschul, 1990) to present the significant regions of similarity to a query sequence.

1.8 *Aspergillus nidulans* as a model organism.

The filamentous fungus, *Aspergillus nidulans*, is one of four historically important ascomycete models which include the yeasts, *Saccharomyces cerevisiae*, *Schizosaccharomyces pombe* and a second filamentous species, *Neurospora crassa*. Seventy-five percent of the known fungi are filamentous. *A. nidulans* is unique among the four model organisms because of its extensive secondary metabolism. *A. nidulans* also is considered a model organism for investigation of molecular and genetic questions of closely related plant and human pathogens and industrially important members of the filamentous ascomycetes (Timberlake, 1990). Examples include *A. parasticus* and *A. flavus*, two of the producers of aflatoxin, a highly toxic DNA intercalating agent which contaminates harvested grain and nuts (Jimenez, 1991; Magnoli, 1998) and the industrially important *Penicillium chrysogenum*, the major producer of the antibiotics, penicillin (Josten, 1998) and griseofulvin (De Carli, 1998). Three *Aspergillus* species, *A. niger*, *A. flavus* and *A. fumigatus*, are common human opportunistic pathogens because they can infect at the site of a trauma such as a burn and can cause aspergillosis, a lung or sinus infection which can become systemic especially in immunocompromised individuals (Fenelong, 1999). *A. niger* and *A. oryzae* are two species used industrially to produce products such as ribonucleases, citric acid, alpha-amylase, invertase, pectinase, detergent enzymes, soy sauce and miso (a soy based food) (Carlile, 1994). Other filamentous ascomycetes such as *Magnaporthe griseae*, the causative agent in barley blight, are

responsible for significant crop losses worldwide (Bennett, 1985).

A. nidulans also is a eucaryotic model system for studying developmental regulation and cell differentiation (Morris, 1992). In the early 1950's, G. Pontecarvo selected *A. nidulans*, a nonpathogenic, easily grown member of the ascomycetes for intensive study, specifically to develop a well-characterized model system (Pontecorvo, 1953). His selection rationale was based on the criteria that *A. nidulans* has a well characterized sexual cycle, has a haploid nuclei for most of its life cycle, has unicellular ascospores, forms heterokaryon, grows on defined medium in liquid or on a solid surface, forms compact colonies on agar, has mature fruiting bodies which do not eject ascospores, and has easily micromanipulated asci from which a large number of ascospores can be obtained. Subsequent work has added two additional properties to this list, the ability of *A. nidulans* to grow as a synchronous culture for asexual development (Timberlake, 1980) and to be transformed after treatment with calcium chloride (Ballance, 1983) or by electroporation (Fromm, 1986)

Research findings have pointed to the success in using *A. nidulans* in a variety of studies. Dean and Timberlake (Dean, 1989) have demonstrated the plant pathogenic potential of *A. nidulans* by successfully establishing infections in cotyledons and fruit. This phytopathogenic character is reinforced by the findings that *A. nidulans* contains the inducible ability to catabolize quinate, a breakdown product of lignin (Hawkins, 1982) and to express two enzymes that are required in catabolism of pectin (Dean, 1989). The presence of the aflatoxin gene cluster lacking the last two enzymes in the pathway recently has been established (Adams, 1996). The entire *A. nidulans* penicillin biosynthetic cluster that originally was exploited in *Penicillium* species has been isolated and characterized (MacCabe, 1990). *A. nidulans* now is considered the model organism for regulatory studies on both of these pathways. Many of the genes involved in *A. nidulans* mitosis also have been cloned and characterized. Several have homologs in other eucaryotes while others are unique to *A. nidulans* (Morris, 1992). *A. nidulans* also is a model system for

septation studies. Momany and Hamer recently found that as in *Drosophila*, the septin encoding gene is essential for *A. nidulans* growth. However, since neither *S. cerevisiae* nor *S. pombe* require septin (Brown, 1996) *A. nidulans* is a better model for studying this system.

Early sequencing results suggest that *A. nidulans* gene sequences will have application to other fungi and higher eucaryotes. For example, the results obtained from the analysis of *A. nidulans* cosmid W06E08 (Kupfer, 1997) showed only two ORFs with high homology to the completely sequenced *S. cerevisiae* ORFs, four with homology to other filamentous ascomycete genes, including the first transposase found in *Aspergillus* which is similar to a class found in *Drosophila*, two with homology to higher eucaryote genes (squid, and mouse). In addition, there are four predicted genes which have no significant similarity to any GenBank entries. An additional genome region of *A. nidulans* sequenced and analyzed as part of this dissertation work shows similar homology results as W06E08 and as well has homologous *A. nidulans* ESTs for ten of the 17 predicted genes. Thus, an EST sequencing project from an *A. nidulans* cDNA library most likely would have a broad application to other eucaryotes. In addition, there could be a number of new genes discovered during such an EST project that would complement the genome information from *S. cerevisiae* and provide a basis for further genomic structural and functional studies of this filamentous fungi.

Chapter II

Materials and Methods

Section 1. cDNA Library

The *Aspergillus nidulans* cDNA library that served as the source of template in the EST project was constructed by Rodolfo Aramayo, Texas A&M University. This library was constructed in 1990 using poly A RNA harvested from *A. nidulans*, strain FGSC A26 (veA1, bio), which had undergone development for 24 hours on a solid surface with an air interface and therefore contained cDNAs from both vegetative mycelial cells and cells involved in asexual reproduction. The cDNAs were cloned directionally such that the 5' end was adjacent to the Eco RI site and the 3' end the XhoI restriction site of the LambdaZap II vector arms (Stratagene Cloning Systems) (Aramayo, 1990).

The *A. nidulans* was grown in minimal medium (Kafer, 1977) plus biotin prepared as follows: 50 ml/l 20x salt solution (120 g/l NaNO₃, 10.4 g/l KCl, 10.4 g/l MgSO₄·7H₂O, 30.4 g/l KH₂PO₄), 1 ml/l trace elements (22 g/l ZnSO₄·7H₂O, 11 g/l H₃BO₃, 5 g/l MnCl₂·4H₂O, 5 g/l FeSO₄·7H₂O, 1.6 g/l CoCl₂·5H₂O, 1.6 g/l CuSO₄·5H₂O, 1.1 g/l (NH₄)₆ Mo₇O₂₄·4H₂O, 50 g/l Na₄ EDTA), 10 g/l D-glucose, 0.5 ml/l 10 mg/ml biotin in 95% ethanol. One liter of minimal medium was inoculated with 4x10⁸ conidia and shaken at 37°C for 24 hours. Then 100 ml aliquots were harvested and transferred to Whatman #1 filter paper (9 cm diameter). The filters were laid down onto glass beads in a Petri dish containing 25 ml of minimal medium with biotin. Petri dishes were placed in a metal tray containing water to provide a moist atmosphere and incubated at 37°C for 24 hours. The synchronously developing cells then were washed from the filter. The cells were lysed by grinding in liquid nitrogen. Total RNA was

isolated by the guanidine isothiocyanate method and the polyA RNA was purified twice by passage over oligo (dT) cellulose (Chomczynski, 1987). First and second strand synthesis, ligation of the cDNA into LambdaZap vectors and packaging was performed using the reagents and protocol provided in the ZAP-cDNA synthesis kit (Stratagene #200402, R. Aramayo, personal communication)

2.1 Characterization and Preparation of cDNAs

The cDNA library was converted from the LambdaZap form, provided by R. Aramayo and R. Prade, to a plasmid form in a mass excision by the following procedure:

1) f1 helper filamentous phage was added at a 1:1 phage:cell ratio and the lambda library at a 10:1 phage:cell ratio to strain XL1-Blue MRF^r (an f1 filamentous phage host) using sufficient lambda library to equal ten times the primary library size of approximately 5×10^5 plaques.

An overnight culture of XL1-Blue-MRF^r cells grown in LB, 0.2% maltose and 10mM MgSO₄ was collected by centrifugation and resuspended at an A₆₀₀ of 1.0 (approximately 8×10^8 cells/ml). Cells, lambda library and helper phage were mixed and incubated in a 50 ml conical tube at 37⁰C for 15 minutes. Twenty ml of LB broth was added and the cells were incubated for 3 hours at 37⁰C with shaking. The tube was heated at 65⁰C for 20 minutes to increase cell lysis.

2) The supernatant containing the released single-stranded filamentous phage containing the minus strand of the excised phagemid, pBlueScript SK- with the cDNA insert was centrifuged at 1000xg for 10 minutes to pellet cell debris and the supernatant was transferred to a fresh conical tube. The f1 supernatant could be stored for up to six months at 4⁰C before any significant loss of titer was seen and an additional excision was necessary.

3) *E. coli* strain SOLR (not an f1 host) was infected with the filamentous phagemid in the absence of helper phage and plated on L broth solid medium containing 100 ug/ml ampicillin, 4 mg/ml Xgal, 5 mg/ml IPTG. To titer the excised phagemids, 5 ul

of the supernatant was mixed with 200 μ l of a overnight culture of SOLR, collected and resuspended at one-half volume in 10 mM MgSO₄, and incubated at 37⁰C for 15 minutes. 100 μ l of the mixture was plated onto a LB amp plate and incubated overnight at 37⁰C. Generally, 2 μ l to 5 μ l of lysate per 200 μ l of freshly prepared SOLR cells was used for single-colony isolation.

2.2 Growth of cDNA Clones and Preparation of Glycerol Stocks

The SOLR colonies containing cDNA phagemids were examined for insert based on the blue /white β -galactosidase selection where white indicates an insert in the multicloning site of the vector. The ratio of white to blue was noted (Table 1). White colonies were picked from the LB plates and inoculated into 1.5 ml Terrific Broth (12 g Bacto-tryptone, 24 g Bacto-yeast extract, 4 ml glycerol per 900 ml) with salts (2.31 g KH₂PO₄, 12.54 g K₂HPO₄ per 100 ml) containing 100 μ g/ml ampicillin in sterile 96 well blocks. After incubation for 18 hours at 37⁰C with aeration, the cells were placed on ice. Two microtiter plates per block were prepared as freezer stocks by adding 40 μ l 50% sterile glycerol per well and 100 μ l of cell culture transferred from each of the 96 wells of the blocks to the microtiter plate with a 12 channel pipettor for both transferring and mixing the cells with the glycerol. The microtiter plates were labeled and stored at -80⁰C. One set of glycerol stocks was sent to the Fungal Genetics Stock Center, Kansas Medical School, Kansas City, KS for archiving and distribution. The remaining 1.3 ml of cell culture was collected via centrifugation at 3000 rpm for 5 minutes in a Beckman GS-6R centrifuge. The cell pellets were frozen at -20⁰C.

Table 1. Summary of *A. nidulans* cDNA Library Characteristics

λ Library Titer	Phagemid Yield from F1 Supernatant	Percent Clones with Insert	Average Insert Size (36 Clones)
7x10 ⁹ pfu/ml	8x10 ⁵ cfu/ml	62%	1.0Kbp (0.5-3.0Kbp)

2.3 Semi-automated alkaline lysis isolation of small-insert DNA

The phagemids were isolated via a modified alkaline lysis protocol (Birnboim, 1979) using a Beckman Biomek 2000 automated workstation and a Hydra96 (Robbins Scientific) as described below.

The cell pellets were thawed for 15 minutes at room temperature and then 200 μ l TE RNase A (50 mM Tris pH8, 10 mM EDTA, 100 μ g/ml RNase A) was added to each well in the 96 well block by a Hydra96. The blocks were placed on a titer plate shaker for 10 minutes at setting 7.5 to resuspend the cell pellets.

A program originally written for the Biomek by Judy Crabtree (Crabtree, 1997) was modified to begin with the addition of 200 μ l of alkaline lysis solution. The modified program "td sds-to-end" was selected and the blocks and reagents were placed on the worksurface as directed in the program layout. Following the addition of 200 μ l of alkaline lysis solution (0.2 N sodium hydroxide and 1% dodecyl sulfate, SDS) and 100 μ l of 3 M potassium acetate, pH 4.8, the blocks were removed from the Biomek and covered with an acetate sealer. Each block was vortexed for 60 seconds, a critical step for formation of a solid debris pellet and cleared lysate.

The blocks were placed on a titer plate shaker at a setting of 6 for 5', then stored at -20° C for 1 hour. This was followed by centrifugation at 3000 rpm for 45 minutes in the Beckman GS-6R centrifuge. The blocks were returned to the Biomek worksurface, and 400 μ l of cleared lysate were transferred to four fresh blocks. One ml of 100% ethanol was added to each sample manually and the blocks were placed at -20° C overnight. The precipitated DNA was collected by centrifugation for 30 minutes at 3000 rpm in the Beckman GS-6R. The pellets were washed twice with 70% ethanol and the blocks drained on paper towels. The Hydra was used to add 100 μ l of sterile deionized water to the pellets which were stored overnight at 4° C following 15 minutes of shaking on a titer plate shaker at a setting of 7.5. Two μ l aliquots were examined on a 1% agarose gel using 0.2 μ g pGEM as a vector standard. The presence of low molecular

weight RNA, insoluble material remaining in the wells and the relative concentration of the isolated cDNA template was examined. If insoluble material was present, the templates were treated by the addition of equal volumes of 7.5 M neutral potassium acetate. The mixture was vortexed for 30 sec. and placed at -20°C for 30 min. The debris pellet was collected by centrifugation for 30 min. at 3000 rpm. The supernatant containing the cDNA was removed to a fresh block and two volumes of 100 % ethanol were added followed by incubation at -20°C for 30 min. The DNA pellets were collected by centrifugation at 3000 rpm for 30 min and washed twice with 70 % ethanol. After drying in vacuo, the DNA was resuspended in 50 ul sterile deionized water and rechecked on an agarose gel before use in a sequencing reaction.

Thirty-six of the first block of clones isolated were examined for insert size by restriction analysis with XhoI and Eco RI. The approximate size of the insert was determined by comparison with the size markers 100 base-pair DNA ladder (Gibco BRL) and Hind III fragments of lambda DNA (Gibco BRL) (Table 1). When multiple bands appeared other than the vector, their sizes were added.

2.4 Optimum Sequencing Conditions

In trial sequencing reactions, a variety of parameters were examined to determine the optimum conditions for DNA sequencing. Both the quality of the sequence data and the number of bases obtained were examined because, for single-pass sequencing in the forward and reverse direction, the extent of data acquired should be maximized to improve the chance that the combined data for the forward and reverse reactions will yield a contiguous segment containing the entire coding region.

The thermocycling reaction employed Taq DNA polymerase catalyzed chain extension using fluorescent dye-terminators containing only one unlabeled primer. The polymerization product was a single-stranded DNA nested-fragment set ending in a dye-labeled dideoxynucleotide. Since dye terminators were used, all four bases can be

included in each reaction so that only a single sequencing reaction is required instead of four as is necessary in dye-labeled primers. In addition, false stops caused by termination when the enzyme prematurely dissociates from the template were not detected because of the lack of incorporated dye terminator and foldback compression was prevented when the extended DNA chain correctly terminated with the bulky fluorescent dideoxynucleotide.

In the trial reactions, two forms of Taq DNA polymerase were examined. The first, Amplitaq FS (Applied Biosystems) contains two modifications: (a) an N-terminal deletion which eliminates the 5'-3' exonucleolytic activity and results in greater processivity of the enzyme, and (b), a residue change from phenylalanine to tyrosine in the ribose selectivity site which increases the affinity for dideoxynucleotides (Tabor, 1995). The thermocycling reactions using Amplitaq FS were performed in the presence of a heat-stable pyrophosphatase which hydrolyzes pyrophosphate maintaining favorable conditions for DNA polymerization. The second, KlenTaqTR enzyme, has both modifications as described for Amplitaq FS, however the N-terminal deletion is not identical (Barnes, 1995; Korolov, 1995). Reactions with this enzyme were performed in the absence of a pyrophosphatase.

Four sets of reactions were tested using the same 24 DNA samples; each enzyme in the presence and absence of 5% dimethylsulfoxide (DMSO) a denaturing agent which can reduce secondary structure in the DNA. Primer, 0.65 μ M and 200-500ng DNA template were used in all cases. The primers used were the M13 -21 universal forward (5'-TGT AAA ACG ACG GCC AGT3') and SK- (5'-CGC TCT AGA ACT AGT GGA TC-3'). The universal forward primer sequence is located in the pBluescript vector 48 bp upstream of the 3' end of the cDNA insertion in the XhoI site and the SK- primer site is located 12 bp upstream of the 5' end of the cDNA insertion in the Eco RI site. Amplitaq FS was a commercially available mixture (PerkinElmer) containing: Amplitaq DNA polymerase FS, Tris-HCl pH 9.2, dCTP, dATP, dTTP, dITP, ddATP-dye terminator

(R6G), ddGTP-dye terminator (R110), ddTTP-dye terminator (6-TAMRA), ddCTP-dye terminator (6-ROX) all rhodamine fluorescent dyes, magnesium chloride, thermal-stable pyrophosphatase. The concentrations for each reaction component are not given by the manufacturer (PerkinElmer, 1995). All Amplitaq reactions were in a final volume of 5 μ l. The KlenTaq TR reaction mix contained: 50.0 mM Tris-HCl pH9.2, 16.0 mM ammonium sulfate, 3.5 mM MgCl₂, 1.0 mM MnSO₄, 150 μ M dATP, dTTP, dCTP, 450 μ M dITP, 1/2000 dilution of Perkin Elmer #401384 "dye deoxy terminators", 1/200 dilution of KlenTaq TR as supplied (Wayne M. Barnes). Twenty μ l reactions were used. FS-containing samples were thermocycled for 25 cycles of 96⁰C for 10 sec, 50⁰C for 5 sec and 60⁰C for 4 minutes while the KlenTaq-containing samples were incubated for 25 cycles of 97⁰C for 50 sec and 65⁰C for 4 minutes.

Unincorporated dye-labeled dideoxynucleotides were removed by chromatography through Sephadex G-50 into a 96 well microtiter plate. The filtered samples were dried in a vacuum oven at room temperature before addition of 1 μ l of a 5:1 formamide:0.1% blue dextran, 1 mM EDTA loading buffer. Slab gel electrophoresis was on 48 cm, low fluorescent glass plates separated by 0.2 mm spacers and containing 5.3% Long-Ranger acrylamide gel mix (FMC) with 8 M urea. Electrophoresis was performed at 2.3 kV, 52⁰C for 10 hr in 1x TBE buffer (10.8 g Tris base, 5.5 g boric acid, 8.4 g EDTA in 1 L) on an Applied Biosystems ABI377 automated sequencer. The results summarized below show the number of high quality bases read. A high quality base is defined as the sequence length up to the position where two bases in five were called as N using the ABI basecalling software.

TABLE 2-Preliminary Sequencing Results-24 Samples

	Taq FS		KlenTaq TR	
Average # of bases	+DMSO	-DMSO	+DMSO	-DMSO
	715	650	586	540

The optimum runs were achieved with the Taq FS chemistry. Since 5% DMSO

has positive effects on sequences which exhibit secondary structure and does not appear to have any negative effect on Taq FS reactions, 5% DMSO was used routinely for sequencing the cDNAs.

Two reverse primers were examined, SK- , 12 bases upstream of the Eco RI site and T3 (5'-CGA AAT TAA CCC TCA CTA AAG3') which is 64 bp upstream. Both were used successfully, however the position of the SK- primer was too close to the insert to enable the program used later in analysis to routinely find the position of the Eco RI site which flagged the beginning of the insert sequence. This was because the signal generated for the first 20-25 bases often was too high to be accurately basecalled. Therefore, the T3 primer was used routinely for the 5' EST sequences.

Roughly half of the ESTs were sequenced with the rhodamine fluorescent dyes described above. Following the introduction by Perkin-Elmer Biosystems of the energy transfer dyes, termed BigDye energy transfer-based terminators (Rosenblum, 1997), the EST sequencing reactions were thermocycled using a 1:16 dilution of the Perkin-Elmer recommended reaction mix of BigDyes containing Amplitaq FS enzyme. The energy transfer dyes are constructed by linking one of two donor dyes, the 5- or 6-carboxy isomer of 4' aminomethylfluorescein (5 Fam or 6 Fam) with a 5-carboxy-dichloro-rhodamine dye. The ddATP contains the donor dye 5-FAM linked to the ddATP acceptor dye dR110-2. 6-FAM is linked to ddCTP acceptor dye dROX-2, ddGTP acceptor dye dR110-2 or ddTTP acceptor dye dTAMRA-2. A typical reaction contained 2-3 μ l template DNA, 1 μ l 25% DMSO, 1 μ l 13 μ M universal forward or 26 μ M reverse primer, 2 μ l diluted terminator:enzyme mix (1 μ l terminator:enzyme mix :3 μ l 5x buffer: 50 mM Tris, pH9.0, 10 mM MgCl₂). The average number of bases, 556 (average of 24 samples) cannot be compared directly to that seen with the dRhodamine dyes since the run time was changed from 10 hours to 6 and the average number of bases obtained with a six hour run was reduced to 550-600.

2.5 Cycle Sequencing Conditions

The dilution of the BigDye components in the thermocycling reactions, was extremely cost effective but required an additional number of cycles to achieve the same intensity of signal seen with the undiluted D-rhodamine dye mix. Therefore, 60 cycles instead of the ABI recommended 30 cycles were used with the same temperature and time as used previously for TaqFS: 95⁰C for 10 seconds, 50⁰C for 5 seconds, 60⁰C for 4 minutes.

All thermocycling reactions were performed in 384 well plates (Cycleplate 384, Robbins Scientific) on the Perkin-Elmer 9600 or 9700 thermocycler. Since the 384 well plates have 96 wells each divided into quadrants, samples from two blocks of 96 cDNA templates could be thermocycled with both the forward and reverse universal sequencing primers simultaneously. The reactions were prepared by a semi-automated procedure using the Hydra96.

The pBluescript-based DNA sequencing template required for the reaction was transferred from the first 96 well DNA template block and dispensed to two of the four quadrants of the 384 plate using the Hydra 96. This process was repeated to transfer a second block of templates into the last two quadrants of the 384 plate. A sufficient volume of the reaction mix for two forward reactions was aliquoted to 96 wells of a 96-well thermocycle plate (Robbins Scientific). This was transferred into two quadrants of the 384 plate. The reverse reaction mix then was dispensed into the remaining two quadrants. Thus, a single 384 well plate contained both the forward and reverse sequencing reactions for each sample in two 96 well sets of subclones or cDNA.

2.6 Unincorporated Dye Terminator Removal.

Following thermocycling, unincorporated terminators and buffer salts were removed from the reaction by gel filtration through Sephadex G-50 mini-columns prepared in 96-well filter plates (Millipore, MASVN6550). To prepare the mini-filtration

columns, 200 μ l dry Sephadex G-50 was added to each well. Then 400 μ l of distilled-deionized water (dH₂O) was dispensed into each well using the Hydra96. The plates were allowed to hydrate at 4⁰C for two hours to overnight. Each 96-well plate was centrifuged for 2.5 minutes at 1500 rpm. An additional 100 μ l dH₂O then was added followed by centrifugation for 2.5 minutes at 1500 rpm. Ten microliters of dH₂O was added to each sample in the 384 well cycle plate once cycling was completed to dilute the reaction. The 384 well plate was centrifuged for 2 minutes at 1500 rpm to collect the liquid to the bottom of the well. The entire volume for each reaction was transferred to the top of one of four 96-well Sephadex G-50 column filter plates using the Hydra96. A 96 well microtiter plate was secured under each filter plate and the filter plate and attached microtiter plate were centrifuged for 3 minutes at 1500 rpm to collect the purified reaction products in the microtiter plate. The reactions then were dried under vacuum without heat and stored at -20⁰C until loading dye was added prior to loading onto the sequencing gels.

2.7 Automated Data Collection

Electrophoresis was performed as described above for six hours on an ABI 377 automated sequencer. Data was automatically collected and analyzed by the Data Collection and Data Analysis software on a Macintosh computer associated with the ABI377 sequencing equipment. Analyzed data then was transferred to the networked Unix-based Sun workstations for assembly and analysis.

Section 2. *Aspergillus nidulans* Cosmids

2.8 Background-Method of Construction of Cosmids

Pulsed-field gel electrophoresis (PFGE) technology has allowed the separation of linear double-stranded molecules up to 10 megabase pairs (Mbp) (Vollrath, 1987). A version of PFGE, contour-clamped homogeneous field electrophoresis (CHEF) was used

successfully by Brody and Carbon (Brody, 1989) to separate the eight chromosomes of *A. nidulans*. The CHEF method employs a hexagonal array of 24 electrodes which produce a homogeneous electric field at an angle of 120° (Vollrath, 1987). The 2.9 Mbp chromosome IV was well resolved from the other chromosomes with this procedure.

Using the CHEF method developed by Brody and Carbon, Prade, et al. (Prade, 1997) isolated chromosomal material for electrophoresis from *A. nidulans* mycelial cells which were treated to form protoplasts. The protoplasts were mixed with an equal volume of 1.4% agarose in a plug mold that then was mixed with an SDS lysis solution, incubated and washed with EDTA. Plugs were inserted into a 0.8% agarose gel and electrophoresed with three pulse intervals, each with increasing duration of the forward pulse, for a period of 156 hours (Brody, 1989). Using this approach, Prade, et al. (Prade, 1997) isolated chromosomal DNA for creating a tiled set of cosmids for each *A. nidulans* chromosome, sub-cloning into the cosmid vector, pWE15, which encodes both kanomycin and ampicillin resistance and contains the colE1 origin of replication.

The three cosmids sequenced in this study were from the chromosome IV tiled set.

Cosmid W06E08 was obtained from Rolf Prade, Oklahoma State University, as dried DNA. The pellets were resuspended in 10:0.1 TE and transformed into CaCl₂ competent ED8767 cells. Cosmids W02H02 and W30B01 were received from Nancy Keller, Texas A&M University, as *E. coli* transformed cells.

2.9 Large scale DNA Template preparation

During the course of this research, two large scale isolation methods were used. Cosmid W06E08 was isolated using the diatomaceous earth-based method (Carter, 1993). This method which is based on the high salt binding and low salt release of DNA is less costly and hazardous than isolation using equilibrium centrifugation in cesium chloride-ethidium bromide gradients (Tanaka, 1977). However, the diatomaceous earth based method had two disadvantages. First, the yield of DNA often was low (Chen,

1997) and second, the method was not amenable to automation. Therefore a modification of the alkaline lysis procedure (Birnboim, 1979), termed a double acetate procedure, also was employed to successfully isolate cosmid DNA since this protocol gave more consistent results than the diatomaceous earth procedure described above and could be successfully automated. Chen and Kupfer (unpublished results) subsequently developed a protocol using the Beckman Biomek 2000 to automate the large scale cosmid, BAC and PAC DNA isolation. Cosmids W02H02 and W30B01 were isolated using this semi-automated double acetate method.

2.10 Large-insert DNA Isolation Using Diatomaceous Earth

A smear of colonies of cosmid W06E08 were picked and used to inoculate a 12x75 mm Falcon tube containing 3 ml LB medium (10g Bacto-Tryptone, 5 g Bacto-yeast extract and 10 g NaCl in 1 liter deionized water, autoclaved) with 100 ug/ml ampicillin. After incubation at 37⁰C for 8 hours with shaking at 250 rpm, the culture was transferred to a 250 ml flask containing 50 ml of the same medium and incubated for an additional 8 hours. All 53 ml of the culture was transferred into a 2 liter flask containing 1 liter of the same medium and incubated for an additional 8 hours.

The cells were harvested by centrifugation at 6000 rpm for 15 minutes in 500 ml bottles in the Sorvall RC5-B using the GS-3 rotor. Cell pellets were frozen at -70⁰C until processed. The cells were thawed and resuspended completely in 35 ml of GET (50 mM glucose, 25 mM Tris-HCl, pH 8.0 and 10 mM EDTA). To the resuspended cells was added 70 mg of lysozyme for a final concentration of 2 mg/ml. After gentle mixing to dissolve the lysozyme, the solution was incubated at room temperature for ten minutes.

Cells were lysed by addition of 70 ml of alkaline lysis solution (0.2 N sodium hydroxide, NaOH, and 1% sodium doedecyl sulfate, SDS) followed by gently swirling until the solution was homogenous. After the mixture was incubated for five minutes on an ice-water bath, 70 ml of 3 M sodium acetate, pH 4.8 was added with gentle inversion

of the bottle several times. The bottle then was placed on an ice-water bath for 60 minutes. The lysate was cleared from precipitated SDS, proteins, membranes and chromosomal DNA by passing through a double layer of cheesecloth into a sterile 250 ml centrifuge bottle, followed by centrifugation at 10,000 rpm for 30 minutes at 4⁰C in the Sorvall RC5-B centrifuge using the GSA rotor. The supernatant was transferred into a 250 ml sterile centrifuge bottle and 20 mg/ml DNase-free RNase A (20mg/ml RNase A in 1 mM, pH 4.5, boiled for ten minutes and cooled slowly to room temperature) was added to a final concentration of 100 ug/ml. Following incubation at 37⁰C for 30 minutes, the supernatant was divided equally between two 250 centrifuge bottles, an equal volume of isopropanol was added to each and mixed by inversion. The solution was held at room temperature for five minutes. The DNA pellet was collected by centrifugation for 30 minutes at 9,000 rpm for 30 minutes. The supernatant was decanted and the pellets drained. Each DNA pellet was dissolved in 10 ml of 10:1 TE (10mM Tris-HCl pH 8.0 and 1 mM EDTA) and transferred to a 50 ml Corning glass centrifuge tube. To each was added 20 ml of defined diatomaceous earth-guanidine-HCl (defined diatomaceous earth suspended as 100 mg/ml in 6 M guanidine-HCl, 50 mM Tris-HCl, 20 mM EDTA, final pH 6.4). This mixture was inverted several times during a five minute incubation at room temperature followed by centrifugation in the Beckman GS-6R tabletop centrifuge at 3000 rpm for ten minutes. The supernatant was decanted and 40 ml of wash buffer (equal volume of 10:1 TE and ethanol) was added to each tube to resuspend the diatomaceous earth-DNA complex. After an additional centrifugation and decanting of the supernatant, the diatomaceous earth-DNA complex was suspended in 40 ml of acetone and centrifuged for 10 minutes. The supernatant was decanted and the pellets were dried in a vacuum oven.

The DNA was eluted from the dry diatomaceous earth-DNA after resuspension in 20 ml of 10:1 TE buffer and incubation at 65⁰C for ten minutes. The diatomaceous earth was collected by centrifugation for 15 minutes at 3000 rpm in the GS-6R. The

supernatant containing the DNA was decanted and the elution step was repeated. The two supernatants were pooled. The DNA was precipitated by adding of two volumes of 95% ethanol containing 0.12 M sodium acetate (NaOAc) and incubating in an ice-water bath for 45 minutes. The DNA pellet was collected by centrifugation for 15 minutes at 3000 rpm. The final pellet was washed with one volume of 70% ethanol and dried. The dried pellet was dissolved in 2 ml of dH₂O and the DNA concentration was determined by measuring the absorbance at 260 nm.

2.11 A Protocol for the Semi-automated Double Acetate Isolation of Large-insert Template DNA

The alternative protocol, a semiautomated version of the alkaline lysis protocol was used to isolate cosmid DNA from W02H02 and W30B01. This protocol, described below, includes an acidic sodium acetate precipitation and a second neutral potassium acetate precipitation. Both appear to be necessary and sufficient for a DNA preparation which can be used to generate shotgun subclones and which, with an additional phenol/chloroform treatment, can be used as a template for direct DNA sequencing.

For each cosmid, a smear of colonies was used to inoculate 3 ml of LB medium containing the 100 µg/ml ampicillin in a 12 x75 mm Falcon tube. The culture was incubated at 37°C for 8-10 hr with 250 rpm shaking. The entire culture was transferred to a 250 ml flask containing 50 ml of the same medium and incubated for 8-10 hr under the identical conditions. All 53 ml of the culture was added to a 2 liter flask containing 1 liter LB medium and antibiotic and incubated for additional 8-10 hr.

The cells were harvested by centrifugation at 7000 rpm for 15 min. in a 500 ml bottle in the RC5-B using the GS3 rotor. After the cell pellets were frozen and stored at -70°C, they were thawed on ice and resuspended in 80 ml of 50:10 TE (50 mM Tris-HCl, pH 8.0, and 10 mM of EDTA, pH 8.0) by gently pipetting up and down. The resuspended cells were dispensed in 200 µl aliquots into four 96-deep well blocks with

repeat pipet.

A series of programs baciso1-baciso5 were written to perform portions of the isolation protocol using the Beckman Biomek 2000. The Biomek was turned on and calibrated. The program baciso1 was opened, the MP200 pipet head, one block of 250 ul tips, and four blocks of resuspended cells were placed on the platform of the Biomek as shown on the program layout. baciso1 adds 200 ul of alkaline lysis solution and 200 ul 3M NaOAc pH4.8 to each well. These steps take 45 min. Reagents for this step are 100 ml of alkaline lysis solution (0.2 N NaOH, 1% SDS) and 100 ml of 3 M NaOAc, pH 4.8 which were added to the two designated reservoirs. The four blocks were removed following the addition of the lysis solution and placed on a titer plate shaker for 5 min. (setting of 5) to mix the lysis solution and to shear the genomic DNA to improve the efficiency of clearing the lysate. The blocks then were returned to the Biomek and the program continued by adding 200 µl sodium acetate to each well. The blocks were removed from the Biomek platform and again placed on a titer plate shaker for 5 min. (setting of 5) and frozen at -20°C for 40 min. Precipitated SDS, cell membrane and chromosomal DNA were collected by centrifuging the blocks in the Beckman GS-6R centrifuge at 3000 rpm for 30 min.

The program baciso2 was selected, and the blocks were returned to the Biomek platform along with four new blocks as shown on the program layout on the computer screen. The Biomek transferred 400 µl of cleared lysate from each well to an empty well in a new block over a 30 minute period. If flocculent material (typically inadequately sheared genomic DNA and protein) inadvertently was transferred to the fresh blocks, the transfer blocks were placed on titer plate shaker for 5 min. and returned to -20°C for 20 min. The 30 min. centrifugation was repeated, the blocks were placed on the Biomek and the baciso2 transfer to fresh blocks was repeated. The blocks containing the cleared lysate were removed from the Biomek and 250 µl of 100% isopropanol was added to each well manually using a repeat pipet. The blocks were covered with acrylic sealers and inverted

three times to mix. The sealers were removed immediately and the blocks incubated at room temperature for 5 min. To collect the DNA pellets, the blocks were centrifuged in a Beckman GS-6R centrifuge at 3000 rpm for 30 min. The isopropanol was decanted and the blocks drained by inversion on paper towels for 10 min.

Program baciso3 resuspended the DNA pellets and added 7.5 M potassium acetate (KOAc) for a second acetate precipitation. The blocks were returned to the Biomek platform as shown on the program layout. Fifty ml of 10:1 TE-RNaseA (10 mM Tris-HCl, pH 7.6, 1 mM of EDTA, pH 8.0, 100 ug/ml RNaseA) and 25 ml 7.5 M KOAc were prepared and placed in the designated reservoirs. The Biomek added 100 μ l 10:1 TE to each well followed by a gentle pipetting up and down twice and then paused. This step took 15 min. The blocks were incubated at 37°C for 10 min. allowing better resuspension of the DNA pellets and RNaseA activity. Following incubation, the blocks were placed on the titer plate shaker at speed 5 for 2 min. to make sure the pellets were completely dissolved. The blocks were returned to the Biomek and baciso3 was continued. The Biomek added 50 μ l of 7.5 M KOAc to each well over a 10 min. period. Then the solution contained in the four blocks was pooled into one block by baciso4, a 15 min. step. Following the pooling there was only one block which contains DNA-acetate solution. The block was placed at -20°C for 45 min. The potassium acetate-cellular debris pellet was collected by centrifugation in the Beckman GS-6R centrifuge at 3000 rpm for 20 min. The block was returned to the Biomek and baciso5 was selected to transfer 550 μ l of DNA-containing supernatant from each well of the old block to a fresh block concluding the Biomek portion of the protocol.

The block then was removed from the Biomek and 1.25 ml of cold 100% ethanol was added to each well of the block using the repeat pipet. The block was covered with an acetate plate sealer and inverted three times to mix. The sealer was removed immediately and the block was incubated in an ice-water bath for 45 minutes. The DNA pellet was collected by centrifugation at 3000 rpm for 30 minutes in the Beckman GS-6R

centrifuge. Each well was washed with 1 ml 70% ethanol and dried briefly in the vacuum oven. To each well was added 40 μ l of dH₂O to dissolve the DNA. The block was placed on a titer plate shaker for 10-15 min. and then placed over night at 4⁰C to allow the pellets to completely resuspend. The block was briefly centrifuged at 1000 rpm to collect the solution to the bottom of each well. The DNA solution was pooled into a 12 x75 mm Falcon tube.

For final purification, the DNA was brought to 50 mM Tris-HCl pH 8.0 with the addition of 1M Tris-HCl pH 8.0. The solution was divided into 0.5 ml aliquots in 1.5 ml Eppendorf tubes and 500 μ l equilibrated phenol (in 50mM Tris-HCl pH 8.0) was added to each tube and mixed gently by inversion. The phases were separated by centrifugation for at 12,000 rpm for 5 min. at room temperature in a microcentrifuge. The upper aqueous phase was remove to a fresh 1.5 ml Eppendorf tube and 500 μ l 1:1 equilibrated phenol:chloroform was added to each tube followed by gentle inversion and centrifugation at 12,000 rpm for 5 min. at room temperature. Again, the upper aqueous phase was removed to a fresh 1.5 ml Eppendorf tube and 500 μ l isopropanol was added to precipitate the DNA. The tubes were inverted gently several times and incubated for 5 min. at room temperature. This was followed by centrifugation for 5 min. at 12,000 rpm at room temperature. The DNA pellets were washed twice with 500 μ l 70% ethanol, dried in a vacuum chamber for 5-10 min. and resuspended in 400 μ l 10:0.1 TE. Any DNA remaining on the sides of the tube was dissolved by pipetting buffer over the sides then the tubes were incubated at 37⁰C for 15 min. and stored over night at 4⁰C to ensure that all the large insert DNA was dissolved completely .

The DNA concentration was measured by determining the absorption at 260 nm. An uncut aliquot and an Eco RI restriction digest of 10-15 μ l of the DNA preparation were examined on a 1% agarose gel for chromosomal contamination, presence of RNA and restriction digestion fragment pattern. If this DNA contained little visually detected genomic DNA, then it was suitable for nebulization and as a template for direct

sequencing. Typical yields per liter of original cell growth for the cosmids was roughly 2.5 mg for the cosmids.

2.12 Cosmid Sequencing Strategy

A blending of sequencing strategies was developed over the period of this dissertation research and was optimized for the three cosmids sequenced in this study. The random shotgun sequencing method was used for the initial phases of sequencing and the directed sequencing approach was used for closure of cosmids W02H02 and W30B01. The directed approach uses specific subclones or the cosmid as template for thermocycling in conjunction with region-specific DNA primers.

2.13 Random Shotgun DNA Subclone Library Construction

The cosmid DNA was randomly sheared by nebulization (Roe, 1995). The size range of the sheared fragments was controlled by the percent of glycerol in which the cosmid DNA was suspended, the temperature, and the pressure of nitrogen gas passed through the DNA solution. The resulting fragments that have "ragged" ends with either a 3' or a 5' overhanging end were made blunt-ended by treatment with the Klenow fragment of DNA polymerase I and T4 DNA polymerase. The fragments were further treated with kinase to make them suitable for blunt end ligation with Sma I cut pUC18 treated with calf alkaline phosphatase. After end-repair and phosphorylation, the fragments were size-selected by electrophoresis on a low melt agarose gel using appropriate size markers. The DNA was recovered from the agarose by melting, treatment with phenol, and then treatment with phenol/chloroform. The fragments were used in a ligation reaction with Sma I cut, dephosphorylated pUC18 and electrotransformed into competent *E. coli*.

2.14 DNA Shearing by Nebulization

The nebulizer (IPI Medical Products, Inc., Chicago, IL) is a commercial product originally designed for use in creating fine droplets from liquid medications or nutrients allowing them to be aspirated by a patient.

A solution of large insert cloned DNA was placed in the bottom of the nebulizer bowl. Nitrogen gas was introduced from the top. The pressurized nitrogen gas was channeled into a small chamber that was in turn connected to a tube which extends through the sample to the bottom of the bowl. The flowing gas creates a vacuum, drawing the sample up the tube and out into the upper chamber where it collides with a protruding plastic surface breaking into fine droplets. The size of the droplets was inversely proportional to the flow rate of the nitrogen and the fragment size is inversely proportional to the viscosity and the temperature. Therefore, glycerol was added to the DNA sample to increase viscosity as well as to prevent sample freezing since the temperature of the nebulizer was reduced (Bodenteich, 1993).

Specifically, nebulization was performed on 50-100 μg of cosmid DNA in a final sample volume of 2 ml with 25% glycerol 1x TM buffer (50 mM Tris-HCl pH8.0, 15 mM MgCl_2) in a -20°C dry ice-ethanol bath. After 2.5 min. at 8 psi nitrogen, the sample was removed from the chamber, divided into four aliquots, ethanol precipitated and subsequently resuspended in 27 μl 1x TM buffer.

2.15 End-fill and Phosphorylation

The resuspended DNA fragments were treated with T4 polynucleotide kinase and Klenow DNA polymerase and T4 DNA polymerase by addition of 5 μl 10x kinase buffer, 5 μl 10 mM rATP, 0.25 mM dNTPs, 1 μl 3 U/ μl T4 polynucleotide kinase, 2 μl 5 U/ μl Klenow DNA polymerase, 1 μl 5 U/ μl T4 DNA polymerase and incubation at 37°C for 30 minutes. The reaction was halted by heating at 70°C for 10 minutes and addition of 10 μl agarose gel loading dye (0.02% bromphenol blue, 5 mM EDTA pH 8.0, 50%

glycerol).

2.16 DNA Fragment Size Selection

The end-repaired DNA fragments were size selected by separation on a 1% low-melt agarose gel. Electrophoresis was performed at 100 mA for 90 minutes with Hind III cut lambda DNA (Gibco BRL) and 100 basepair ladder (Gibco BRL) as size markers. Samples were removed by slicing agarose gel pieces from the 1-2 Kbp and 2-4 Kbp ranges. The gel pieces were heated to melt the agarose, equal volumes of TE equilibrated phenol was added and the sample tubes were vortexed. The extraction was repeated with 1:1 phenol:chloroform. The aqueous phase was ethanol precipitated and the DNA pellet was washed twice with 70% ethanol. After drying in vacuo, resulting pellets were resuspended in 11 μ l dH₂O.

2.17 Ligation of Fragments and Transformation of Plasmids

Examination of the relative concentration of the recovered sample was performed by electrophoresis of 1 μ l of the DNA fragment with 0.1 and 0.2 μ g pGEM as concentration standards and Hind III digested lambda and 100 basepair ladder as size standards. The remaining 10 μ l of fragment DNA was diluted by two-fold and a series of ligation reactions were set up using 2 μ l, 1 μ l and 0.5 μ l as the DNA fragment template volume with 2 μ l Sma I cut CIP dephosphorylated pUC18 (10 ng/ μ l), 1 μ l 10x ligation buffer, 1 μ l T4 DNA ligase (400 U/ μ l) in a 10 μ l reaction volume. Incubation at 40°C for 16 hours was followed by transformation into *E. coli* strain XL1-Blue MRF^r by electrotransformation. This procedure allowed the uptake of DNA by the passage of an electrical current at 2.5 KV through a chamber containing cells and DNA suspended in 50% glycerol. The procedure was performed at 40°C. Two μ l of ligation mix was added to 40 μ l of electrocompetent cells and incubated on ice for one minute. The cells and DNA were transferred to a cuvette and placed in the pulser chamber (Bio-Rad).

Immediately following the five microsecond pulse the cells were diluted with 1 ml cold YENB medium (7.5 gm yeast extract, 15 gm Bactotryptone in 1 liter) and transferred to a small Falcon tube. Recovery was for 60 minutes at 37⁰ C with shaking. Following recovery the cells were collected by centrifugation for 3 minutes at 3000 rpm. The medium was poured off and 200 ul fresh YENB was added along with 30 ul 20 mg/ml 5-bromo-4 chloro-3 indolyl β -D-galactosidase in dimethylformamide (XGal) and 30 ul 25 mg/ml isopropyl-thiogalactoside (IPTG). One hundred μ l of the cell mixture was plated on a LB plate containing 100 μ g/ml ampicillin (LB amp100). White colonies were picked to 1.5 ml LB amp100 medium in 96 deep well blocks following 18 hours incubation at 37⁰C. The blocks were incubated for 18 hours at 37⁰C with shaking at 300 rpm for maximum aeration. The cell pellets were collected by centrifugation at 3000 rpm for 5 minutes and stored at -20⁰C.

2.18 Isolation of Subclone Template DNA

The same protocol and method was used to isolate cosmid subclone templates in a 96-well format as was used for isolation of cDNA template DNA.

2.19 Directed Phase of Cosmid Sequencing-Closure

The criteria for finishing was modified after W06E08 was sequenced and prior to sequencing W020H02 and W30B01 due to the availability of a program, primOU, modified in our laboratory by Steven Kenton, ACGT, and originally obtained from Southwest Medical Center, Dallas as the primo program. In addition, the acquisition of a MerMade (Southwest Medical Center, Avantech, Dallas) oligonucleotide synthesizer enabled the inexpensive synthesis of large numbers of single-stranded oligomers which could be used as primers in thermocycling reactions.

For W06E08 shotgun sequence data was collected from 980 reactions. This was sufficient to allow the data to be assembled into a database using Phred/Phrap (Ewing,

1998; Green, copyright 1994-1996) which required no custom synthetic primer-directed sequencing for finishing.

For cosmids W020H02 and W30B01, 768 and 576 thermocycle sequencing reactions were completed, respectively. Fifty-two 20-mer primers were designed by primOU for W02H02 and 61 for W30B01 with a melting temperature between 59-61⁰C, and a distance from the ends of the contiguous sequences of not less than 90 bases and not more than 1000 bases for closure.

Section 3. Computer Methods for Data Analysis

2.20 EST Sequence Analysis

The collection of EST sequences presented a data type which differed from genomic sequences in several ways. Although each sequence represented an expressed gene, in most cases there were two sequences from each of the expressed genes only some of which overlapped. As additional cDNAs were sequenced, the number of contiguous sequences increased rather than decreased in contrast to a genomic sequencing project. To accommodate this difference, each EST was handled as a separate file which allowed each to be submitted to the GenBank, analyzed for homologs in the GenBank databases, and made publicly available on the ACGT website (<http://www.genome.ou.edu>). However, the EST sequences also were assembled to allow ESTs from the same gene to align and to generate a consensus sequence in the Unigene database. The strategy for analyzing the EST database is shown in Figure 4. Three directions were taken to yield: 1) high quality ESTs for submission to GenBank 2) a method for determining the endpoint of library sampling and 3) an assembled Unigene database reduced in sequence redundancy.

For each EST, an experiment file (Staden) was created containing the trace file (the raw sequence data) and library specific information. Automatic processing of each EST sequence was done via a series of scripts obtained from LaDeana Hillier,

Table 3. Clip and Clean EST processing scripts.

1. embellish_template	extract information from the template name, get library name from experiment file
2. reformat-scf.uwphred	reformat the trace file
3. the-big-one	call bases with ABI and phred and determines which sequences have overall poor quality (N ratio of 1:5). Makes the quality start and stop estimates based on trace quality, cut at first base <15
4. getscf_field2expfile	add the information from the trace to the experiment file
5. embellish_template_2	take the dye terminator information and the library name to extract information about the vector, adapter sequence and primer position.
6. clip-seq-vec	use vep_vector end point (Staden, 1992) to find the sequencing vector and mark those sequences which are completely sequencing vector
7. clip_left_seq_vec	repeat attempt to find the left cutoff point, using adapter sequence information and distance from primer. Tag if the poly T is not found on 3' end.
8. clip-seq-wep-left	cut the vector sequence off the left end
9. clip-seq-wep-right	cut the vector on the right end if detected, this indicates short enough insert to read through in single pass
10. check-wrong-adapter	is the wrong adapter sequence present, example if 5' adapter sequence seen and name indicates a 3' EST. A tag fails the EST.
11. blastn_vec_check	check for the vector again, trim sequence if necessary BlastN: S=133 S2=133 M=5 N=-11 W=8
12. extend_seq	can sequence be extended past the conservative initial quality estimate to the second tier of reasonable quality, are there high quality bases to the right of the first base with quality 15.
13. check_processor	checks for sequence length <100 bases, fail the short sequences.
14. screen.p	BlastN against the <i>E. coli</i> genome database, the <i>A. nidulans</i> mitochondrial sequence, <i>A. nidulans</i> ribosomal sequences. Contaminants are failed. S=170, S2=150, M=5, N=-11

15. expESTBlastx BlastX against non-redundant protein database
Matrix=blosum62, filter=seq
16. reversed checks for reversed clones
17. check_processor_2 check if traces judged by the-big-one "overall poor
quality" are worth keeping by checking similarity
information with other ESTs. Use Blast
information to extend the estimate of good quality
sequence.
18. flip_qz_qr bookkeeping to ensure the QR(quality right cut)
always contains the right most cutoff point of the
sequence, that if there is a QZ(quality extend) it is
the hiqual_stop.
- 19.exp2dbest create a dbEST submission file and place in
directory for GenBank submission and placement
on website

Table 4. Sequences used for screening mitochondrial and ribosomal sequences.

Ribosomal sequences:	<u>Accession number</u>	<u>definition</u>
	u77377	18S
	u40122	26S, partial
	u29859	28S, regions D1, D2
	u93686	5.8S
	x03564	5S, type 1
	x03567	5S, type 2
Mitochondrial sequences:		
	x00790	cytochrome oxidase subunit 1
	x01507	ATPase subunit 6, tRNA Arg, Asn
	x06960	cytochrome oxidase subunit 3, tRNA tyr
	x06961	L-rRNA, ATPase subunit 6, cytochrome oxidase subunit 3, 7 ORFs, tRNA pro, thr
	x07795	tRNA met , his
	x15441	NADH dehydrogenase subunit 3, cytochrome oxidase subunit 2
	x15442	ORF A3

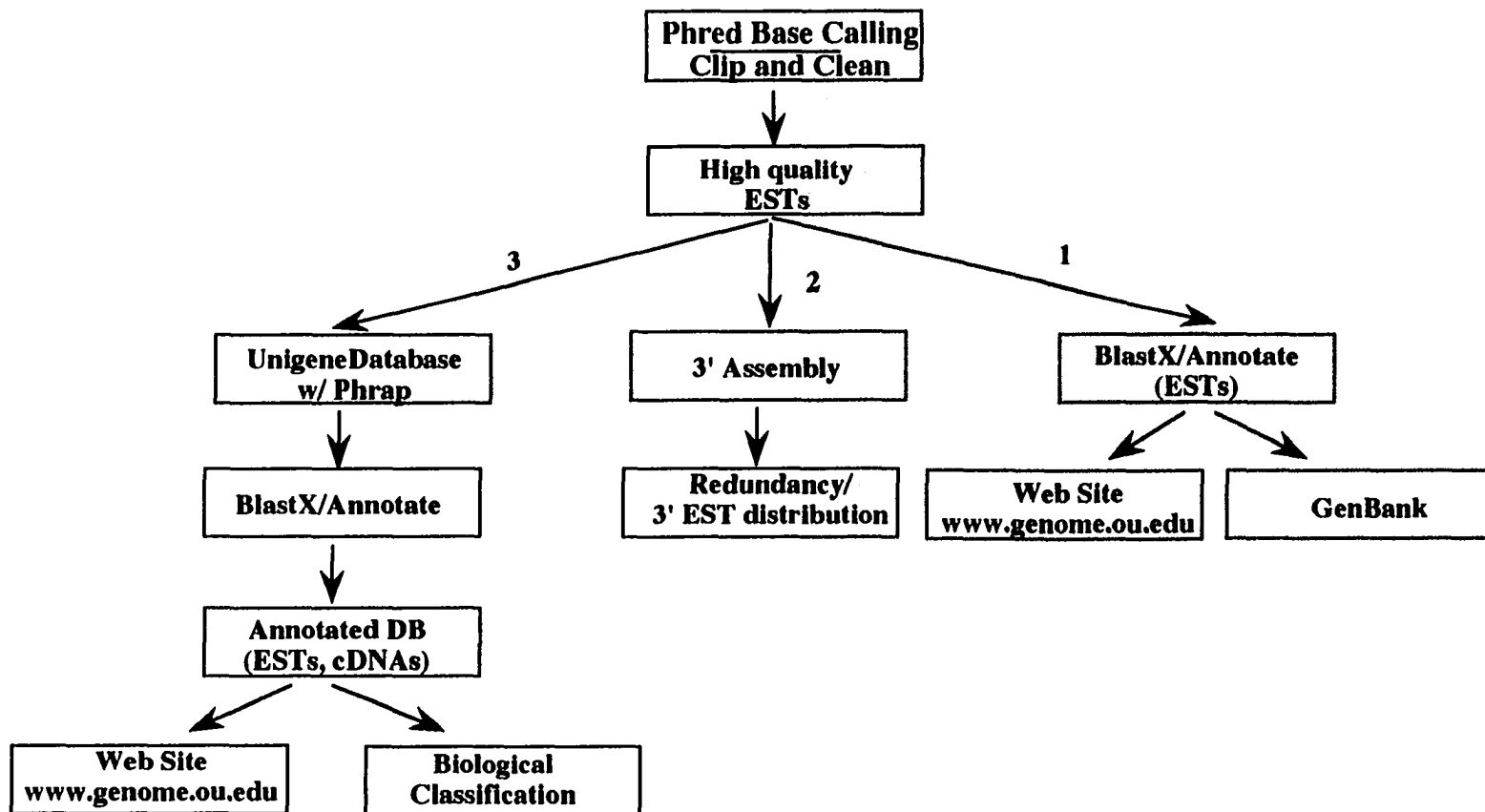


Figure 4. EST database analysis strategy. The arrows indicated the three directions for analyzing the *A. nidulans* ESTs. 1. The process of placing the high quality ESTs on the ACGT website and submitting them to Genbank. 2. 3' EST only assemblies for determination of library redundancy as sampling progressed. 3. The creation and annotation of the Unigene database. See text for detailed explanation.

Washington University, St. Louis and modified by Honshing Lai, ACGT. These scripts were linked or piped to allow passage of the EST from one script to the next. The scripts were piped in the order shown in Table 3, which also gives a brief description of the function of each script. "The-big-one" is a key script which recalls the bases using the editing program Phred (Ewing, 1998) and applies a quality value to each base called from 1-100. This quality evaluation was used in several scripts for an overall quality evaluation and endpoint determination. Table 4 lists the mitochondrial and ribosomal sequences obtained from GenBank used to screen against contaminants. The *E. coli* genome sequence was obtained from F. Blattner, University of Wisconsin prior to its publication. Figure 5 shows in a graphical format the order in which the ESTs sequences contained in the experiment files were examined by the piped scripts in the processing protocol to yield the high quality ESTs. In addition, the steps in the processing where a sequence could be removed from further consideration and moved automatically to a no-pass directory are noted.

Those ESTs which fail to pass the scripts were removed to specific no-pass directories that contained vector only, low quality, short, wrongend, ribosomal, mitochondrial, and *E. coli* sequences. Those which passed were termed the high quality ESTs.

The programs used in the Clip and Clean processing were the sequence editing program Phred (Ewing, 1998) which ranks each called base on a scale of 1-100 with a larger value indicating a higher quality. The Phred algorithm is based on peak height/width ratio and relative spacing as described earlier. BlastX (Altschul, 1997), which compares the six-frame conceptual translation products of a nucleotide query sequence against a protein sequence database and BlastN (Altschul, 1990), which compares a nucleotide query sequence against a nucleotide sequence database. The latter two programs use the Basic Local Alignment Search Tool algorithm. The parameters used with BlastN were:

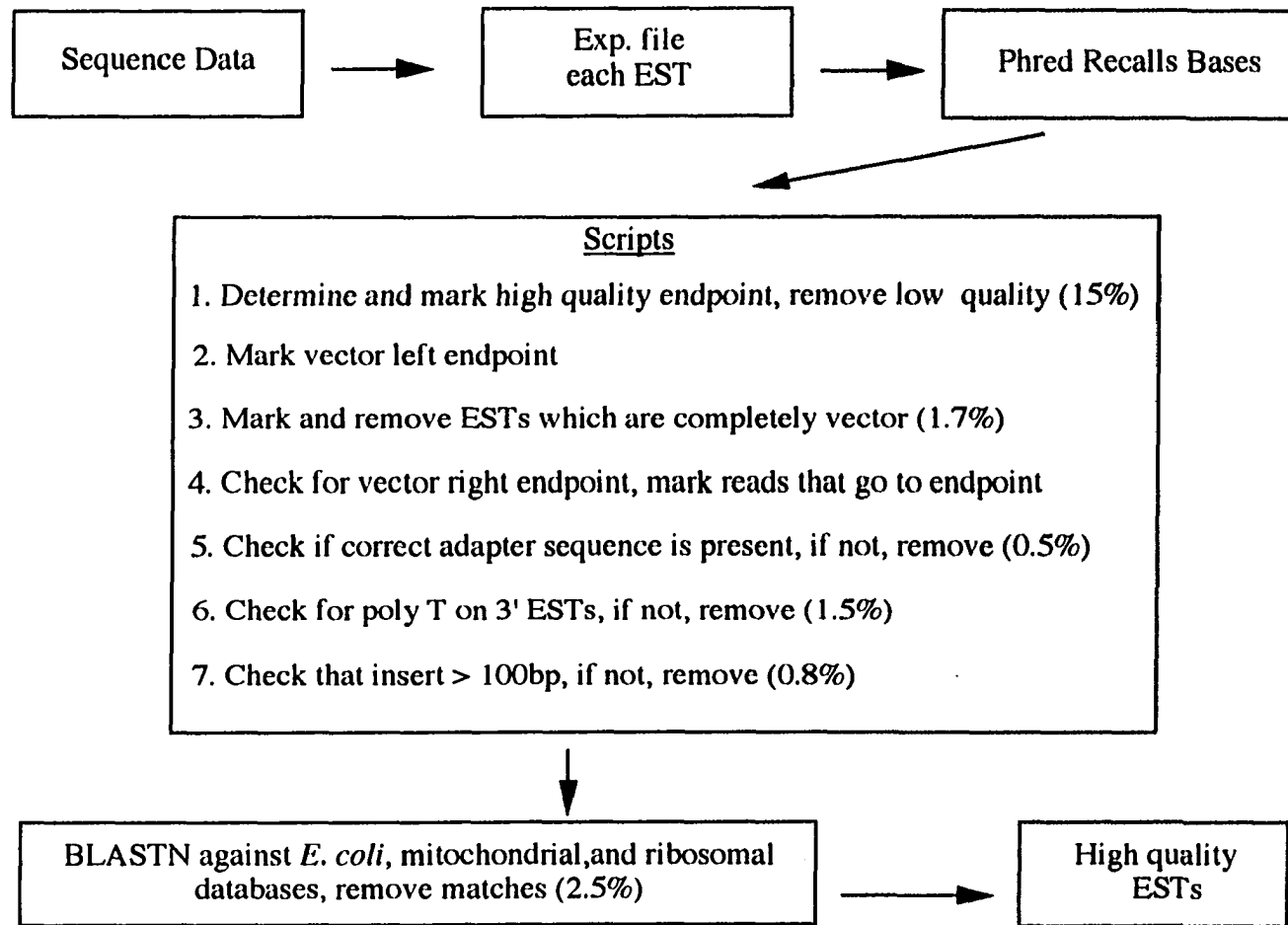


Figure 5. The "Clip and Clean" EST processing protocol. The boxes delineate steps in the processing of the EST sequence data to identify those sequences which pass the established criteria for high quality ESTs, see text.

S-limits the reported database sequence to those with at least one High Scoring Sequence Pairs (HSP) of the S setting,

S2-limits the reporting of individual HSPs to those with the S2 setting

M-score for a match

N-penalty for a mismatch

and the BlastX parameters were:

Matrix-amino acid blocks substitution matrix BLOSUM62 (Henikoff, 1991), the Blast default matrix, based on observed substitutions between segments that were less than 62% identical.

Seg filter, masked the low complexity sequences in the query sequence, default filter for BlastX

S-the cutoff for HSP score

Those sequences which were placed into the wrongend and low quality categories were examined manually. Those in the wrong end directory are checked for: 1). incorrect naming, a set of reactions identified by the wrong primer name. 2). A very short or absent poly A sequence tract with the expected 3' end primer and cloning sequence 3). a 5' reaction with the correct primer or cloning sequence not identified by the scripts. In all three instances the sequences showing these characteristics were removed from the wrong end directory and placed in the high quality database and in the first instance the EST name was also corrected. The ESTs in the low quality directory were examined to determine if reactions from a particular 96-well block of cDNA templates were of low quality. If so, the templates were reprecipitated with 7 M neutral potassium acetate as described in section 2.3 and the sequencing reaction repeated.

The high quality ESTs were submitted to the GenBank EST database (dbEST) by batch submission. Concurrently, a BlastX homology search was performed on each EST sequence. The complete report was saved and the top five homology hits with an

HSP of > 99 and a p value of $<e^{-4}$ were reported in the EST file that was placed on the ACGT website.

2.21 3' Assemblies of the EST Database

The level of redundancy in the cDNA library sampling was determined by cumulative 3' EST assemblies using the assembly program, Phrap (Green, copyright 1994-1996). The parameters of minmatch score=14, minscore=80 were empirically determined to be optimal for the assembly of the EST sequences, which were all greater than 100 bases in length, into relatively short consensus sequences (as compared to genomic) that required high stringency match. The percent redundancy was determined first by obtaining the number of individual gene alignments produced by each assembly and then by calculating the percent which represented new sequences. The inverse of this percentage gave the percent of redundant sequences seen. A best fit curve was generated from plotting the number of 3' ESTs vs the percent redundant sequences using CA-Cricket Graph III (Computer Associates Inc.). A report showing these results is presented in the Results and Discussion section.

2.22 3' and 5' Assemblies of the EST Database

Both the 3' and 5' EST sequences were assembled with the same minmatch and minscore as described above. This yielded a Unigene database assembly that was saved in two directories. Those ESTs which did not assemble with any others were considered singlets and stored in a singlets_dir; those that assembled with other EST sequences were considered members of a cluster, ESTs representing the expressed gene. The consensus sequences derived from the clustered ESTs were given "contig" identification names and were stored in a contigs_dir. The singlet and consensus cluster sequences (the contigs) were submitted for a BlastX search using the default Blosum 62 matrix, seg filter, High Scoring Sequence Pair (HSP) >99 and p value of $<e^{-4}$. Following the

BlastX search each of the two directories contained three files for every Unigene member. One file contained the sequence of the EST or consensus sequence in fasta format, a second file contained the complete BlastX output, and the third file contained a table with the BlastX header lines only.

2.23 Biological Function Outline

The analysis of the *A. nidulans* Unigene database entailed the organizing of the singlet and cluster members into a functionally related schema based on that developed by Monica Riley for *E. coli* (Riley, 1997) and improved by Selkov, et al. (Selkov Jr., 1998). This schema resulted in an *Aspergillus nidulans* Categories of Cellular Function as outlined in Appendix II. A BlastX search was performed on all members of the Unigene database, and the results were determined to be significant if they had a HSP >99 and p value < e^{-4} . The process of developing the Biological Function Classification for the Unigene database is shown graphically in Figure 6. Initially, a preliminary keyword list was developed, termed Keyword list 1, using a variety of sources that included the major metabolic pathway enzymes from biochemical texts (Bohinski, 1987; Stryer, 1995), pathway enzymes from the Metabolic Pathways Database (Selkov Jr., 1998) as well as reports from GenBank (Benson, 1996) Medline and Entrez (Schuler, 1996). The list was used with a series of scripts designed by James White, ACGT, to automatically scan the Unigene files containing the BlastX output descriptor lines for matching keywords to create an output file termed Keyword.hits that was organized into contig numerical order displaying the match. Following manual editing to examine for the multiple presence of a contig in the output file when a BlastX output which matched more than one keyword, the keyword list was corrected to yield Keyword list 2 and three output files were created: Keyword.hits 2, a list of contigs and BlastX descriptor lines which had matching keywords; Keyword.nohit, a list of those contigs which were not selected by a keyword, and Keyword.better, an exception file containing contigs with keyword matches but with

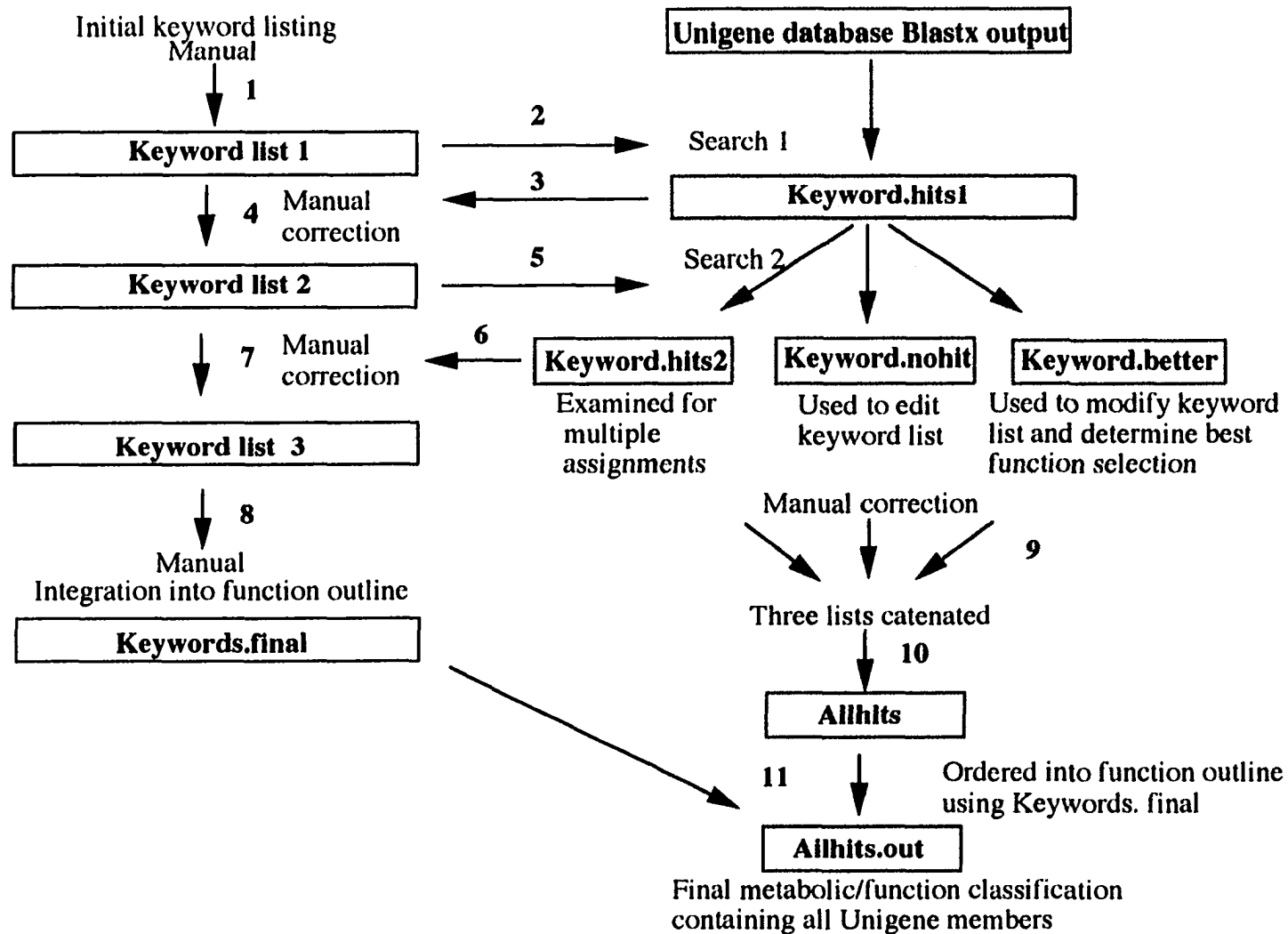


Figure 6. Steps used in creating the *Aspergillus nidulans* Biological Function Outline. Manual steps are noted. Other steps were done automatically with Perl scripts. In bold are the files created in the steps.

significant hits with better HSP and p-values for homologies not included in the keyword list. The Keyword.nohit and Keyword.better were examined manually for either addition or modification of the keyword list, yielding Keyword list 3. The Keyword list 3 was manually integrated into the Categories of Cellular Functions outline creating Keywords.final (Appendix I) containing 1375 keywords, 1041 primary keywords, 478 of which contain comments and 334 alternate keywords which allowed combining BlastX headers containing different spellings or terms for the same gene, protein or enzyme. The three files, Keyword.hits2, Keyword.nohit and Keyword.better were manually edited and then catenated into one file, Allhits. This file was automatically ordered into Keywords.final to yield Allhits.out which contained the Biological Function Classification including both the Contigs and singlet members of the Unigene database. The output from this is reported in Results and Discussion and listed in Appendix II.

Assignment of function was done using the homolog with the best HSP and p values, i.e. the best hit, as the source of gene assignment. The assignments are based solely on sequence homology as no biological testing was performed and therefore are "predicted" until biologically confirmed. In some cases, the best hit was a hypothetical protein or ORF while other hits in the blast report were to genes with known functions. Each instance of this was examined in the Keyword.better file. If a less significant homolog showed sequence similarity over the same region as the best hit and had a HSP value of >99 and p value < 10^{-5} then it was used to assign a function to the Unigene member. A notation (*) was placed next to the contig name to designate when this occurred (Appendix II).

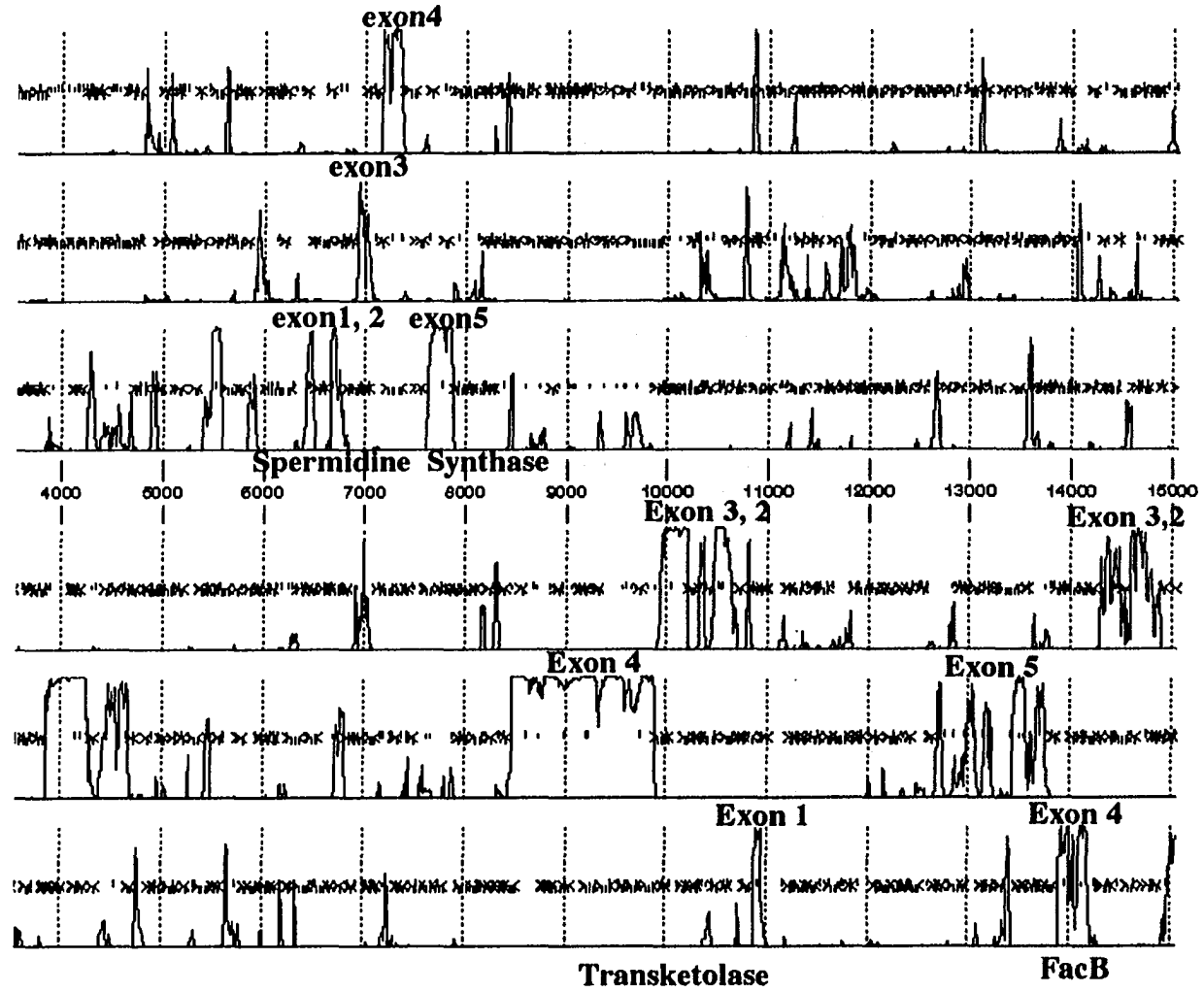
2.24 Annotation of *Aspergillus nidulans* Chromosome IV Cosmids

The sequence of cosmids W30B01 and W02H02 was completed as part of this dissertation research. Because the cosmids had a large overlap the data from both projects was combined to form a contiguous sequence for this genomic region for further

analysis (Results and Discussion).

Gene detection typically is more accurate if both intrinsic and extrinsic approaches are employed. The GeneMark program allowed an intrinsic approach to gene prediction which included evaluating particular properties of the DNA sequence without explicit referral to other sequences (Borodovsky, 1994). This approach required a training set based on *A. nidulans* non-redundant protein database entries to produce a matrix, *E.nidulans_3.mat*, that was kindly provided by Dr. M. Borodovsky. GeneMark produced a set of predicted exons that then were submitted for a Powerblast (Zhang, 1977) search using both BlastN and BlastX algorithms of the GenBank databases. The output from both the GeneMark analysis and the Powerblast search were visualized by Diana, a program developed at the Sanger Centre, Hinxton, UK and modified by James White, ACGT. This program allows examination of a DNA sequence when provided with files containing intrinsic data, such as that provided by GeneMark and extrinsic data, such as that obtained from BlastX results. Figure 7, part A shows the six frame output from GeneMark for a region containing the exons for three predicted genes, spermidine synthase, transketolase and acetate regulatory factor (FacB). The peaks indicate regions of coding potential as defined by GeneMark, the larger ticks indicate potential methionine ATG translation start codons while the smaller ticks indicate the alternative GTG codon which can serve as an alternative start codon. The lower ticks indicate one of the three stop codons TAA, TAG, TGA, and < or > mark potential intron/exon borders. Figure 7, part B shows the graphic output from Diana after integration of both the BlastX information and the GeneMark exon data for the same region as an example of the data used for annotation of the cosmid sequences reported in Results and Discussion.

A.



Chapter III

Results and Discussion

3.1 Introduction

The goals of the EST project were to obtain 3' and 5' end sequences of a majority of the expressed genes represented in the *Aspergillus nidulans* vegetative and asexual cDNA library, to determine their relative expression level and their probable identity, to establish a metabolic overview based on the probable identity of the expressed genes based on the EST sequences, and to demonstrate the usefulness of the EST sequences in annotation of genomic sequences.

3.2 EST database overview

The *A. nidulans* EST database was created by obtaining the sequence of both the 3' and 5' ends of isolated cDNA clones. Each EST trace file was assessed for quality by a series of automated, sequential scripts. A file, containing the sequence and all additional information pertaining to that EST was created for each sequence. After the sequences were examined for homology to the non-redundant protein database at the NCBI, this information was added to the EST file. The separate files then were combined into a single directory on the Unix-based Sun computer to form the EST database. The database was made publicly available by placing it on the Advanced Center for Genome Research (ACGT) website (<http://www.genome.ou.edu>) and depositing it into the GenBank EST database (dbEST).

A double-stranded cDNA does not necessarily represent a complete copy of the mRNA from which it was synthesized. Variable 5' ends can result if the first strand synthesis reaction does not result in a copy of the entire mRNA. In contrast, the 3' end which contains the 3' untranslated region and 3' end of the coding region, is anchored in a fixed position at the poly A tail since a poly dT primer was used to begin the first strand synthesis. Thus, because the 3' end sequences, which represent a fixed region of the

cDNA and include the 3' untranslated region are gene specific sequences, they were used to determine the complexity of the cDNA library and also the sequence redundancy at various points in sampling the library. The homologous 3' ESTs were aligned into groups where each group represents a cDNA family, i. e. a set of clones transcribed from the same gene. The number of ESTs in each group was an estimate of the abundance of a gene's transcript present in the library. In addition, the probability of detecting a new gene was estimated by periodically examining the redundancy in the 3' assemblies. A plotting of this probability helped determine when the productive endpoint of library sampling was reached.

Because the 5' ESTs were not anchored by the poly A tail as were the 3' ESTs and therefore were from variable positions of the original mRNA, they either could represent the 5' untranslated region and/or the coding region depending on the efficiency of the first strand synthesis and the size of the cDNA insert. The sequence of an entire cDNA could be obtained if the assembly of 5' and 3' ESTs resulted in a consensus cDNA sequence from an abundant cDNA and the 5' ends of the aligned EST sequences were sufficiently staggered. Since one goal of this study was to determine the abundance and distribution of EST species, library subtraction to eliminate redundant copies was not attempted. Thus, these results yield an estimate of the relative level of transcription of the corresponding genes at the time of mRNA harvest. In addition, the ESTs and the resulting consensus sequences provided a means for gene identification which facilitated annotation of genomic sequences because the ESTs often reveal the correct translation start site and intron-exon borders. This was demonstrated by the annotation of three cosmids from *A. nidulans* Chromosome VIII sequenced as part of this dissertation research and a gene cluster region of Chromosome VI.

Several conclusions could be drawn from analysis of EST databases. First, the abundance of different EST species reflected the level of representation of that cDNA in the library. Second, alignment of ESTs into contiguous sequences was extremely useful

in predicting genes in genomic sequences. Third, determining the sequence homologs in the public databases not only classified the cDNA represented by the EST but allowed the organization of the data into a biological function-based scheme such as that designed by Monica Riley for *E. coli* (Riley, 1997). This approach has been used as an organizational scheme for the genes identified during the genomic sequencing of *Haemophilus influenzae* (Fleischmann, 1995), *Methanococcus jannaschii* (Bult, 1996), and *Mycoplasma genitalium* (Bult, 1996; Fraser, 1995) and for the *Saccharomyces cerevisiae* genome (<http://wit.mcs.anl.gov/WIT2>). Classification of the expressed cDNA therefore presents a snapshot of the genes expressed at the time of harvest and serves as an index of the *A. nidulans* expressed genes.

3.3 EST Database Quality Summary.

Table 5a and b presents the EST sequencing results. Table 5a lists the number of 3' and 5' reactions which passed the Clip and Clean processing scripts called the high quality ESTs, as well as the no-pass sequences, those which did not (see Clip and Clean in Materials and Methods). Table 5b breaks down the no-pass ESTs into the specific categories in which each did not pass the processing criteria. The success rate for the 3' sequences was lower than that for the 5' sequences since the presence of the Poly A tract on the 3' end of a cDNA resulted in a number of lower quality sequence reads. This is because enzyme slippage occurs in the poly A homopolymer regions and the resulting shift of one or more bases in the sequence beyond the homopolymer region caused the trace to appear to have multiple overlapping sequences (PerkinElmer, 1995). The difference in success rate between 5' and 3' sequences also has been noted previously by Hillier et al. (Hillier, 1996) where their average success rate was 76% for 5' and 63.5 % for 3' cDNA sequences from 20 human tissue libraries. As can be seen in Table 5a, the present study had an efficiency of 89% and 78% for 5' and 3' end sequences, respectively. The resulting *A. nidulans* EST database represents 8645 cDNA clones.

Both the 3' and the 5' ESTs are present in the high quality database for 3847 cDNAs, only 3' ESTs are present for 1987 cDNAs and only the 5' ESTs are present for 2811 cDNAs. Sequences falling into the overall low quality no-pass category were examined manually and the 5' sequencing reactions were repeated (Materials and Methods). The continued failure of a subset of these reactions may be due to GC or AT rich sequence regions through which the 1:16 BigDye ampliTaQ FS reaction chemistry used for this work was unable to sequence.

Table 5. *Aspergillus nidulans* 24 hour vegetative/asexual cDNA library data summary. Final release 12-15-98

A.	Total Number of ESTs		12,482
	Number of megabases sequenced		4.268
		<u>5'</u>	<u>3'</u>
	High quality	6658	5824
	No pass	792	1611
	Total	<u>7450</u>	<u>7435</u>
	Success	89%	78%
B.	<u>Clip/Clean no pass:</u>		
	Completely vector	69	109
	Too short (<100b)	92	313
	Wrong end	33	240
	Overall low qual.	450	880
	E. coli	141	68
	Mitochondrial	5	-
	Ribosomal	2	1

3.4 Submission of EST Data

The Clip and Clean processing created two files, a homology file which was placed on the ACGT publicly accessible web site and a GenBank entry file, which lacked the homology information. Figure 8 shows an example of the output created for each EST sequence. The homology information includes the ten highest homology sequence

TYPE: EST
 STATUS: New
 CONT_NAME: Bruce Roe, University of Oklahoma, broe@ou.edu
 CITATION:
 LIBRARY: Aspergillus nidulans 24hr mixed vegetative and
 developmental cDNA
 EST#: c9a04a1.r1
 CLONE: Aspergillus nidulans c9a04a1
 SOURCE: Rodolfo Aramayo, Texas A&M University, raramayo@bio.tamu.edu
 P_END: 5'
 SEQ_PRIMER: T3
 HIQUAL_START: 1
 HIQUAL_STOP: 225
 DNA_TYPE: cDNA
 PUBLIC: unpublished
 COMMENT: This clone is available from the Fungal Genetics Stock Center;
 contact the curator, Dr. Kevin McCluskey
 (fgsc@kuhub.cc.ukans.edu), for further information.

HOMOLOGy:

gnl d1007414	(D32070)	heat-shock protein 30	(HSP30..+1 262	5.5e-22	1
gi 168820	(M55672)	heat shock protein 30	[Neuro..+1 212	2.1e-16	1

SEQUENCE:

```

ATTAGACGCTTCAAACAAAGCATCCTCGCAATCCACAAGGCATCAATTTCCCTCGACATT
CTCATAAACAAACCCACGTCACAGGTACAATGGCTTTCTTCCCCCGCTACTGCTCAGGCG
ACTTCGCCCTTTGTTTCAGCTCCTCGACGACTACGATATGCACCAGGCCACCCGCCGAC
CAAACAAGAAGGTCACCAACGTGAGAACATTTGTTCTAAATTTGACGTCTACG
||
  
```

55

Figure 8. An EST homology file. This format without the homology information is used to prepared a GenBank entry file for submission to the Genbank dbEST.

matches found by a BlastX search of the NCBI nonredundant protein database. To date five batch submissions have been made to the NCBI EST database (dbEST) where they have been assigned accession numbers (Table 6). The April 1999 release from the NCBI dbEST listed the *A. nidulans* ESTs as the ninth largest collection of organism-specific ESTs.

Table 6. EST Accession Numbers and Deposit Dates into the GenBank dbEST

<u>Accession Numbers</u>	<u>Submission Date</u>
AA783056-AA788459 AA788523-AA788570	February 1998
AA965289-AA966707 AA966906-AA966919	May 1998
AI007487-AI007508	June 1998
AI209337-AI214027	October 1998
AI327546-AI328148	December 1998

3.5 Assessing Library Redundancy.

Since the *A. nidulans* library was oligo (dT) primed from the mRNA template's poly (A) tails, the 3' EST sequence provides the 3' end of a gene. Assembly of the 3' ESTs into homologous clusters revealed the complexity of the library as well as the number of new genes detected while library sampling was in progress. These 3' assemblies were based on the presence of the 3' untranslated region which was assumed to be gene specific. Those sequences which aligned during a Phrap assembly were considered to be transcripts from the same gene (Figure 9) and therefore grouped into a cDNA cluster.

The end sequencing of the cDNA library is an example of sampling with replacement. This differs from the sequence data obtained for a cloned genomic region where each position of the sequence is represented in the randomly generated subclones and the distribution of sequences can be mathematically expressed by a Poisson

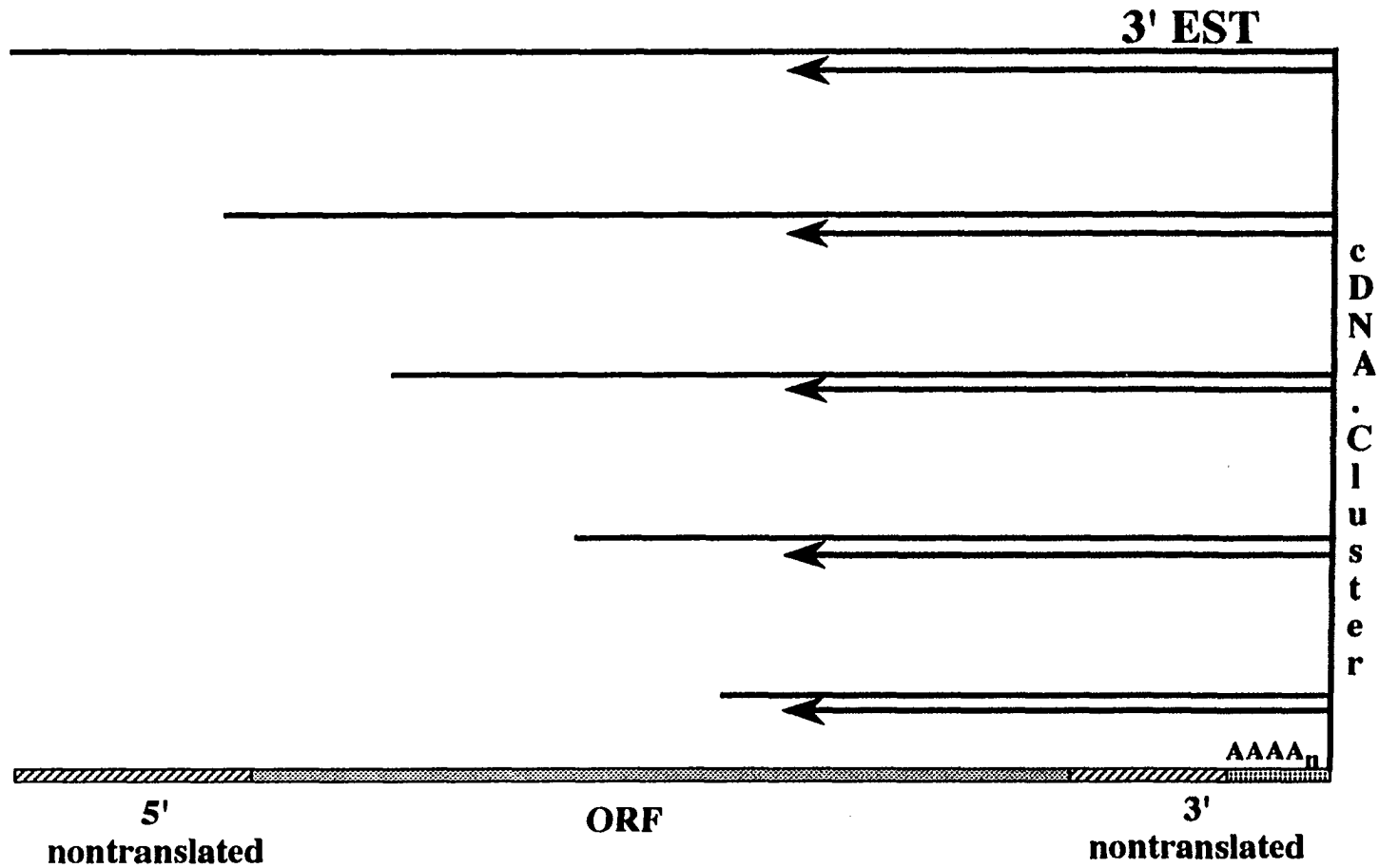


Figure 9. 3' EST Assembly of ESTs into a cDNA cluster. The lines represent the relative length of a cDNA to the entire length of the complete mRNA (Bar). The arrows represent the 3' ESTs sequenced from the cDNA templates.

distribution as described by Lander and Waterman (Fleischmann, 1995; Lander, 1988). Here, the calculations yield the theoretical number of sequences required for a certain fold coverage of a target sequence and the probability of completely sequencing the target region with that fold coverage. However, a cDNA library represents only transcripts and hence only the genes. Populations will be differently represented depending on the number and distribution of the mRNAs at the time of harvest, which in turn is dependent on the growth conditions. Finally, the probability of selecting and end sequencing representative cDNAs must be considered.

Table 7 lists the number of singlets, i.e. those ESTs which were not grouped with others and therefore represent nonredundant sequences, and clusters, i.e. those ESTs which were aligned or "clustered" by Phrap and represent transcripts from the same gene. Cumulative assemblies were performed at intervals once 200 new 3' EST sequences were obtained. The number of genes represented at each assembly point is given in Table 7 where the percent redundant sequence is simply the inverse of the percent of new genes found. A plot of the total number of 3' ESTs against the percent of redundant sequences is shown in Figure 10. As was expected, the percent of resampled sequences initially was low since most of the EST sequences represented newly sampled genes. As the sample size increased, the number of redundant sequences also increased and many of the new EST sequences could be clustered. This suggested that as expected, a population of redundant sequences was present in the library. Extrapolation of the data showed that a plateau occurred when an approximately 70-75% redundancy level was reached. The experimental evidence was consistent with the theoretical calculation since with 5824 3' EST sequences the redundancy approaches 70%. Since new genes were still being detected, but at a fairly low level, at least several thousand additional ESTs would have to be sampled to exhaust the library and find the rare cDNAs (Figure 10). It was not the purpose of these present studies to discover all the expressed genes in *A. nidulans*, but rather to determine the identity of the genes expressed in the

Table 7. Determination of the percent new genes by cumulative 3' EST assemblies of the *Aspergillus nidulans* database.

$$\% \text{ New Genes} = \frac{(\text{New S} + \text{C}) - (\text{Old S} + \text{C})}{\text{New Reads}} \times 100 = \frac{(300) - (165)}{200} \times 100 = 67.5$$

Total Reads	Singlets (S)	Clusters (C)	S + C =# genes	% New Genes	% Redundant
200	144	21	165	82.5	17.5
400	249	51	300	67.5	32.5
600	354	72	426	63.0	37
800	443	95	538	56.0	44
1000	526	120	646	54.0	46
1200	586	168	754	54.0	46
1400	630	214	844	45.0	55
1600	696	243	939	47.5	52.5
1800	755	272	1027	45.5	54.5
2000	799	311	1110	41.5	58.5
2200	857	336	1193	41.5	58.5
2400	920	362	1282	44.5	55.5
2600	971	398	1369	43.5	56.5
2800	1005	437	1442	36.5	63.5
3000	1053	474	1527	42.5	57.5
3200	1106	503	1609	41.0	59
3400	1131	552	1683	37.0	63
3600	1106	610	1716	16.5	69
3800	1161	630	1791	37.5	62.5
4000	1205	661	1866	37.5	62.5
4200	1229	695	1924	29.0	71
4400	1272	725	1997	36.5	63.5
4600	1316	747	2063	33.0	67
4800	1325	788	2113	25.0	75
5000	1376	815	2191	39.0	61
5200	1403	839	2242	25.5	74.5
5400	1429	867	2296	30.5	69.5
5600	1469	888	2357	27.0	73
5800	1507	911	2418	30.5	69.5

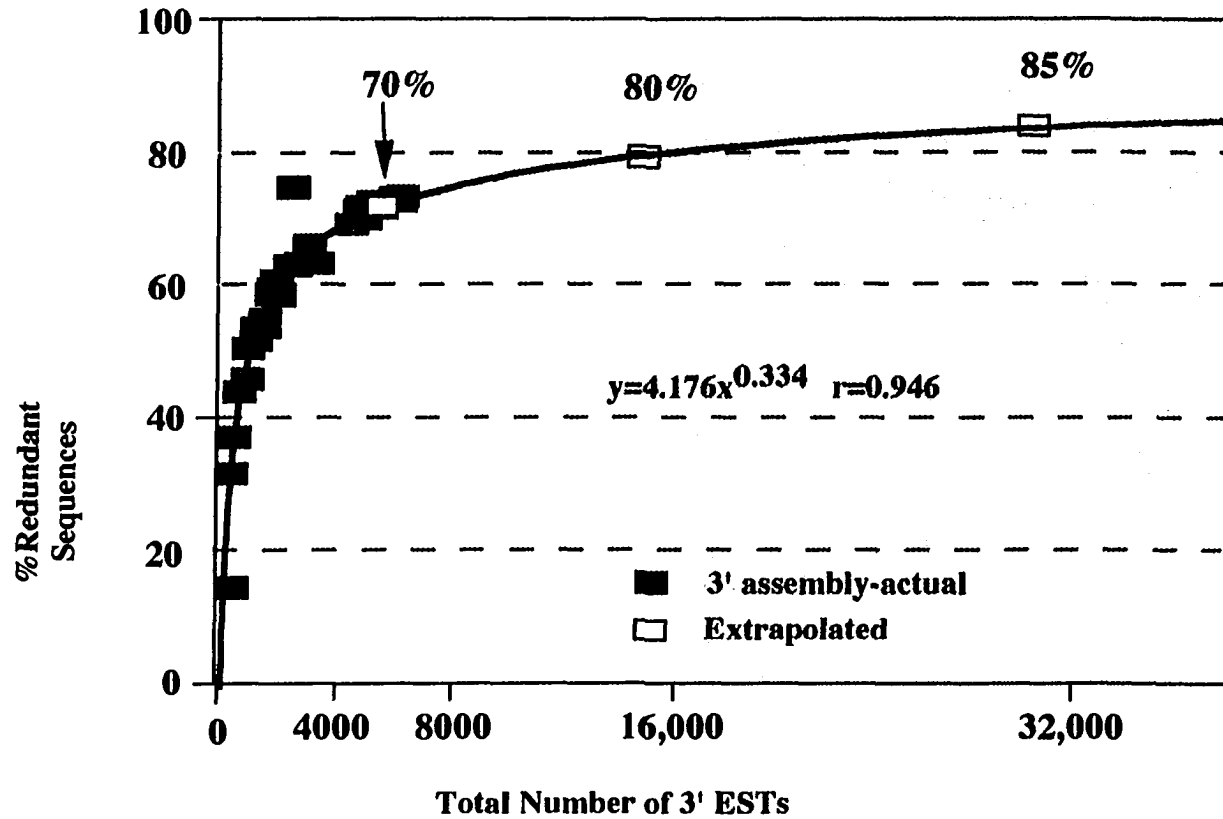


Figure 10. The percent of redundant sequences determined by cumulative 3' EST assemblies. The gray boxes represent the percent redundant at each assembly as determined in Table 8. The white boxes are extrapolated points determined by the formula given which was generated from the actual data points.

vegetative/asexual cDNA library. This library remains a good source of new gene information but the new information only can be obtained by acquiring a high number of redundant sequences or by switching to more efficient subtracted libraries. In addition, this redundancy would be expensive, and yield few new genes if sampling extended beyond the 70% redundancy level. As a comparison, in the libraries examined at Washington University Genome Sequencing Center, sampling also was discontinued when the level of redundancy reached 70% (Hillier, 1996).

Although beyond the scope of this present research, new, previously unidentified genes in the library could be detected by subtractive hybridization of the highly redundant members of the cDNA library (Adams, 1995) or by library normalization to reduce the frequency of the highly expressed populations (Bonaldo, 1996). Both of these techniques would allow a deeper examination of the library but would not yield the relative expression information which was a goal of the present research.

3.6 The Unigene Database.

Figure 11 shows the same set of theoretical cDNAs that represent the cDNA cluster shown in Figure 9 but with both the 3' and 5' ESTs aligned. The 5' ends terminate at variable positions depending on the success of the first strand synthesis so that staggered 5' ends often are present when the homologous EST sequences are assembled with Phrap. Such a multiple sequence alignment assembly yields a consensus sequence that is more informative than the individual EST sequences in two ways. First, the redundancy often yields a consensus sequence with multiple coverage that because of multiple sequence reads results in a more accurate sequence, and second, the consensus sequence could be extended in the 5' direction to yield a larger contiguous sequence than that obtained with the individual staggered 5' end sequences.

In this present study, the database assembled as described above and in Materials and Methods contained 1866 clusters with from two to 363 ESTs and 2429 single ESTs

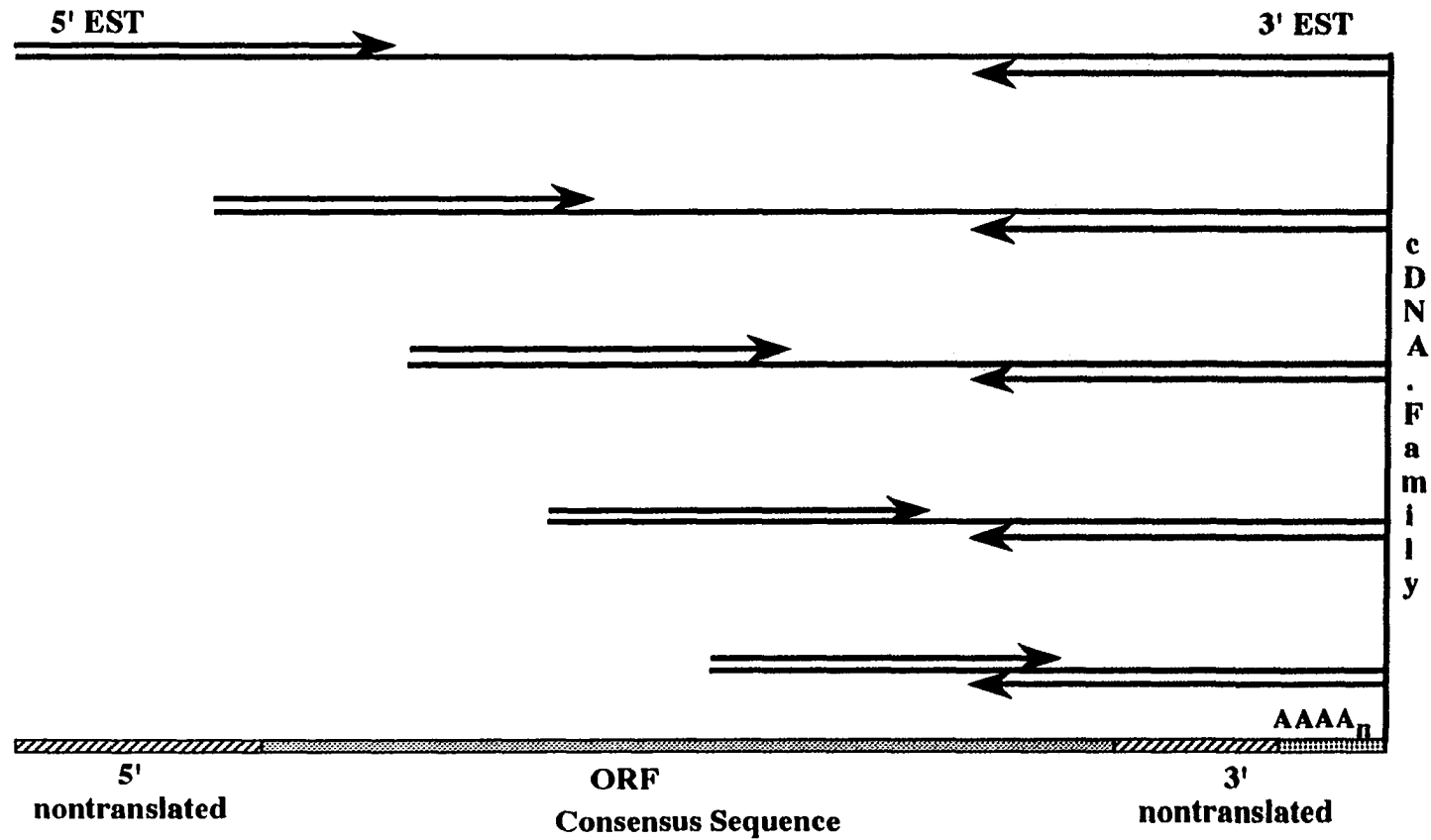


Figure 11. cDNA consensus construction by assembly into a unigene database containing both 3' and 5' ESTs. This figure is related to figure 9. The arrows represent the pairs of EST sequences generated for each cDNA. The lines represent the 3' anchored cDNAs and their relative coverage of the complete transcript. The bar represents the consensus sequence resulting from assembly of the ESTs.

resulting in a total of 4295 unique members (Table 8). This Phrap assembled data results in a so called Unigene database, a term borrowed from the grouped human ESTs found at the NCBI (Boguski, 1995). The final *A. nidulans* Unigene database had 1219 clusters (65%) that contain both 5' and 3' ESTs and therefore a consensus sequence containing at least a portion of the 5' end of a cDNA sequence, although it should be noted that this is not necessarily the complete 5' end of the gene. There also were 647 consensus sequences which were not assembled from both 5' and 3' sequences, 427 (23%) consensus sequences assembled from only 3' reads and 220 (12%) consensus sequences assembled from only 5' reads. Note that the higher number of 3' only EST sequences was expected since they have a common end, contiguous with the poly A tail, while the 5' ends were staggered and not necessarily overlapping.

Previous studies have determined common characteristics of mRNA and EST databases (Bishop, 1974 ; Soares, 1994). Reassociation kinetics analysis studies of mammalian somatic cell mRNA indicated that the population was distributed into three frequency classes, a very abundant class that consists of 10-15 mRNA species which represent 10-20% of the total mRNAs, an intermediate abundant class with 1000-2000 mRNAs that represent approximately 50% of the total mRNAs and a low abundance class containing the remaining 30-40% of the mRNAs (Bishop, 1974). The percent redundancy in an EST database therefore is expected to become greater than 60% once most of the mRNAs from the first two abundance classes are identified (Soares, 1994).

The *A. nidulans* Unigene database also has the three abundance classes suggested by Bishop (Bishop, 1974) (Table 9). The very abundant class was a group of 15 EST clusters, each of which consisted of greater than 0.5% of the ESTs in the Unigene database . The abundant class was made up of clusters of greater than two ESTs and represented genes sampled more than once. The contiguous sequences containing one (singlets) or two ESTs (representing a clone pair), made up the low abundance class and represented genes which were sampled only once.

Table 8. Assessing library complexity using assembly of both 3' and 5' EST sequences with Phrap version 98. The three frequency classes total 4295 unique Unigene members. Cluster size gives the number of ESTs belonging to the cluster. Horizontal lines delineate the three frequency classes.

<u>Cluster Size</u>	<u>Frequency of Cluster</u>	<u>Cluster Size</u>	<u>Frequency of Cluster</u>
1	2429 Low	33	3 Abund-
2	855	34	2 ant
3	316 Abund-	36	1
4	176 ant	37	1
5	110	39	2
6	55	40	2
7	47	41	1
8	28	43	1
9	46	45	2
10	21	47	1
11	20	51	1
12	14	52	4
13	13	53	2
14	4	54	1
15	9	55	1
16	12	57	1
17	3	63	1 High
18	4	64	1
19	5	65	2
20	4	71	1
21	7	77	1
22	2	80	1
23	4	81	1
24	2	83	1
25	1	86	1
27	2	88	1
28	3	112	1
29	2	124	1
30	1	137	1
31	1	145	1
		363	1

Table 9. Gene expression levels by EST abundance in the Unigene database for 12,490 entries.

<u>Total Reads</u>	<u>Class</u>		<u># Clusters/Singlets</u>
1704 (13.6%)	Very abundant	63-363 ESTs > 0.5%	15
7602 (60.9%)	Abundant	3-62 ESTs 0.02-0.5%	1038
3184 (25.5%)	Low Abundance	1-2 ESTs < 0.02%	755 +2429 Singlets

The 15 members of the very abundant class represented almost 14% of the ESTs and were noticed early in the library sampling. The low abundance class represented just over 25% of the ESTs suggesting that additional groups of genes represented in the library at a much lower level were sampled but at a level lower than the 30-40% suggested by Bishop (Bishop, 1974). However, the 70% redundancy found by 3' EST assembly for the last sampling point (figure 10) suggested that close to 100% of the cDNAs from the first two abundance classes have been found (Soares, 1994).

3.6 Determining the Number of Genes Identified in the Unigene Database.

To obtain an estimate of the number of genes represented in the EST database, both the 5' and 3' EST sequences were aligned using Phrap (Green, copyright 1994-1996). As described above, the resulting Unigene database contained two types of data. The first were the sequences that occurred only once in the Unigene database and are termed the singlets. The second was the contigs which are the consensus sequences derived from the clusters, the aligned EST sequences which have homology as determined by the Phrap assembly criteria (Material and Methods section 2.20). In addition, 367 of the 2429 EST sequences in the singlet category had their clone pair sequence aligned in a consensus sequence. There also were 718 clone pair members in

the singlets group whose sequences did not overlap and so could not align with each other to allow formation of a consensus but represent sequences from the 5' and 3' end of the same cDNA clone. Both of these categories were taken into account in determining that 1703 genes were represented in the singlets data category as shown in Table 10. An examination of the ESTs contributing to the consensus sequences showed that 10,061 ESTs were assembled into 1866 clusters. There were 742 clusters which shared a clone pair EST with other clusters. These were subtracted from the total to yield 1495 aligned EST consensus sequences representing 1495 unique genes. By totaling the 1703 genes represented in the singlets and the 1495 genes represented by the aligned ESTs it was concluded that 3198 genes were represented in the Unigene database. Since the estimate for the total number of genes present in the *A. nidulans* genome ranges from 8,000 to 12,000 (Kupfer, 1997; Timberlake, 1978) and see 3.30 Determining Gene Density), these 3198 genes in the Unigene database represent roughly one third of the *A. nidulans* predicted genes. Therefore, approximately one-third of the *A. nidulans* genes are expressed under the specific growth conditions which yielded the cells harvested for the cDNA library, a number consistent with the typical mammalian cDNA library which, even after subtraction and/or normalization yields only slightly greater than 30% of the postulated number of genes (Marra, 1999).

3.7 Identity of the Unigene Database Members, a Biological Classification for Cataloging the Expressed Genes.

An important goal of this study was to identify the genes represented in this vegetative/asexual *A. nidulans* cDNA library by comparison to known genes in the public databases. The predicted identity of the sequenced cDNAs and thus the expressed genes allows grouping by cellular roles and molecular families to facilitate data analysis. Several organizational schemes have been developed for this process (Riley, 1997;

Table 10. An estimate of the gene number in the Unigene database.
containing 12,490 ESTs.

Total ESTs in Singlets-		2429
Singlets with pair member in Clusters-		<u>-367</u>
		2062
3'and 5' EST pairs in Singlets-	$718/2=$	<u>-359 pairs</u>
Number of genes represented in Singlets-		1703
<hr/>		
10,061 aligned ESTS-		1866 Clusters
Number of Clusters sharing pair member	$742/2=$	<u>-371</u>
Number of genes represented in Clusters-		1495
<hr/>		
		1703
		<u>1495</u>
Total genes represented by Uni gene database		3198

White, 1997; Selkov, 1997). The Riley scheme was the earliest and one of the most comprehensive. It was designed for *E. coli* and provided for an exact cataloging of all proteins, and was based on both sequencing data as well as the extensive biological information available for this well-studied bacterium. More recently, genomic data from several additional organisms has been published and incorporated into versions of this classification scheme modified to reflect organism-specific information (Bult, 1996; Fleischmann, 1995; Fraser, 1995). Similarly, the genomic data for *Saccharomyces cerevisiae* and more than 30 bacterial and archeal species have been grouped into a comparable schema developed by Overbeek, et al. (WIT2 version 2.3, <http://www.cme.msu.edu/WIT2/>; Overbeek, 1997) called WIT2. In the study presented here, the seven major classes from the organizational scheme of Riley were used and a recent WIT2 analysis has been performed by Overbeek's group (Ross Overbeek, personal communication). When useful, each class was expanded to provide categories for all the Unigene members (Table 11). The bacterial genomic classification scheme of Riley lacks categories for asexual development, and a large number of metabolic functions that reflect the chemoheterotrophic and adsorptive nature of fungi. Although no scheme can be complete and different interpretations, such as placement of some functions in more than one category can be argued, this present fungal specific scheme given below with Table 11 and detailed in Appendices I and II followed established models and allowed presentation of the assembled *A. nidulans* EST data in an organized fashion that easily was accessed, modified, and analyzed.

Each cluster in the *A. nidulans* Unigene database was examined for homologs in the GenBank non-redundant protein database using the same parameters as for the individual ESTs (Materials and Methods). The singlets were not reexamined since the results of a BlastX homology search was completed earlier on the individual ESTs. Appendix II contains the complete cataloging of all the database members, both clusters (consensus) and the singlets. As can be seen, each of the 1788 Unigene

Table 11. *Aspergillus nidulans* categories of cellular functions.

- I. Bioenergetics and Metabolism
 - A. Metabolism of Carbohydrates
 - 1. macromolecules
 - 2. energy reserve biosynthesis
 - 3. sugars
 - 4. Calvin cycle
 - B. Metabolism of Amino Acids
 - 1. biosynthesis
 - 2. degradation
 - C. Metabolism of Nucleotides and Nucleic Acids, Purines, Pyrimidines
 - 1. purine metabolism
 - 2. pyrimidine metabolism
 - 3. nucleotide metabolism
 - D. Biosynthesis of Lipids, Fatty Acids, Sterols
 - E. Aromatic Compound Metabolism
 - F. Sulfur Metabolism
 - G. Phosphate Metabolism
 - H. Nitrogen Metabolism
 - I. Metabolism of Cofactors
 - J. Energy
 - 1. glycolysis
 - 2. gluconeogenesis
 - 3. pentose-phosphate pathway
 - 4. tricarboxylic acid pathway
 - 5. fermentation
 - 6. glyoxylate cycle
 - 7. beta-oxidation of fatty acids
 - 8. metabolism of energy reserves (glycogen, starch, trehalose)
 - 9. respiration
- II. Cell Growth, Cell Division
 - A. Cell Walls
 - B. Biomembranes
 - C. Cytoskeleton, Organelle Biogenesis
 - D. Cell Cycle Control
 - E. Mitosis/Cytokinesis
 - F. Meiosis
- III. DNA Synthesis
 - A. DNA Replication
 - B. DNA Modification and repair
 - C. DNA Packaging
- IV. Gene Expression
 - A. Transcription
 - 1. RNA polymerase
 - 2. regulation, includes asexual development regulatory pathway
 - 3. processing
 - 4. tRNA synthesis
 - 5. degradation
 - B. Protein Biosynthesis
 - 1. Initiation, elongation, termination factors
 - 2. ribosomal proteins
 - 3. post-translational modifications

- 4. folding and targeting
- 5. Turnover
- V. Cell Processes
 - A. Cell rescue, cell defense, osmotic adaptation, starvation response, development (asexual, sexual), includes antibiotics, toxins
 - B. Cell signaling, Signal transduction
 - 1. kinases and second messengers
 - 2. G proteins
 - 3. cAMP
 - C. Transmembrane Transport
 - D. Classes of Enzymes, general cellular role
 - 1. oxidoreductases
 - 2. transferases
 - 3. hydrolases
 - 4. lyases
 - 5. isomerases
 - 6. ligases
 - 7. synthetases
 - E. Non-enzymatic classes
 - 1. zinc finger motif
 - 2. leucine zipper motif
 - 3. other regulatory proteins
- VI. Unclassified (significant homolog but function uncertain in *Aspergillus nidulans*)
- VII. Unidentified (significant match with ORFs)
- VIII. No significant homolog

sequences with a significant homolog in the non-redundant protein databases was assigned a potential function based on BlastX scores (see Materials and Methods) and placed into the categories of cellular functions (Table 12). Those Unigene sequences with no significant homolog were not listed individually because of their number but are enumerated in section VIII. Because the listing is presented in an outline form, keywords, which represent the words used to search the BlastX descriptors in each report are shown in brackets (< >) under each section to distinguish them from headings. They are the words used to search the descriptors in each BlastX report (Material and Methods). The clusters were identified by a contig number followed by a contig identifier, for example Contig750_q0b02a1.f1, while the singlets were listed by their EST identifier, for example c4d02a1.r1.

Figure 12 displays a graphical representation of the percentage of Unigene members in each of the seven Riley categories plus those with no known homology (i.e. the no match category). Only 31% of the Unigene members could be given a functional assignment based on sequence homology. An additional 4% were unclassified, i.e. those with a homolog having a defined function in another organism but an unclear role in *A. nidulans*; while 9% had homology with ORFs or predicted proteins, which in the case of strong matches with Unigene members lent validation to the predicted genes, but gave no indication of the function in either organism. The remaining 56% of the Unigene sequences had no significant homology to any gene or protein in the GenBank databases. These results were in good agreement with recent findings from *Neurospora crassa*, a related multicellular ascomycete where Nelson et al. (Nelson, 1997) reported that 57% of the ESTs from three tissue specific libraries had no significant homologs in the databases using a significance cutoff of $p < 10^{-4}$, a somewhat less stringent criteria than that used for *A. nidulans* (HSP > 99 and $p < 10^{-4}$). Therefore, the present data suggests that a significant number of the sequences in the *A. nidulans* Unigene database represent newly discovered expressed genes.

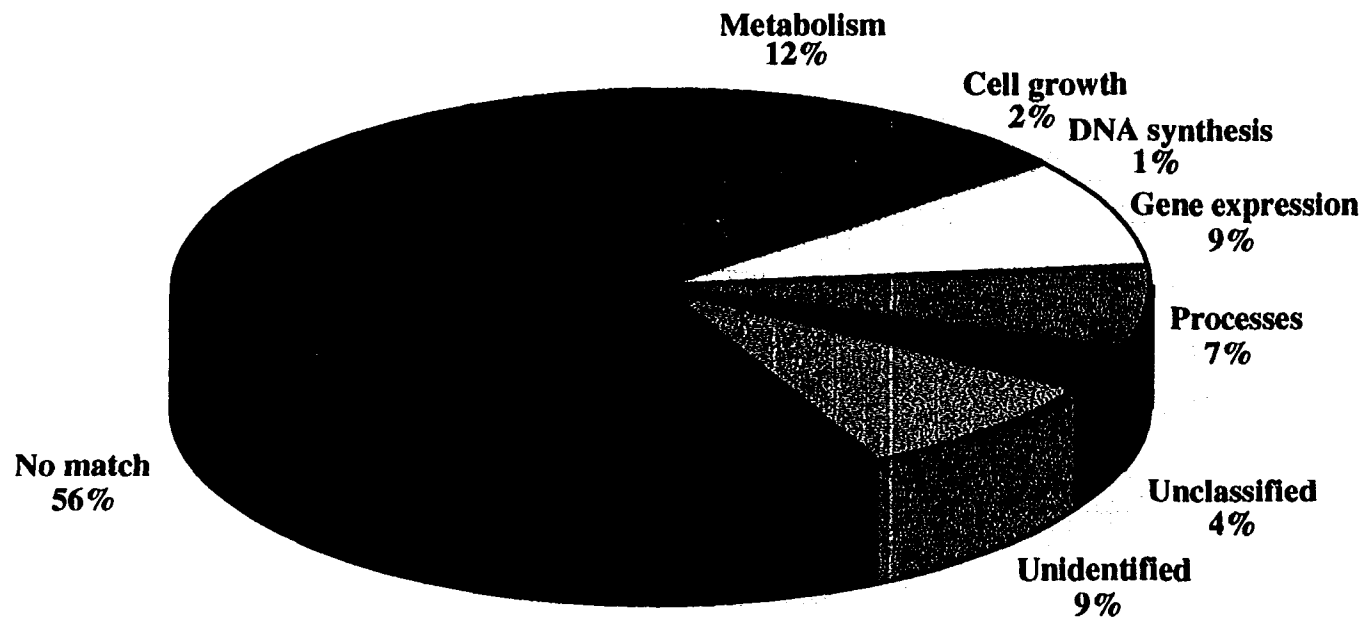


Figure 12. The Unigene database biological function classification by percent of members falling into each of eight categories

3.8 Unigene Representation in the *A. nidulans* Biological Classification Scheme.

In addition to the rapid detection of expressed genes, EST sequencing allows for determining the relative levels of gene expression by their representation in the EST library. The alignments which generated the *A. nidulans* EST Unigene database also revealed the number of homologous EST sequences in each cluster as well as the relative position of each EST in the consensus sequence as shown below in Figure 13.

```
Sequence Contig923
Assembled_from z4b03a1.f1.comp -557 382
Assembled_from z4b03a1.r1 -73 516
Assembled_from a5f07a1.f1.comp -46 1057

Sequence Contig1255
Assembled_from d3c05a1.r1.comp -430 770
Assembled_from d3c05a1.f1 -81 464
Assembled_from k0f07a1.f1 -68 524
Assembled_from k0f07a1.r1.comp 200 642

Sequence Contig1453
Assembled_from v4g03a1.f1 -117 723
Assembled_from v3g03a1.f1 -75 567
Assembled_from b0h05a1.f1 -65 466
Assembled_from u4b10a1.r1.comp 376 1063
Assembled_from b0h05a1.r1.comp 563 1006
```

Figure 13. An excerpt from the relationships file of the Unigene database showing the identity of the EST members in selected clusters and their relative position in the alignment used to generate a consensus sequence. Comp indicates that the complimentary sequence was required for alignment.

Thus, the percentage of genes assigned to each of the seven categories could be compared to the percentage of transcripts as represented by the EST clone pair sequences. In general, as shown in Figure 14 there was a direct correlation between the percent of the total genes and their relative expression levels. The notable exception was with the highly expressed genes in the "Gene Expression" category. Here, the percent of assigned genes (21.7%) was lower than the percent of total transcripts (32.6%). Table 12 indicates the number and percent of genes as well as the gene expression levels in the subcategories. A large number of genes, 286, were assigned to the protein biosynthesis category where the

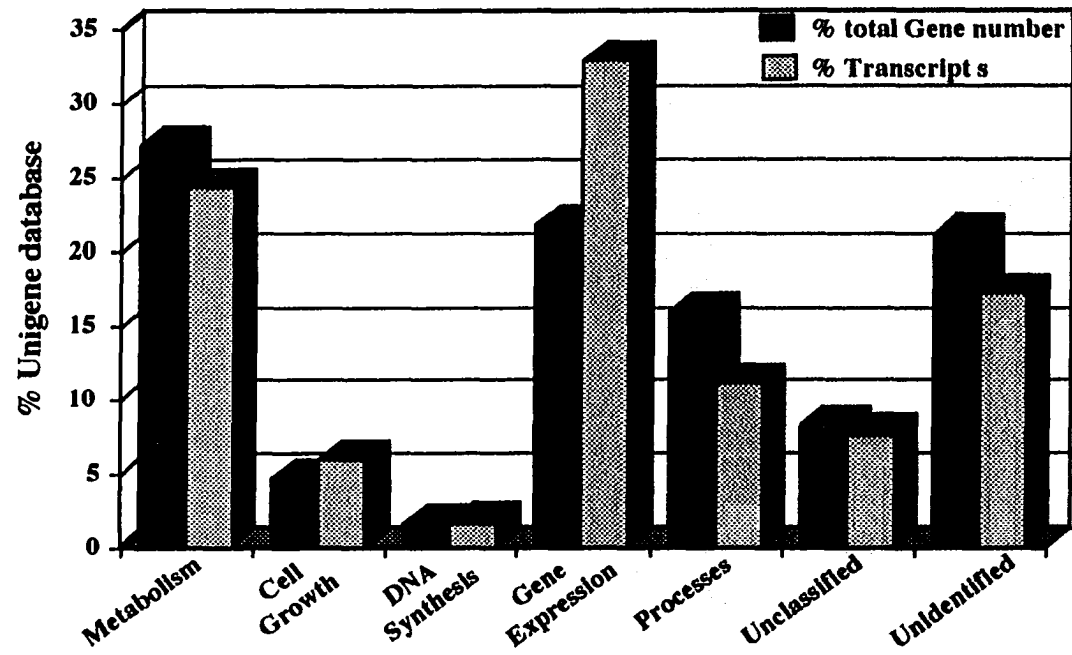


Figure 14. Gene vs transcript representation in the seven function categories for 3198 genes and 8612 cDNAs.

Table 12. Comparison of gene number and gene expression level for the categories of cellular functions for 1775 genes and 4604 cDNA clones . The percent values in the subcategories are the percent of genes or cDNA clones in seven categories.

I. Bioenergetics and Metabolism											Total	%Assigned		
	Carbo- hydrates	Amino Acids	Purines, Pyrim- idines	Lipids, Fatty Acids, Sterols	Aromatic Com- pounds	Sulfur	PO ₄ -	Nitrogen	Cofactors	Energy				
Genes	79 4.4%	47 2.6%	22 1.2%	43 2.4%	6 0.3%	2 0.1%	1 .05%	7 0.4%	27 1.5%	247 14.0%	481	27.1		
cDNAs	232 5.7%	100 2.5%	45 1.1%	84 2.0%	12 0.3%	2 0.05%	2 .05%	33 0.8%	64 1.6%	551 13.6%	1125	24.4		
II. Cell Growth, Cell Division														
	Cell Walls	Bio- membranes	Organelle, Cytoskeleton	Cell Cycle	Mitosis, Cytokinesis	Meiosis								
Genes	12 0.68%	7 0.4%	30 1.7%	16 0.9%	14 0.8%	2 0.1%						81	4.6	
cDNAs	14 0.3%	14 0.3%	64 1.6%	23 0.6%	20 0.5%	2 0.05%						137	3.0	
III. DNA Synthesis														
	Replication	Modification Repair	Packaging											
Genes	9 0.5%	8 0.45%	9 0.5%										26	1.5
cDNAs	15 0.4%	8 0.2%	50 1.2%										73	1.7
IV. Gene Expression														
	Transcription	Protein Biosynthesis												
Genes	100 5.6%	286 16.1%											386	21.7
cDNAs	193 4.2%	1308 28.4%											1501	32.6

V. Processes

	Adaptation	Signalling	Transport	Enzymes	Non-Enzyme		
Genes	73 6.2%	76 6.5%	96 8.1%	25 2.1%	11 0.9%	281	15.8
cDNAs	142 3.5%	130 3.2%	168 4.1%	47 1.1%	16 0.4%	503	11.0

VI. Unclassified

Genes						145	8.2
cDNAs						347	7.5

VII. Unidentified

Genes						375	21.1
cDNAs						788	17.1

Total in no assignment category:

genes: 2034

cDNA:4008

level of measurable gene expression is high at 15.2% of the total assigned clones. Ribosomal proteins and chaperones are two classes of highly expressed genes in the protein biosynthesis category that contributed to this high level, representing 4.9% and 6.1%, respectively, of the total cDNA clone population. The other highly represented class was bioenergetics and metabolism, where over one-fourth of the genes were assigned and nearly one quarter of the transcripts were represented by cDNA clones.

Comparison of the *A. nidulans* Unigene database with the 1879 EST sequences reported for *Neurospora crassa* (Nelson, 1997) showed a slightly different gene expression pattern. Three hundred seventy-three clones from the three *N. crassa* libraries examined were placed into the seven Riley categories with the result that 45% of their cDNAs were metabolism related and 32.6% were involved in protein synthesis. It is interesting to note that no chaperone proteins were discovered in the *N. crassa* libraries and that the protein synthesis category consisted primarily of ribosomal proteins. Nelson et al. (Nelson, 1997) suggested that because only a small number of sequences were collected, only the very highly expressed genes have been observed. However, it remains a notable difference that no chaperone class of proteins was detected since this class represented a significant fraction of both the very abundant and abundant groups in the *A. nidulans* library.

3.10 An examination of the highly expressed Unigene members.

Table 13 lists the significant BlastX homologs for each of the 15 clusters making up the very abundantly expressed gene category. Nine consensus sequences have a significant homolog but only seven have predicted functions. Three of the clusters are members of the HSP30 family, a class of small chaperone proteins of the *S. cerevisiae* HSP20 family (Kusakabe, 1994), one cluster represents ubiquitin, a protein involved in protein turnover (Ozkaynak, 1984), one is a homolog of the spore-wall fungal hydrophobin, dewA, produced during asexual development (Stringer, 1995), one cluster

Table 13. The homologs of and EST representation in the very abundantly expressed class of the Unigene database.

Contig Number	Total ESTs in Contig	% of ESTs	Homologs with BLASTX HSP>99
1866	363	2.9	Heat shock protein 30, <i>Aspergillus nidulans</i>
1865	145	1.2	Ubiquitin, <i>S. cerevisiae</i> , <i>Candida albicans</i>
1864	137	1.1	Heat shock protein 30, <i>A. nidulans</i>
1863	124	1.08	Metalloproteinase, <i>A. fumigatus</i>
1862	136	1.0	Heat shock protein 30, <i>A. nidulans</i>
1861	112	0.9	Spore-wall fungal hydrophobin, <i>A. nidulans</i>
1860	88	0.7	ORF W02A2.g <i>C. elegans</i>
1859	86	0.69	Chitinase, <i>A. nidulans</i>
1858	83	0.66	Glucose repressible gene, <i>Neurospora crassa</i>
1857	81	0.65	no significant homologs
1856	80	0.64	no significant homologs
1855	77	0.62	no significant homologs
1854	65	0.52	no significant homologs
1853	64	0.51	no significant homologs
1852	63	0.5	no significant homologs

was chitinase, an intracellular enzyme which may be involved in cell wall remodeling, growth and development (Takaya, 1998), and one was a metalloproteinase, which is an extracellular peptidase involved in protein degradation (Ramesh, 1995). Two of the clusters had homology to genes which have no known function; one to a *C. elegans* ORF and the other to a glucose repressible gene of *N. crassa*. The remaining six very abundantly expressed clusters had no significant matches in the GenBank databases. Each cluster represents expressed sequences from a single *A. nidulans* gene. Therefore, roughly 40% of the very abundantly expressed genes found in the *A. nidulans* library have no known homologs. It is noteworthy that 39 of the 100 most highly expressed genes represented in the Unigene database have no significant matches in the GenBank databases, suggesting that a large number of new genes are represented in the *A. nidulans* Unigene database. An expanded discussion of the members of the Unigene very abundant class is given below.

3.11 Heat Shock Protein 30 Representation in the Unigene Database.

The Heat shock protein (HSP) 30 representation in the *A. nidulans* cDNA library was striking, as it included three of the 15 very abundant class and three additional HSP30 homologs from the abundantly expressed class. Although all six had their highest alignment score with accession number D32070, the only *A. nidulans* HSP30 in GenBank, they represented six different HSP30 genes as discussed below and contributed significantly to the protein expression category. A total of 684 HSP30 sequences were present which represented 5.5% of the total EST population from only six distinct EST clusters (Table 14). The relationship between these six *A. nidulans* families and to other HSP30s also was examined. Heat shock proteins in general serve as molecular chaperones that are reversibly associated with nascent proteins to assist their transport in the cell, their translocation across membranes and their proper folding. They also are produced in response to a variety of cell stresses and growth conditions such as

Table 14. HSP30 representation in the EST Database.

Family	EST#	Percent in Database
<u>Contig 1866</u>	363	2.9%
Contig 1864	137	1.1%
Contig 1862	112	0.9%
Contig 1820	28	0.22%
Contig 1812	23	0.18%
<u>Contig 1800</u>	21	0.17%
Total ESTs	684	5.5%

elevated temperature, glucose starvation, and chemical treatments (Gething, 1992; Lindquist, 1988). All HSPs have well conserved primary sequences and have been classified into either low molecular weight, HSP30, or the high molecular weight, HSP60, 70 and 90, families (Kusakabe, 1994). HSP30 occurs abundantly in plants while HSP70 predominates in animals (Lindquist, 1988). Kusakabe et al. (Kusakabe, 1994) isolated and sequenced a cDNA (Accession number D32070) from a heat stressed *A. nidulans* culture. Northern hybridization experiments revealed that there was a high level of HSP30 mRNA in *A. nidulans* grown at normal culturing temperature (30°C). Kusakabe et al. (Kusakabe, 1994) also found that the corresponding gene did not have heat shock response elements upstream of the coding region, suggesting constitutive expression.

To further investigate these HSP30 ESTs, each of the six HSP30 contiguous sequences in the Unigene database was examined for an open reading frame and translated. An amino acid alignment was done using the GCG Bestfit program (Genetics Computer Group, Ver. 5) for each against D32070. The percent identity for each alignment are shown in Table 15 along with the HSP and p values from the BlastX search.

Table 15. Unigene HSP30 members comparison with GenBank entry D32070.

<u>Unigene identifier</u>	<u>HSP</u>	<u>p value</u>	<u>% identity to D32070</u>
Contig1866	816	1.2 e-80	87.6
Contig1864	933	4.8 e-93	100
Contig1862	291	2.9 e-43	60
Contig1820	832	2.4 e-82	90
Contig1812	612	3.7 e-59	92
Contig1800	855	8.9 e-85	91.6

Clearly, Contig1864 represents the same gene as accession number D32070 characterized by Kusakabe since it displayed 100% identity. A multiple alignment, performed to examine the six for regions of sequence identity using the GCG program Pileup (Genetics Computer Group, 1996), is shown in Figure 15. When analyzed by a Blocks search

(Henikoff, 1991), each HSP30 matched block BL010031B that contains an FPK motif and domain 473, a Prosite (Bairoch, 1995) pattern for the HSP30 family which contains a consensus sequence also found in alpha-crystallin (Plesofsky-Vig, 1995). Both characteristics were marked in Figure 15 and showed that six families of closely related HSP30 proteins were present in *A. nidulans*. The closest homolog to the *A. nidulans* HSP30 family in the databases was *Neurospora crassa* accession number P19752 which had a 42% sequence identity to Contig 1864 and a 43.4% sequence identity to Contig1862, the most distantly related contig in the *A. nidulans* Unigene family. The *N. crassa* homolog was found in association with the mitochondria during periods of glucose limitation and elevated temperature (40⁰C) (Plesofsky-Vig, 1995). It is tempting to speculate that since the *A. nidulans* culture from which the library was grown on minimal media with 1% glucose at high temperature (37⁰C) (R. Aramayo, personal communication) rather than 30⁰C (Kusakabe, 1994), that under respiratory limitations at the elevated temperature populations of HSP30 were induced. In contrast, the *N. crassa* cDNA libraries studied by Nelson et al. (Nelson, 1997) were grown at 25⁰C and no HSP30s were represented in the ESTs which they generated. Thus, in addition to the constitutive population seen by Kusakabe, additional HSP30s possibly were induced at the elevated temperature. It would be interesting to examine directly the HSP30 population in an developing culture grown at 25⁰C or 30⁰C as well as a vegetative only culture grown on rich media with 2% glucose to investigate the conditions which modulate HSP30 induction.

The relationship of the *A. nidulans* HSP30 Unigene members to HSP30 genes in other organisms was examined using the clusters of orthologous groups (COGS) provided by NCBI. Clusters of orthologous groups describe divergent evolutionary relationships between proteins with a common ancestor. The clonal theory of evolution is based on the analyses of rRNA sequences and holds that all genes are passed directly to each succeeding generation (Lake, 1999). Variations in these genes result in altered Fig

	1				50
D32070	MSLFRTIPTP	GDFAPLFRLL	DDYDNHRSAR	G...H.ASVQ	SFAPREFVRE
1864	MSLFRTIPTP	GDFAPLFRLL	DDYDNHRSAR	G...H.ASVQ	SFAPREFVRE
1820	MSLFRTIPTP	GEFAPLFRLL	DDYDVHRSTR	G...Q.TVVQ	SFAPREFVRE
1866	MSLFRTIPTP	GEFAPLFRLL	DDYDVHRSTR	G...Q.TVVQ	SFAPREFVRE
1800	MSLFRTIPTP	GDFAPLFRLL	DDYDNHRSAR	G...H.ASVQ	SFAPREFVRE
1812	MSLFRTTTPSV	SSFAPLFRLL	DDYDNHLASR	NWGH.H.TSVR	SFSPREFVRE
1862	M-AFFPRYCS	GDFAPLFRLL	DDYDMHQATR	RPNKKVTNVR	TFVPKFDVYE
	51		<u>FPK motif</u>		100
D32070	SNEAYHLDGE	LPGIPQSNID	IEFTDPQTLV	IKGRSEREYH	SSSDDNKNDQ
1864	SNEAYHLDGE	LPGIPQSNID	IEFTDPQTLV	IKGRSEREYH	SSSDDNKNDQ
1820	SNEAYHLDGE	LPGIPQSNIE	IEFTDPQTLV	IKGRSEREYH	.SNDENKAEQ
1866	SNEAYHLDGE	LPGIPQSNIE	IEFTDPQTLV	IKGRSEREYH	.SNDENKAEQ
1800	SNEAYHLDGE	LPGIPQSNID	IEFTDPQTLV	IKGRSEREYH	.SNDENKAEQ
1812	TSDTYHLDGE	VPGVAQKID	IEFTDPQTLV	IKGRVERQYH	SGNTDDTQKQ
1862	QGDYRYLDGE	LPGVQSNI	IEFTDPQTLV	IKGHSKRNYH	HKSEPDTDDK
	101				150
D32070	ADTE.....NQAR	GESSEVAKTGE	KQVSTKKAAN
1864	ADTE.....NQAR	GESSEVAKTGE	KQVSTKKAAN
1820	AETE.....KPVQ	GESSEVAKTGE	KQISTKKAAN
1866	AETE.....KPVQ	GESSEVAKTGE	KQISTKKAAN
1800	AETE.....KPVQ	GESSEVAKTGE	KQISTKKAAN
1812	RQVE.....DENE	SSSNEVAKTSE	KQMTKSASSE
1862	SETSSVKSLQ	PTVEDWDEME	DATPAVEQTP	SLGPKEKAVE	KNSSTRSQEP
	151		<u>Domain hsp20</u>		200
D32070	KSRYWVSERS	VGEFQRTFTF	PTRVNQDDVK	ASLKDGI ^R LSL	VVPKAVPPTA
1864	KSRYWVSERS	VGEFQRTFTF	PTRVNQDDVK	ASLKDGI ^R LSL	VVPKAVPPTA
1820	KPRYWVSERS	VGEFQRTFTF	PTRVNQDDVK	ASLKDGI ^R LSL	VVPKAVPPTA
1866	KPRYWVSERS	VGEFQRTFTF	PTRVNQDDVK	ASLKDGI ^R LSV	IVPKAVAPSA
1800	KPRYWVSERS	VGEFQRTFTF	PTRVNQDDVK	ASLKDGI ^R LSV	IVPKAVAPSA
1812	KPRYWVSERS	VGEFQRTFSF	PSRVDQDRVR	ASLRDGI ^R LSV	VVPKEAPPNA
1862	AYKFWASERL	VGEFSRTFAF	PTRVDQDAVR	ASLNNGI ^R LSV	VLPKEPAPQL
Consensus	KWHRMERS	SGKFMRRFRL	PENVKVDEIK	ASMENGLTV	TVPK
	201				
D32070	<u>KKITIQ</u>				
1864	<u>KKITIQ</u>				
1820	<u>KKITIQ</u>				
1866	<u>KKITIQ</u>				
1800	<u>KKITIQ</u>				
1812	<u>KKITIQ</u>				
1862	KKVRVE				

Figure 15. The alignment of six HSP30 families found in the Unigene database aligned with the single representative from Genbank, AC# D32070. In red are the positions conserved in all *A. nidulans* members. The Prosite HSP30 consensus shown has the identity positions highlighted in blue.

phenotypes and the appearance of new species. The evidence from completed genome sequences has argued against the clonal theory as being the only method of gene acquisition. The examination of functional groups of proteins from eucaryotes, procaryotes and archaea suggest that both eucaryotic and prokaryotic genomes may be chimeras, obtaining transcription and translation related genes from an archaea while eucaryotes obtained the housekeeping genes from a procaryote (Rivera, 1998). Tatusov et al. (Tatusov, 1997) describe a comparison of proteins encoded in eight complete genomes from five phylogenetic lineages and delineated 824 clusters of orthologous groups (COGs) based on consistent patterns of protein sequences. These 824 COGs have between three and over 100 ortholog members, where orthologs are genes in different species that evolved from a common ancestral gene and result from a horizontal transfer (Koonin, 1997). Paralogs are genes that have arisen within a genome by duplication (Henikoff, 1997). Orthologs and hence the COG members are assumed to have retained the same function while paralogs have evolved new functions which may or may not be related to the original.

Koonin's group have determined a series of COGs and made them publicly available on the National Center for Biotechnology Information (NCBI) web site (<http://www.ncbi.nlm.nih.gov/COG/cognitor.html>). Any sequence can be compared to the COG database to determine its similarities to previously determined orthologous groups. Comparing individual HSP30 members of the *A. nidulans* Unigene database with the COG database revealed that the HSP30 Unigene members had significant homology to COG0071 a molecular chaperone COG containing four members, one each from *Methanococcus jannaschii*, an Archaea, *Senecocystis* a cyanobacteria, and two paralogs from *Saccharomyces cerevisiae*, the Ascomycete yeast. All four COG members had the conserved FPK motif within their consensus regions. The GCG multiple alignment program Pileup (Genetics Computer Group, 1996) was used to create a relational dendrogram for the *A. nidulans* HSP30 family, the *N. crassa* HSP30 and the

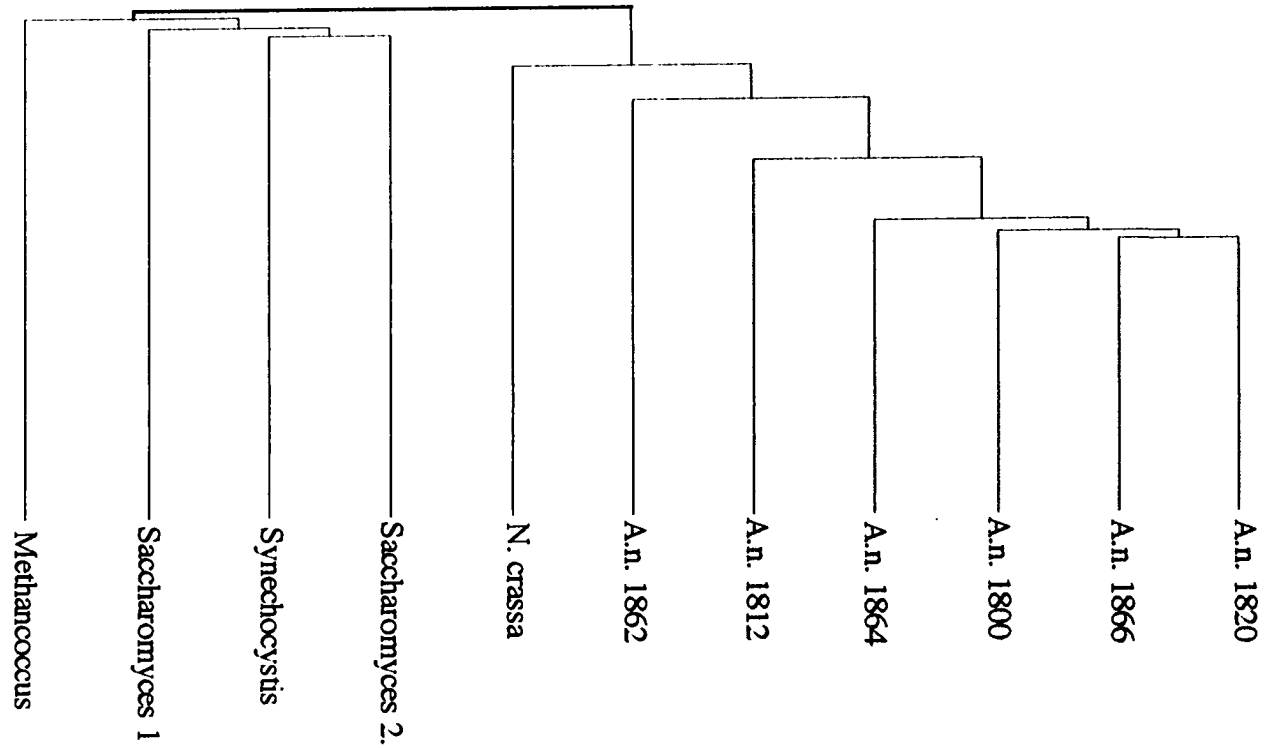


Figure 16. A dendrogram showing the sequence based relationship of the filamentous fungal HSP30 members to the members of the NCBI chaperone COG. A.n. is *Aspergillus nidulans*.

COG members. The results are shown in Figure 16. The filamentous ascomycete members, *N. crassa* and *A. nidulans*, formed a well-defined group and appeared to be comparably related to all four of the NCBI COG members. Since *S. cerevisiae* is not only a fungus but also an ascomycete, it is surprising that there is no difference in the filamentous ascomycete relatedness to the two *S. cerevisiae* and either the bacterial or archaeal representatives as might be expected. As seen by the distances from branchpoints in the dendrogram (Figure 16), the *Saccharomyces* HSP members of the COG were as closely related to the other COG members as they were to the filamentous ascomycete HSP30s. This observation suggested that the HSP30 progenitor had entered all three evolutionary groups by horizontal gene transfer. However, since there is a distant relationship between the HSPs of the multicellular ascomycete and the two *Saccharomyces* HSPs, this transfer most likely occurred after the evolutionary divergence of single and multicellular ascomycetes that occurred approximately 2.5 million years ago (Berbee, 1992).

3.12 Ubiquitin

Another member of the highly expressed clusters, Contig1865, had equivalent and high BlastX homology to three GenBank members, *Candida albicans* (Z54197), *Saccharomyces cerevisiae* (D29456), and *Nicotiana tabacum* (AJ223328) all three had an HSP of 1457 and $p=1.4e^{-148}$ and are ubiquitin homologs. In eucaryotes, ubiquitin is a highly conserved 76 residue protein involved in targeting intracellular proteins for degradation by conjugation of ubiquitin to the proteolytic substrates in an ATP-dependent reaction (Ozkaynak, 1984). The *A. nidulans* homolog had four perfect 76 amino acid repeats arranged in the head-to-tail fashion also seen in the yeasts and tobacco (Ozkaynak, 1984). This is an unusual structure for a precursor protein. Generally, spacer sequences are found between each copy which are lost during processing. There is a single amino acid difference, a serine for a threonine, in three of the repeats in the other homologs. In

the fourth repeat this residue is a threonine in all four organisms leading to 301/304 identical amino acids in this *A. nidulans* polyubiquitin compared to the fungal and tobacco homologs. It was not surprising to find that ubiquitin was expressed at high levels in the *A. nidulans* library since it is a key component in protein turnover. It is interesting to speculate that since the culture from which the library was derived was grown on minimal medium for 24 hours that the high level of ubiquitin was necessary to prepare the cells for this growth stage or that it could be part of a mechanism for providing carbon and nitrogen for a nutrient limited culture.

3.13 Metalloproteinase

Contig1863 was homologous to an *A. fumigatus* mep20 metalloproteinase (accession number JC4379). This metalloproteinase is a thermostable, zinc binding, extracellular protease which cleaves basic proteins such as histones and protamine. Comparison of the mep20 amino acid sequence with the Unigene homolog protein revealed a 57% sequence identity with the *A. fumigatus* sequence and conservation of the zinc binding domain AQDQATTTL**HEFTH**APGUY including the active site shown in bold type. The presence of this domain suggests that the *A. nidulans* Unigene member is a homolog of the *A. fumigatis* mep20 gene. The biological role of mep20 (Ramesh, 1995) and another homolog in *A. oryzae* (Tatsumi, 1991) is not clear but Tatsumi assumes that mep20 "serves to utilize substrates in the environment" since *A. oryzae* secretes a large amount, 300mg/liter, under normal culture conditions. Its high level of representation in the EST database from a culture entering carbon and nitrogen starvation is consistent with action as a "scavenger" during a time of increased cell death and lysis degrading external carbon and nitrogen sources for their absorption into the starving cell.

3.14 Dew A, a fungal hydrophobin

Contig1861, another very abundantly expressed cluster member has 100% amino

acid and DNA homology with the *A. nidulans dewA* protein, accession number P52750. The *dewA* protein is a spore wall specific hydrophobic protein which appears on the surface of the conidia and is one of several fungal hydrophobins needed for cell-surface hydrophobicity (Stringer, 1995). Since the *A. nidulans* culture was harvested during conidiation, it is not surprising that this gene was highly expressed in the cDNA library.

3.15 Chitinase

Contig1860 matched with 100% identity over 398 of 416 amino acids of the *A. nidulans* chitinase, GenBank accession number D87063. This nonsecreted chitinase which depolymerizes fungal wall chitin efficiently resulting in high molecular weight polysaccharides (Reyes, 1989) is thought to be expressed during morphogenesis (asexual development) (Takaya, 1998). This high degree of homology suggests that Contig1860 represents a chitinase similar to D87063 and is highly expressed because of the asexual development the *A. nidulans* culture was undergoing at the time of harvest. However, as can be seen in Figure 17A, the alignment of the first 42 residues of Contig1860 with D87063 only is possible if there is an 18 residue gap in Contig1860. The remainder of the alignment was 100% identical. The alignment of the cDNA version of the chitinase with the genomic conceptual translation suggests that there may be alternately spliced versions of this chitinase gene or that an intron, in the same reading frame, was included in the genomic translation and the cDNA reveals the true translation product. The second possibility is strongly suggested by examination of the nucleotide sequence shown in Figure 17B from accession number D87063 for the region covering the 18 residue gap. Both the 5' and 3' ends show the conserved intron border sequences, shown in bold (Mount, 1982).

3.16 The glucose-repressible gene

Another very abundantly expressed Unigene member, Contig 1859 which has

A. dbj|BAA35140| (D87063) chitinase [Emericella nidulans]
 Length = 416

Score = 831 bits (2124), Expect = 0.0
 Identities = 398/398 (100%), Positives = 398/398 (100%), Gaps = 18/398 (4%)

```
Contig1860  MSGYKTVGYFVNW-----AIYGRNYPQDLPAEKLTHILYAFANVRP 42
              MSGYKTVGYFVNW              AIYGRNYPQDLPAEKLTHILYAFANVRP
D87063      MSGYKTVGYFVNWVRTSCLLP IYISFTNDRQAIYGRNYPQDLPAEKLTHILYAFANVRP 60
```

B. 5' **gtacgt** **acttcctgtctcctaccaat**atataatctttcactaacgat **cgacag** 3'

V R T S C L L P I Y I S F T N D R Q

Figure 17. Comparison of Contig1860 with *A. nidulans* chitinase, accession number D87063. A. partial amino acid comparison of the translated Unigene chitinase homolog and the GenBank *A. nidulans* chitinase, chiB. The dashes represent the gap in the sequence alignment in Contig1860 required to align to D87063. B. The genomic region corresponding to amino acids 14-31 of accession number D87063. The translation is shown above the nucleotide sequence. Letters in bold are the conserved 5' and 3' intron border sequences.

59% amino acid identity and 73% conservation with *grg-1*, glucose repressible gene of *N. crassa* (Figure 18). A search of the Blocks database gave no matches in the database so it is not known which regions are important for function. This glucose repressible gene codes for a protein of unknown function but the level of *grg-1* mRNA increases over 50 fold during the first hour of glucose deprivation (McNally, 1988). Since in *N. crassa*, a number of activities are expressed when glucose is limiting including invertase, amylases and high-affinity glucose transport systems (McNally, 1988) the overexpression of a *grg-1* homolog is not surprising. If contig1859 is indeed a homolog of *N. crassa grg-1* its presence in the very abundant expression group would be consistent with the decreased levels of available glucose 24 hours after plating on the minimal medium.

```

sp|P22151|GRG1_NEUCR GLUCOSE-REPRESSIBLE GENE PROTEIN >gi|3014|emb|CAA32907|
(X14801)
      grg1 [Neurospora crassa]
      Length = 71aa

Score = 85.8 bits (209), Expect = 1e-16
Identities = 41/69 (59%), Positives = 51/69 (73%)
Frame = -3

1859: 344 METVKNAVNYVSESVQAGATASKE'TNKNVAKDSASLTSRATAAKDAVVDDKDEKSHDA 165
      M+T+KNA NYV + VQGA ATASKE NK+VAKDS+ + +R AA DA+ DK E HDA
grg-1:1  MDTLKNAAANYVGDKVQGATATASKEANKDVAKDSNQGVTGLRLNAAGDAISDKVSENKEDA 60

~1859: 164 KADVHKEAA 138
      KA+ HK+ A
grg-1: 61  KAEAHKQGA 69

```

Figure 18. Alignment of the *A. nidulans grg-1* homolog (1859) with the *N. crassa grg-1* protein. (+) means a conservative change in sequence. Alignment shows high level of identity especially at 5' end.

3.17 *C. elegans* ORF W02A2.g

Contig1858 represents a gene which has homology with 13 GenBank entries. All of the entries are ORFs either from *C. elegans*, *S. cerevisiae* or *E. coli* with significance scores ranging from $1.3e^{-13}$ to $1.4e^{-08}$. However, the actual homology was in all cases

only over a small 5' region of 54 amino acids from the 198 amino acid translation product from Contig1858. The best alignment was with the *C. elegans* ORF W02a2.g shown in figure 19 A. A search of the Blocks database showed a match with block BL01309, that represented a domain from the uncharacterized protein family UPF0057 (figure 19 B). Although the function of the gene product represented by Contig1858 in the Unigene database is not clear, it does represent a new *A. nidulans* gene belonging to a protein family which has not been previously reported in a multicellular fungus.

A.

```
>gnl|PID|e1188370 (Z82286) W02A2.g [Caenorhabditis elegans]
Length = 594
```

Minus Strand HSPs:

```
Score = 193 (67.9 bits), Expect = 1.3e-13, P = 1.3e-13
Identities = 35/54 (64%), Positives = 45/54 (83%)
```

```
C 1858: 1 MPFTASDICKLIFAFILPPLGVFLERGCADFLINICLTILGWIPGIIHAIYII 54
      MP T +DI K I A +LPP+GV++E+GCGAD +INI LTILG+IPG+IHA +II
W02A2.g: 1 MPITCTDIPKFCALLLPPIGVWMEKGCADLVINIVLTILGFIPGVIIHACFII 54
```

B.

```
BL01309 INILLTILGYIPGIIHACYVI
      ||| ||||| ||||| |||
C 1858 INICLTILGWIPGIIHAIYII
```

Figure 19. The characterization of Unigene member Contig 1858. A. The small region of homology between Contig1858 and the *C. elegans* ORF W02A2.g is shown. No other region in the *A. nidulans* 198 residue translation product aligned. The highlighted region is the conserved uncharacterized domain found in all other homologs. B. Shows the alignment of the domain found in Contig1858 with the Block BL01309 representing an uncharacterized protein family (Henikoff, 1991).

3.18 Additional examination of "no match" Unigene members.

It is clear that Unigene members with no significant homologs in the GenBank databases represent new genes and that it would be useful to obtain some indication of their cellular function. Therefore, each of the six Unigene members in the highly abundant class which showed no matches with a BlastX score of 100 or greater and a p value $< e^{-4}$ (Table 13) were examined in greater detail. To examine further if there were

any significant homologs in the databases, each was translated and the translation product was used for a BlastP search without the low complexity default masking function seg. For Contig1857, the BlastP showed a weak similarity to mucin and a blocks database search (Henikoff, 1991) showed a partial match with a subset of the selectin superfamily motifs that represented a transmembrane domain termed sushi:

```

mouse-FSFSSQCT
      |   |||
1857 -FCYRSQCT

```

Thus, there is some evidence that Contig1857 may be a membrane protein with a transmembrane domain.

The BlastX report for Contig1852 showed homology to amino acids 144-298 of a thyroid hormone responsive gene and the Downs syndrome region 1 alternatively spliced exon 1 with a score of 50 and e value $< 5e^{-5}$. A blocks search indicated a match with two regions of block BL00354 from the high mobility group 1 (HMG) DNA binding domain, a cell cycle dependent human embryogenesis transcription factor:

```

human-KRGRGRPRKQ
      | || || | The RGR motif
1852-RRRRGQPRQQ

human-AEEYGNTSSDSSDED
      || |   || The ESE motif
1852 -EEESGGGGGKSDEQ

```

A ProDom search (Sonnhammer, 1994) revealed a different domain in Contig1852 that had homology to domain 82878 of the *Drosophila* DNA binding protein k10. This domain contains the Wilms tumor motif that has been associated with developmental transcriptional regulation and binds to the DNA sequence CGC CCC CGC. In addition, since the domain 82878 overlaps block BL00354, Contig1852 may encode a gene product which may be developmentally regulated, have DNA binding capability, and thus may itself regulate transcription.

The remaining four Unigene members had no significant regions of homology to any known proteins, domains or COGs as revealed by BlastP, domain searches, or

search with the NCBI COGs database and thus their function remains unknown. Finally, all six Unigene member were examined with the Prosite program (Bairoch, 1995) from which Blocks is derived and the Stanford University motifs Identity program (Nevill-Manning) but this gave no additional information for any of the six Unigene members examined.

3.19 Summary

Over 8500 clones were isolated and almost 13,000 high quality ESTs were generated by sequencing both ends of the cloned inserts during the course of this dissertation research. A series of automated scripts was developed in collaboration with Hongshing Lai, ACGT informatics group, to trim each EST to high quality endpoints and to remove any contaminating sequences. A GenBank submission file was created for each high quality EST that was submitted to the GenBank dbEST database. In addition, each high quality EST was examined by a BlastX search against the nonredundant protein databases and the sequence as well as the homology information was made publicly available on the ACGT website to facilitate their further study.

The 3' ESTs were assembled into a Phred/Phrap database that allowed construction of a graph which displayed the percent redundant sequences, was a measure of library sampling and a means to determine a sampling endpoint.

A Phred/Phrap assembly of both the 5' and 3' ESTs create the *A. nidulans* Unigene database from which the approximate number of genes, 3198, represented in the library could be determined. A further analysis of this database revealed three expression levels, approximately 14% of the Unigene members belonged to the abundantly expressed group, 61% belonged to a moderately expressed group and 25% belonged a rarely expressed group. When the Unigene database members with known functions were placed into biological function groups, 44% of the Unigene members could be assigned functional groups such as bioenergetics, gene expression or cellular processes. The majority of the Unigene members, approximately 56%, had no homologs and

represented new genes.

An examination of the highly expressed genes revealed that six of the 15 very abundantly expressed Unigene members had no homolog in the GenBank databases, consistent with the observation that a significant number of new genes were represented in the Unigene database. Of the members with significant homologs, many were members of the HSP30 gene family. A study of these HSP30 related ESTs showed that the assembly had aligned them into six families, each representing a gene, five which had not been previously reported. A comparison of these six HSP30 sequences to the NCBI COG database revealed their homology to the chaperone COG but surprisingly they were not closely related to the *S.cerevisiae* orthologs. Six of the other very abundantly expressed genes had homologs which, in general, were related to a cellular response to growth under conditions of carbon and nitrogen limitation, leading to asexual development. Finally, an attempt to determine any potential function for the six remaining very abundantly expressed Unigene members via a conserved domain search had mixed results since two of the six showed either conserved motifs or domains that suggested a possible sub-cellular location and/or function while four did not.

3.20 *Aspergillus nidulans* Cosmid Sequencing.

Since one purpose of the above EST studies was to aid in genomic sequence annotations, the next section will describe the sequencing and annotation results for three *A. nidulans* chromosome VIII cosmids. Each completed cosmid initially was examined by a BlastN search of the GenBank nonredundant database which included the *A. nidulans* ESTs sequenced as part of this dissertation research. The results demonstrate the application of EST sequences to annotation of genomic DNA sequences.

3.21 Cosmid W06E08 Analysis

Sequencing of the Chromosome VIII cosmid was undertaken as part of a pilot

project to determine if the minimum tiled library created by Prade et al. (Prade, 1997) was suitable for the proposed genomic sequencing of *A.nidulans* . In addition, the sequence was used to estimate the approximate gene density and total gene number for *A. nidulans*. This particular cosmid was selected since it had been shown by hybridization to contain the development specific transcription factor, *brlA* (R. Prade, personal communication) whose sequence was known (Prade, 1993). The cosmid sequence determined by fluorescent sequencing methods could be compared with the region previously sequenced by radiolabelled methods to check the accuracy of both sequences. The environment of the *brlA* gene also was of interest to compare the sequence of the region with the existing genetic map (Clutterbuck, 1997) as well as for comparison with the sequences in the EST Unigene database.

The sequence of the 38,807 basepair cosmid, W06E08, was completed (Table 16) and, after annotation using the gene-finding program, GeneMark (Borodovsky, 1994) coordinated with the results of a BlastX (Altschul, 1990) search, was submitted to GenBank (accession number AC000133). A summary of this work was published (Kupfer, 1997). However, when the EST database became available, the cosmid sequence was examined for homology to these ESTs. Subsequently two clones, c5h06 and o4h01, were found to align with the *brlA* gene region. Included here is a summary taken from the published work as well as an additional study done on the two *brlA* cDNA clones.

A GeneMark examination of the W06E08 sequence suggested thirteen ORFs. Eight of the ORFs had homologs in the GenBank non-redundant database. The five others had coding potential as revealed by examination using the *A. nidulans*-specific GeneMark matrix *E.nidulans_mat3* (figure 20). Thus, the gene density for this cosmid was calculated to be approximately 1/ 2.9 Kbp.

Table 16. DNA sequencing summary for *A. nidulans* cosmid W06E08

<i>A. nidulans</i> strain:	FGSC4	Cosmid Vector:	pWE15
Chromosome source:	VIII	Subcloning Vector:	pUC18
Total thermocycling reactions:			980
	Forward:		490
	Reverse :		490
Gel readings in database:			851
Gel readings in contiguous sequence:			842
	Forward:		475
	Reverse:		367
Unpassed gel readings: (pUC vector or partial pUC)			51
E. coli			9
Insert size for cosmid W06E08:			38878 bp

3.22 ORFs and Exon Identification

The following is an expanded discussion of the homologies found to each of the predicted open reading frames of W06E08 also presented in Figure 20 and Table 17.

The *opa* ORF showed homology to a number of proteins with CAX repeats that encode glutamine residues. Because it was possible that this was a spurious match due to the presence of repeated DNA the glutamine residues in the translated sequence were masked and the sequence again was analyzed by a Blast search. However, a similar set of BLAST matches was found that included the mouse *mopa* gene as the best match homolog. Gerber et al. (Gerber, 1994) found that over 80% of the genes encoding a (CAX)_n repeat were transcription factors and that the glutamine repeats reside within the trans-activating domain. This (CAX)_n motif, also called an *opa* repeat, was first identified in homeotic genes of *Drosophila* (Schneuwly, 1986) and subsequently have been found in developmentally regulated proteins such as notch, engrailed and bithorax (Wharton, 1985). The *A. nidulans* homolog contains one 16 residue glutamine repeat as well as a 19 glutamine interrupted repeat and a total of 137 glutamine residues in the 483 amino acid polypeptide. This glutamine signature suggests that the *opa* ORF most likely encodes a polypeptide involved in transcription activation, and is related to genes primarily known in metazoans.

The *pot* ORF was flanked by 47 bp perfect inverted repeats and showed homology to three fungal transposases of the mariner class first identified in *Drosophila* (Farman, 1966). This is the first time the presence of a transposon-like element has been detected in *A. nidulans* and a recent finding suggests that additional copies are present in the genome (Li, M. G., Nicosia, D., Scazzocchio, C., personal communication) (see also discussion of cosmid sequence W02H02-W30B01 below).

The *brlA* ORF corresponds to bristle, a transcription factor that is necessary and sufficient for asexual development in *A. nidulans* (Adams, 1988; Prade, 1993) that had been previously reported (Prade, 1993). The gene region contained two overlapping Fig

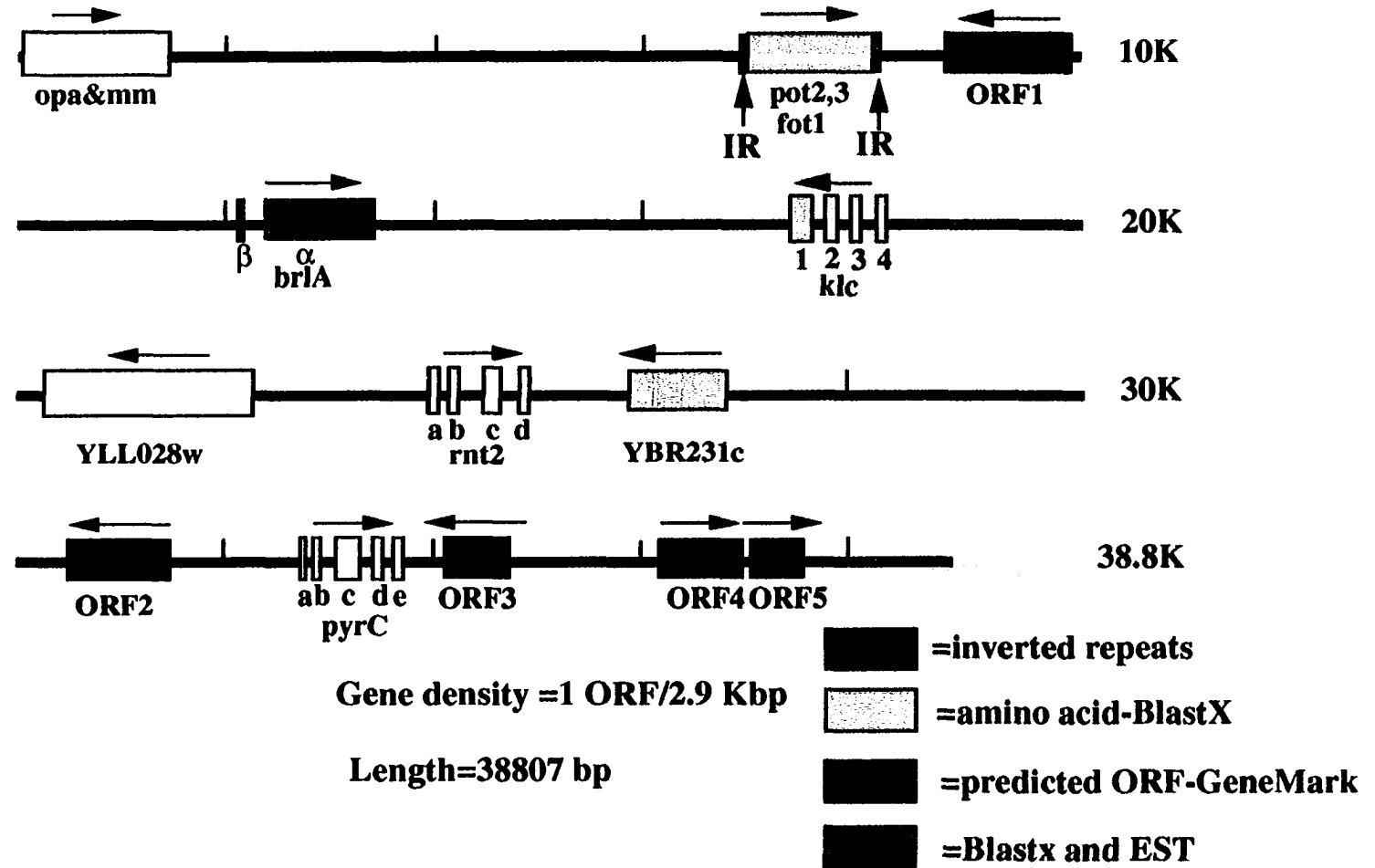


Figure 20. The ORF map of *Aspergillus nidulans* cosmid W06E08. Bars represent open reading frames or exons. Arrows indicate direction of transcription.

Table 18. Annotation of *A. nidulans* cosmid W06E08.

Significant Sequence Homology and Accession Number	% Similarity	Translation End Points	Size (a. a.)	Comments
<i>mopa</i> , differentially expressed Murine mRNA (M16362)	51	2>1453	483	diff. expressed in fetal and adult tissue possible transcription factor.
mastermind- <i>Drosophila viridis</i> (M92914)	47			
<i>pot3</i> , transposase, <i>M. griseq</i> (U60989)	62.7			<i>A.nidulans</i> inverted repeats (6575-6622 and 8397-8443)
<i>pot2</i> , putative transposase, <i>M. gridea</i> (Z33638)	49.3	6669>8335	555	
<i>pot1</i> , transposon, <i>Fusarium oxysporum</i> (X64799)	45			
<i>brlA</i> , regulator of conidiation, <i>A. nidulans</i> (L25858), cDNAs:c5h06, o4h01	100	*a-11176>11302 b-12315>13615	432	Transcription factor.
<i>klc</i> , kinesin light chain, <i>Loligo paelii</i> (squid) (L24440)	49.2	*1-17640<17950	103	4 ORFs with homology to <i>KLC</i> , coiled-coil and imperfect repeat region. Potential pseudogenes.
	44.3	2-17296<17702	136	
	43.5	3-16946<17308	124	
	45.7	4-16712<16926	71	
YLL028w, <i>S. cerevisiae</i> (Z73133)	64.7	20577<22107	510	ORF of unknown function Cycloheximide resistant protein
<i>cyhr</i> , <i>Candida maltosa</i> (cyhr_canma)	55.8			
<i>rnt2</i> , ribonuclease T2, <i>A. oryzae</i> (P10281)	73.4	a-24269>24520 b-24571>24780 c-24815>25135 d-25203>25255	277	fungal ribonuclease of T2/RH/M family, conserved cys residues.
YBR231c, <i>S. cerevisiae</i> (S46107)	51.1	25850<27008	386	ORF of unknown function.

Table 18R.continued

<i>pyrC</i> , dihydroorotase, <i>Ustilago maydis</i> (X63181) 68.2	a-32883>33167 b-32994>33087 c-33178>33633 d-33685>34163	450	third step in pyrimidine biosynthesis.
ORF1-No significant homology	9163<9826	220	
ORF2-No significant homology	36479>37069	197	
ORF3-No significant homology	37090>37737	216	
ORF4-No significant homology	34206>34878	223	
ORF5-No significant homology	31062<32134	356	

*-letters indicate exons and numbers indicate pseudogenes.

transcription units, α and β , one intron, and one ORF of unknown function (mORF) (Prade, 1993). The two transcription units result in two forms of the protein, the α form, which predominates during vegetative growth and the β form, which is the predominant species during asexual development. The β form differs from the α form by the addition of 13 amino acids at the 5' end.

A BlastX examination of this region with the EST Unigene database showed two EST clones, c5h06 and o4h01 which had homology to the *briA* gene. Each clone therefore was resequenced from both the 3' and 5' ends, and the sequences were assembled using Phrap to improve the accuracy of the cDNA sequences. Although neither clone contained the full length cDNA and thus the amino acid alignment was incomplete at the 5' end, the sequences that were present matched the genomic sequence with 100% identity. Because of the incomplete 5' end, it could not be determined whether the α or the β form was represented by these EST clones.

The *klc* region of this cosmid was a complex site consisting of four ORFs each with homology to a coiled-coiled motif and four 42-residue imperfect tandem repeats which are present in all kinesin light chain proteins (Wedaman, 1993). Kinesin light chains associate with heavy chains and, when coupled with ATP hydrolysis, move vesicles and organelles unidirectionally along microtubules. Since two of the *klc* ORFs overlap in different reading frames (ORFs 3 and 4 in Table 18) and a third region (ORF2) contains a stop codon, these three ORFs most likely are pseudogenes. ORF1 remains a potential gene coding region for kinesin light chain however, no ESTs had homology with this region to support this interpretation.

The *drt* region had its highest BlastX homology with a *S. cerevisiae* ORF, YLL028w, that encoded a product of unknown function. However, significant HSP scores also were found for a number of integral membrane drug resistance translocase genes, including *Candida maltosa* shown in Table 18. The highest alignment scores were with transmembrane proteins, where eleven of the twelve found in the *C. maltosa*

cycloheximide resistant protein (Sasnauskas, 1992) are present in the drt ORF. The hydrophobic residues characteristic of a transmembrane region are conserved in the drt ORF, suggesting that it most likely is an integral membrane protein belonging to the major facilitator drug resistance translocase family.

The mt2 region encoded a polypeptide with significant amino acid sequence similarity and conservation of cysteine residues involved in disulfide bonds characteristic of the *A. oryzae* ribonuclease rnt. This ribonuclease belongs to the fungal T2/H/M RNase family which preferentially cleaves 3' to A residues (Kawata, 1988).

The next ORF had homology to YBR231c, an *S. cerevisiae* ORF with unknown function. Since no other homolog was observed in the GenBank, no potential function could be assigned to the protein encoded by this ORF.

The adjacent region had homology to the *Usilago maydis* pyrC gene which produces dihydroorotase, the enzyme catalyzing the third step in pyrimidine biosynthesis, i.e. the closure of the pyrimidine ring in a dehydration reaction. The location of this gene downstream of the brlA gene corrected the relative positions of these two genes on the *A. nidulans* genetic map (Clutterbuck, 1997).

Finally, none of the five remaining ORFs that could encode proteins longer than 190 amino acid residues, had any significant homology to any entries in the GenBank databases.

3.23 Cosmids W02H02 and W30B01 sequence analysis

Cosmid W02H02 and W30B01 are overlapping cosmids from Chromosome VIII of *A. nidulans* that were of interest because hybridization studies revealed that they most likely contained genes involved in spermidine biosynthesis (N. Keller, personal communication). These two cosmids were from the minimum tiled cosmid library created by Prade et al. (Prade, 1997) and mapped distal to brlA on the same arm of Chromosome VIII.

The polyamine, spermidine, is involved in an early step of the sterigmatocystin biosynthetic pathway. This pathway has been partially characterized and the region of Chromosome IV containing this gene cluster has been sequenced (Brown, 1996 and section 3.26). The related species *A. parasiticus* and *A. flavus* are responsible for enormous grain crop losses each year because they produce the related toxin, aflatoxin (Lee, 1992) in infected harvested grain. The region of *A. nidulans* Chromosome VIII containing the spermidine synthase gene, which catalyzes the fifth and last step in spermidine production from arginine and methionine was of interest for three reasons. First, several aflatoxin genes from both *A. parasiticus* and *A. flavus* have been studied and found to occur in a gene cluster similar to the *A. nidulans* sterigmatocystin cluster. It therefore was of interest to learn the precise position and sequence of any additional genes involved in producing the toxin. The second was to obtain the sequence of the encoded toxin genes for future mutational studies. The third was to determine the gene environment of the *A. nidulans* spermidine synthetase gene as a prelude to sequencing the similar region of *A. parasiticus* which recently began in the Roe laboratory.

These overlapping cosmids span 44,998 basepairs and subsequent analysis of the results of a BlastX search of the non-redundant GenBank databases and the Unigene database revealed the structural features discussed below.

Table 18 describes the number of reactions needed to complete the sequence of each of these cosmids to an error rate of fewer than one base in 10,000 as determined by Phrap version 98.

A slightly different approach was used to sequence these cosmids than that used for W06E08. The recent availability of inexpensive internal primers and programs for optimum primer selection (Materials and Methods) allowed a shift from shotgun (random) sequencing to a directed phase earlier in a sequencing project and more rapid completion of the project using fewer sequencing reactions. Table 18 lists those reactions containing the universal forward and reverse primers used in the shotgun phase of sequencing and

Table 18. DNA sequencing summary for *A. nidulans* cosmids W30B01 and W02H02.

<i>A. nidulans</i> strain:	FGSC4	Cosmid Vector:	pWE15
Chromosome source:	VIII	Subcloning Vector:	pUC18
		<u>W02H02</u>	<u>W30B01</u>
Total thermocycling reactions:		830	641
Forward:		384	288
Reverse :		384	288
Internal Primer:		52	65
Gel readings in database:		786	554
Gel readings in contiguous sequence:		605	470
Forward:		245	189
Reverse:		240	220
Internal primer:		52	61
Unpassed gel readings (pUC vector or partial pUC):		155	60
<i>E. coli</i> :		26	9
low quality:		0	15
Final quality- in errors/10,000 bases		0.66	0.21
Insert size for cosmid		40554 bp	37312 bp
Accession number		AC005299	AC004395

the number of primers used in directed sequencing. Comparing the total number of sequencing reactions needed to finish the cosmids shows that 22 reactions/Kbp were required to complete W06E08 as compared to 19 reactions/Kbp for W02H02 and 15 reactions/Kbp for W30B01. A directed approach was used after 4x96 random samples were cycle sequenced for W02H02 and 3x96 samples for W30B01. These results indicate that adding a directed phase to the sequencing approach earlier resulted in fewer reactions being needed to obtain the finished sequence. Since there was a higher percent of *E. coli* and vector contamination in the W02H02 subclones than in W30B01, the data suggested that a shift to directed cycling after the same number of random samples as performed for W30B01 would result in fewer reactions to reach a similar error rate for W02H02 and the original database would have fewer contaminating sequences.

The cosmid sequences were aligned and extensive overlap found. Since 7685 base pairs are unique to W02H02 and 4443 base pairs are unique to W30B01, there is an overlap of 32,870 base-pairs and a combined length of 44998 base-pairs of Chromosome VIII. There was a single difference between the two cosmids in the overlap region where the G residue inserted in clone W02H02 position 21459, was missing from W30B01 position 13774. An example of a subclone sequence from each cosmid is shown in the Consed (Gordon, 1998) view of each region in Figure 21 where the arrow indicates the G residue. Each position was sequenced from three separate subclones verifying that both sequences were correct and that these are valid sequence differences between the two cosmid clones.

3.24 ORFs and Exon Identification

Table 19 and Figure 22 give a summary of the annotated features in the 45 Kbp region defined by the overlapping cosmids W02H02 and W30B01. Both BlastX and GeneMark (Borodovsky, 1994) were used in a combined extrinsic and intrinsic examination to determine these genomic features (Material and Methods). The features

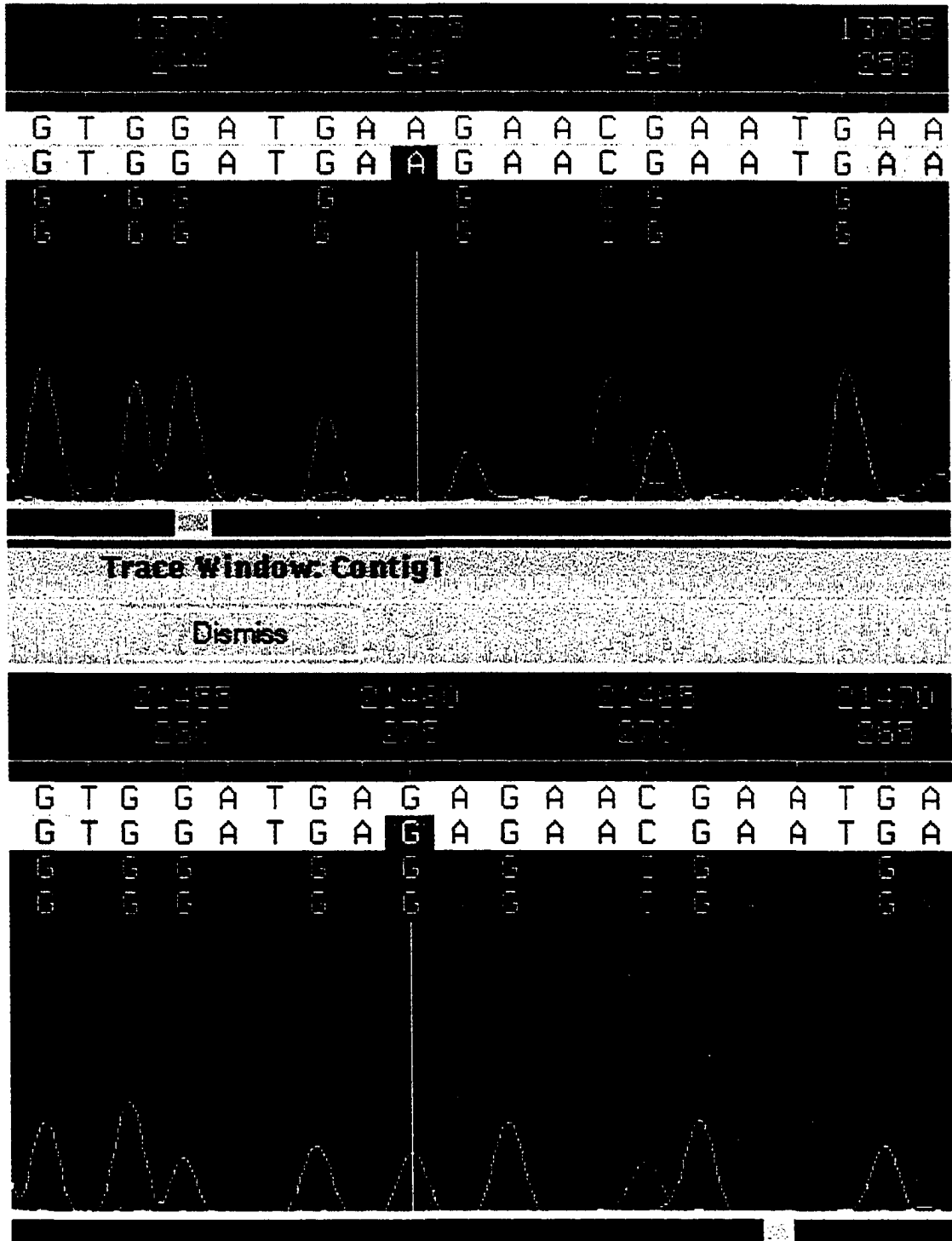


Figure 21. Comparison of two subclone sequences a2e12h2.f1 and a2a06b1.f1 from overlapping cosmids W30B01 (top panel) and W02H02 (bottom panel) showing an equivalent region in each. Cosmid W30B01 shows a deletion of a G residue between residues 13773 and 13774 (highlighted in red) which is present at position 26459 (highlighted in red) in Cosmid W02H02.

Table 19. Annotation of the 45 Kbp region of *Aspergillus nidulans* chromosome VIII defined by overlapping cosmids pW02H02 and pW30B01. Probability is given for each exon when multiples are present. EST homology is for each cDNA aligning with the given cosmid region.

Gene Homolog/Accession #	Probability Scores HSP p value	Translation endpoints	Size in amino acids	cDNA	Unigene sequence endpoints	EST % Homology
1. Transposase, tcl-like z29098 <i>D. hydei</i> , w02h02 only	68 4e-10 68 4e-10	2987>3901 3945>4366	446	e0b11a1	11631, 12103	365/376 97%
2. Glucoamylase precursor p08640 <i>S. cerevisiae</i> , w02h02 only	144 2e-6	5895>8549	885	-		351/351 100%
3. yLR063w s61636, <i>S. cerevisiae</i>	182 2e-19	10716>11069	118	j4c04a1	10734, 11103	351/351 100%
4. Prohibitin-like p40961, <i>S. cerevisiae</i>	591 5e-95 390 3e-36	12074<12397 11498<12013	280	y3b11a1	12397, 11749	338/338 100%
5. CytoC oxidoreductase subunit VII u20790, <i>N. crassa</i>	70 1e-10	13833>13905	24	-		-
6. Spermidine synthase q09741, <i>S. pombe</i>	ND 176 9e-16 112 4e-6 264 7e-23 302 6e-27	14120>14150 14310>14490 14576>14724 14832>15051 15326>15604	320	h4a05a1	14331, 15957	303/306 98%
7. Transketolase ql2630, <i>Kluyveromyces</i> <i>lactis</i>	1553 1e-159 1553 1e-159 1553 1e-159	18549<18602 18143<18285 18004<18088 15415<17934	934	y4a09a1 m0d06a1	18608, 15414	381/381 100% 381/381 100%

8.Acetate reg. DNA binding protein (FacB) u56097, <i>A. nidulans</i>	731 2e-72 413 1e-74 606 5e-59 606 5e-59 1986 3e-205	22655<23138 22282<22595 21943<22227 21537<21894 20324<21486	868	z4e02a1 20841, 20323	471/473 99%
9.spac3c7.01c z99568, <i>S. pombe</i>	none 123 4e-7 118 1e-6	26871<27161 26306<26776 25618<26229	458	-	
10.Thioredoxin-2 GTP binding protein u40843, p42942 <i>S. cerevisiae</i>	none 394 1e-36	29968<30060 29414<29632	40	-	
11.Expressed Gene 1	none none	30952>31065 31116>31409	136	d1d11a1 30882, 31485 i8e05a1 z1e12a1 m7b11a1 s3d05a1 z1a05a1 i3f04a1 c7g05a1	393/393 100% 361/362 99% 271/273 99% 524/525 99% 517/517 100% 468/469 99% 448/448 100% 369/369 100%
12.Expressed Gene 2	none	32285>33019	245	w7d08a1 32210, 32872 e7e03a1	182/184 98% 493/493 100%
13.High mobility group-like protein 2 p32495, <i>S. cerevisiae</i>	none 250 e-21	34052<34333 33532<33888	213	c5f04a1 34178, 33268	374/375 99%
14.Spindle assembly check-point MAD1 p40957, <i>S. cerevisiae</i>	281 5e-36	35602>37383	594	c5g07a1 35406, 37317	260/267 97%

15.yCR030 al021730, <i>S. cerevisiae</i>	164 2e-9	37755<38882	376	-
16.ORF1	none	39612<40355	248	-
17.ORF2 (w30b01 only)	none	42378<44555	726	-

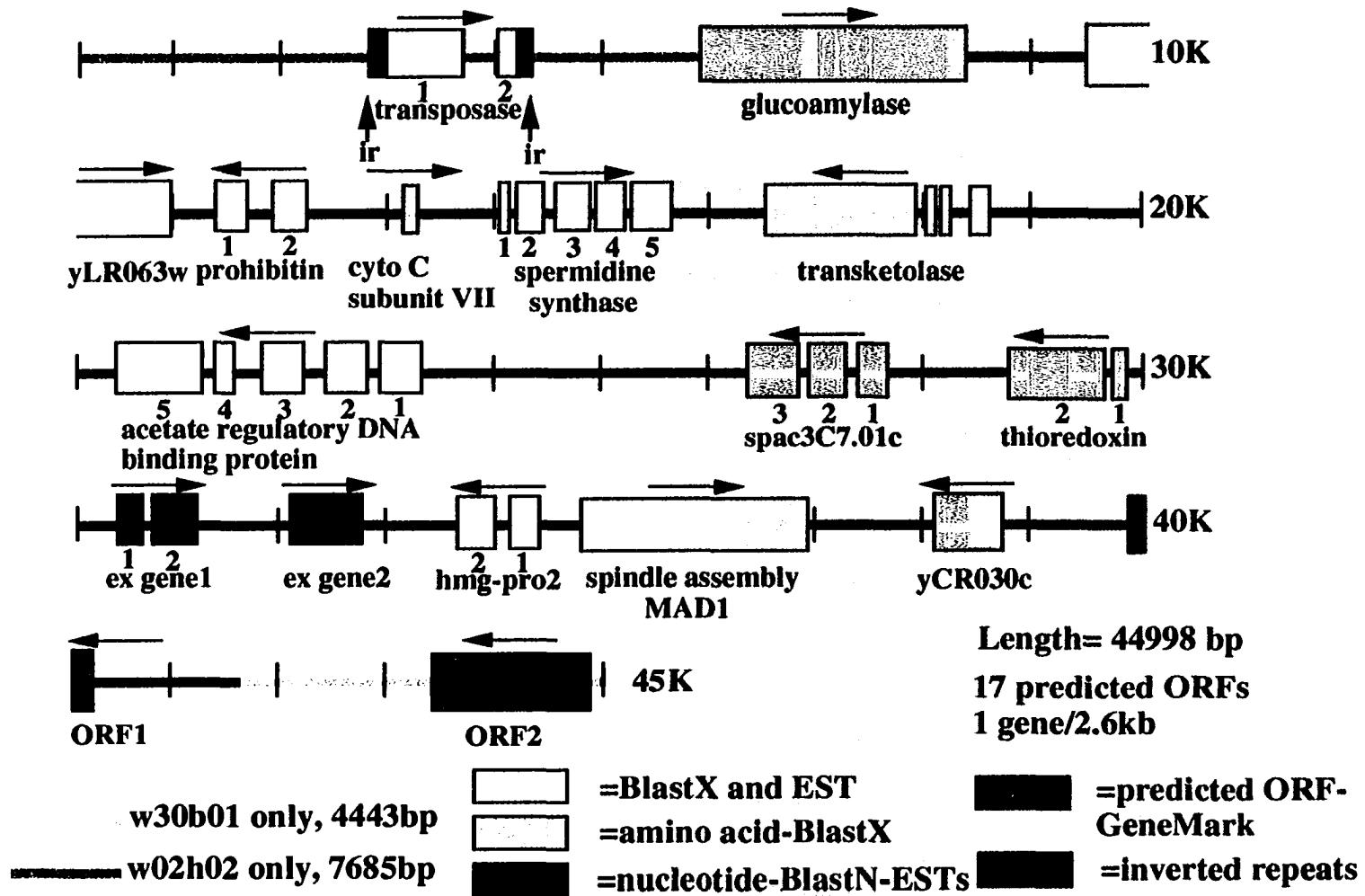


Figure22. ORF Map of *Aspergillus nidulans* Cosmids w30b01-w02h02. Bars indicated the cosmid sequence. Boxes indicate coding regions of genes and exons. Arrows indicate direction of transcription. Key defines the colors used for boxes and bars.

listed in Table 19 represent the BlastX homologies with the highest HSP and best probability scores. The importance of the EST database for genomic annotation was confirmed by a BlastN search showing a number of homologs from the *A. nidulans* EST database to the 45 Kbp W30B1-W02H02 region.

These matches are listed in Table 20 along with the percent of the individual EST homology. The corresponding Unigene member also was identified and, in all cases, additional sequencing of at least one of the clones was completed when the 3' and 5' sequences did not overlap each other in the Unigene database. Figure 22 displays the annotated region covered by the two overlapping cosmids with the predicted ORFs and their proposed gene assignment. The overlapping cosmid annotation is discussed below.

ORF1 had significant homology to a transposable element from *Drosophila hydei* which is a member of the Tc1-like transposons first characterized in *Caenorhabditis elegans* (Franz, 1992; Rosenzweig, 1983). The *D. hydei* transposable element, *Minos*, has perfect 255 nucleotide inverted repeats and two non-overlapping open reading frames which code for a 346 amino acid protein (Franz, 1992). Analysis of ORF1 revealed that perfect 244 nucleotide inverted repeats were present at the ends of two adjacent open reading frames with an ATG at the 5' end of the first, and the expected splice sites at the proposed intron/exon borders (Table 21). These two exons coded for a protein with 446 amino acids. The GCG Pileup alignment (Figure 23) of *minos* and the proposed transposon showed a 37% identity between the two amino acid sequences. There was conservation of uncharged residues, primarily in the 5' end of the *A. nidulans* protein which may be important for correct protein folding. This is the first report of a transposase of this class in *A. nidulans*.

A comparison of the tc-1 homolog nucleotide sequence with the Unigene database revealed a cDNA with high homology to the second exon. The 3' and 5' EST sequences of clone e0b11a1 overlapped to yield a single sequence whose GCG Bestfit alignment with the transposon is shown in figure 24. Mismatches between single pass EST

Percent Similarity: 36.986 Percent Identity: 30.822

| = IDENTITY
: = Similarity

```

1
Minos MSQYSMOKNF RLLQISRSLA TMVRGKPISK EIRVLIRDYF KSGKTLTEIS
Transpos ----- -MPRGGFHPV ELRVQVLTLA AIGFSTEKIS

51
Minos KQLNLPKSSV HGVIQIFKKN G..... ..NIENNIAN RGRTSAITPR
Transpos KSLNLSPRTV QSIVKGRDR GYRPEVSLRV QLEFVEDRKR SGRPVEITEA

101
Minos DKRQLAKIVK ADRRQSLRNL ASKWSQOLAK LSSESGRDKL KSIGYGFYKA
Transpos TQNTVITSVT ADR..AGREK LSEILAYEAG ISHSSVLCIL HSHGFVIAKP

151
Minos KEKPLLTLRQ KKKRLQWARE RMSWTQRQWD TII FSDEAKF DVSVDTRKR
Transpos SWKPGLTEAA CLRLEFCLA HQHWTLEDWK RVI FTDETGV ILGHRRGAIR

201
Minos VIRKRSEYH KDCLKRTKF PASTMVWGCM SAKGLGKLF IEGTVNAEKY
Transpos VWRTVKDSHT RNCVRRRWKA CSDFMVWGCF SYNKKGPLHI YKPETAAMRK

251
Minos INILQDSLIP SIPKLLDCGE FTFQODGASS HTAKRTKNWL QYNQMEVLDW
Transpos QADIEIEAMN RELEPLCREE WELATGLSRV HLRPNRGRVP KWNWNEKNGK

301
Minos PSNSPDLSPI ENIWWMKNQ LRNEPQRNIS DLKIKLQEMW DSI SQEHCNK
Transpos LIRKGGGID WWRYQTVCSL ISIIYYRLK PLLI PFAKEC .MIERPNTIV

351
Minos LLSSMPKRVK CVMQAKGDVT QF-----
Transpos LEDSAPAHCH RIQQHVYKAE DVQKILDWPG NSPDLNAIEP CWAWMKRRTT

401
Minos -----
Transpos SRGAPRDKKT GEAEWRQAWA DLPQETIQHW IERLICHIQI VIELEGGNEY

451
Minos -----
Transpos KEGREDRDTR SWAGRRIKG

```

Figure 23. The Pileup alignment of the *A. nidulans* Tc-1 element and the *Drosophila* *Minos* transposon. Highlighted bases are conserved uncharged residues.

Length: 140
 Gaps: 0
 Percent Similarity: 92.143 Percent Identity: 90.714

| = IDENTITY
 : = 2
 . = 1

Transposon.P x E0b11fr.P

```

      .           .           .           .
306 LKPLLIPFAKECMIERPNTIVLEDSAPAHCHRIQQHVYKAEDVQKILDWP 355
      |||||  |||||  |||||  |||||  |||||  |||||  |||||  |||||  |||||  |||||
      2 LKPLLILFAKECMIERLNTIILED SAPAHCHQIQQHIYKAEDMQKILDWP 51

      .           .           .           .
356 GNSPDLNAIEPCWAWMKKRTTSRGAPRDKKTGEAEWRQAWADLPQETIQH 405
      ||  |||||  .  |||||  ||  ||  |||||  ||  |||||  |||||  |||||  |||||
      52 GNLPDLNAIKPCWAWMKKHTISRSAPRDKKTGEVECRQAWADLPQETIQH 101

      .           .           .           .
406 WIERLICHIQIVIELEGGNEYKEGREDRDTRSWAGRRIKG 445
      |||||  |||||  |||||  |||||  |||||  |||||  |||||  |||||  |||||
      102 WIERLICHIQIVIELEGGNEYKEGREDRDTRSWAGRRIKG 141
  
```

Figure 24. The Bestfit sequence alignment of the *Aspergillus nidulans* tc-1-like transposon and cDNA clone e0b11a1 translation product showing strong homology with the second exon of the genomic transposon with the overlapping EST sequences of e0b11a1.

sequences and a genomic sequence are not unexpected. As can be seen in Figure 24, the two sequences have significant homology, this genomic region most likely encodes an expressed tc-1-like transposase.

This result is of great interest since there has been no evidence given in the literature for an active transposon in *A. nidulans*. An alignment with the fot1 class transposon discovered in cosmid W06E08 showed no sequence homology to this tc-1 class *A. nidulans* transposon confirming that these are representatives of two distinct classes of transposons, and that the tc-1 homolog appears to be actively transcribed. Interestingly, although Fot1 homologs has been found in related ascomycetes (Nyyssonen, 1996), homologs for the second, tc-1, have been detected only in higher eucaryotes (Merriman, 1995)

ORF2 had a significant homology with a glucoamylase of *S. cerevisiae*. This is a secreted enzyme which attacks both α -1,4 and α -1,6 linkages of starch to yield glucose and is used commercially to produce high glucose syrup (Carlile, 1994). The alignment of the *A. nidulans* and *S. cerevisiae* glucoamylase proteins shows a conservation of the serine/threonine residues in the region from 210-572 of the yeast glucoamylase and an overall sequence similarity of 31% (Figure 25). Thus, this open reading frame encodes a possible glucoamylase that had no corresponding ESTs in the *A. nidulans* Unigene database. This was not unexpected since, at the time of mRNA harvest for the library, glucose was the sole carbon source and glucoamylase is generally an inducible enzyme.

ORF3 showed homology to yLR063w, a *S. cerevisiae* open reading frame with unknown function. ORF3 also had significant homology to the EST clone j4c04a1. Eighteen bases at the 5' end of the EST clone are missing but the otherwise perfect nucleotide match indicates no introns in ORF3 nucleotide sequence and verifies this region as containing an actively transcribed gene.

ORF4 has significant homology to an *S. cerevisiae* prohibitin, a protein with anti-proliferative activity (Berger, 1998) that is important in determining the length of the cell

Identities = 72/372 (19%), Positives = 121/372 (32%), Frame = +3

cosmid: 5754 GGEVTLVWRS SDVRSLEQLKTTSESSTSTSSSTSVSSTTTSSSTSQT*RMASISLPLQPOTA 5933
GG + SS S +TS SST+TSSTSSSTTTSSSTSS++ +S + P P T

S. cere: 206 GGTKSSTTTSESSTTTSTSESSTTTSSSTSSSTTTSSSTSSSTSSSTTAPATPTT 265

cosmid: 5934 -----PSFPFYFDGADPAPSGHRRRLTMSTPLPNPFFVFPARDEDEPKQDLDTTNGR 6095
P+PP P+ + T T K T +

S. cere: 266 SCTKEKPTPPTTCTKEKPTFPFHDTTPTCK-----KKITTSKTCTK 309

cosmid: 6096 PPLPAFSFNPGSVGSNQAPAPAPSNBRM-SGHRROYSEFVGGDQLIIPGNTAAGQTSDET 6272
P + + + S+ AP P PS+S S S +P +++ S

S. cere: 310 TTTFVPTPSSSTTESSSAPVPTPSSSTTESSSAPVTSSTTESSSAPVPTPSSSTTESSSA 369

cosmid: 6273 PMASSSTTVSS SVFRWSSTTXS SQDLDTIS SVDLTAISNALDLKPYVESAPCTSDMTR 6452
P+ SSTT SSS SSTT SS T SS ++ S SAP TS+

S. cere: 370 PVTSSTTESSSAPVTSSTTESSSAPVPTPSSTTESSSAVTSSTTESSSAPVTSTTES 429

cosmid: 6453 ERRP----SLEPSQLLPHSATVLSRPTPPASPQLNLNEAS PSSQIPKNERLENPRCTPT 6620
P + E S S+T S P +P + E+S + + + TP+

S. cere: 430 SSAPVTSSSTTESSSAPVTSSTTESSSAPVPTPSSSTTESSSAPVTSSTTESSSAPVPTPS 489

cosmid: 6621 SFAPESQNAIRS SVLARKSDTAITSPKAESSAASQQPRPRPTADA-SLMLELGGSTMT 6797
S ES +A +S S + +P + ++ +S P P P ++ S ++ ST

S. cere: 490 SSTT-ESSAPVTSTTESSSAPVPTPSSSTTESSSAPAPTPSSSTTESSSAPVTSSTTE 548

cosmid: 6798 DNSSPTKRPNSAAGHSRSEKMS S 6869
+S+P P+S+ S S SS

S. cere: 549 SSAPVPTPSSSTTESSSSTPVTSS 572

Figure 25. Comparison of the *A. nidulans* glucoamylase homolog with *S. cerevisiae* AC# P08640 showing the conservation of threonine and serine residues (in bold) in a thr/ser signature region.

cycle (Coates, 1997). In yeast, the accumulation of the prohibitin protein has been associated with a shift to senescence, and in humans, prohibitin has been characterized as an antitumor factor (Coates, 1997). The *A. nidulans* protein encoded by ORF4 has 70% identity with the yeast prohibitin protein, (GenBank Accession number p40961). EST clone y3b1 1a1 included the entire mRNA sequence. Aligning the consensus Unigene sequence with the genomic sequence defined an intron, and matched with 100% accuracy the first exon and 98% the second. A 280 amino acid protein which was similar in size to the 284 amino acid yeast protein was predicted.

The region from 13834-13905 defined a small ORF which GenMark indicated as having coding potential. There was a single match in the GenBank databases which had low homology to the *N. crassa* subunit VII of cytochrome C oxidoreductase (Lobo-Hajdu, 1996). There were no Unigene sequence homologies to verify that this region was expressed, so no additional conclusions could be drawn.

Since both cosmids previously had hybridized to a spermidine synthase probe (Nancy Keller, personal communication) it was expected that the region coding for this gene would be detected. Therefore, it was not surprising when ORF6 had homology with an *Schizosaccharomyces pombe* spermidine synthase in GenBank. The EST clone, h4a05a1, also aligned with this region and the sequence revealed four of the five predicted introns. However, this EST clone did not include the extreme 5' end where the first small exon is located. GeneMark did not predict this exon and it only was determined based on the alignment of the translated products of the *S. cerevisiae* and *S. pombe* spermidine synthases. Figure 26 shows that this 10 amino acid-encoding exon contains the methionine start codon and had a sequence that was conserved between both the spermidine synthase of *S. cerevisiae* (accession number Q09741) and that of *S. pombe* (accession number Q12074).

ORF7 had a very significant level of BlastX homology to the 5' region of the transketolase of the single-celled ascomycete, *Kluyveromyces*. This enzyme catalyzes

```

1           |2
A. nidulans MS--EITHPTIKDGFSEQSE-MWPGQAMNLRVNQILHHEKSKYQDVLVFESSDYGTVLV
S. cerevisiae M-AQEITHPTIKDGFSEISDTMWPGQAMTLKVEKVLHHEKSKYQDVLIFKSTTYGNVLV
S. pombe MSVQELSHPLIKDGFREINN-MWPGQAMTLKVKKVLVYAGKSKYQDVLVFESETYGHVLV
* * ** ***** ** ***** * ***** ** ** **

          |3
A. nidulans LDNVIQCTERDEFSYQEMITHLAMNSHPNPKKVLVIGGGDGGVLRVVKHETVEEAILCD
S. cerevisiae LDNVIQCTERDEFAYQEMIAHLALNSHPNPKKVLVIGGGDGGVLRVVKHESVEEAWLCD
S. pombe LDGGAQCTERDEFSYQEMIAHLALNSHPNPKKVLVIGGGDGGVLRVVKHECVEEAILCD
** ***** ***** ** ***** ***** ***** ***** ***** **

          |4
A. nidulans IDEAVIRVSKKYLPGMSIGFQHPNVKVHVGDGFELKQRQNEFDVIITDSSDPEGPAESL
S. cerevisiae IDEAVIRLSKEYLPGMAASYSHPKVKVHVGDGFQFLRDYQNTFDVIITDSSDPEGPAETL
S. pombe IDEDVIKVKQYLPGMSAGFNHPNVKVIIGDGFKFLQDYQNTFDVIITDSSDPDGPAAEL
*** ** ** ***** ** ***** ** ** ***** ***** **

          |5
A. nidulans FQKPYFELLRDALRDGGVITTTQAENQWLHLPLIADLKKACNEVFPVAEYAYTTIPTYPSG
S. cerevisiae FQKEYFQLLNSALTEKGVITTTQAESMWLHLPLIKDLKKACSEVFPVAEFVYTTIPTYPTG
S. pombe FQKPYFELLSDALRGGVITTTQAECMWLHLGVISNVLTAVKTVFPVVEYAYTTIPTYPSG
*** ** ** ** ***** ***** * . * ***** * ***** *

A. nidulans QIGFMVCCKDANRNVKEPVRTWSREEEERLCRYYNQDIHRASFVLPNPFARKALGN----
S. cerevisiae TIGFMVCSKDKTCNVKKPLREISDEKEAELYRYYNKKIHEASFVLPPTWAAKELN----
S. pombe SIGFVVACKDASIDLKEPLRKWSPEEENKLCRYYNSEIHAASFVLPPTFARDVVDKATSS
*** * ** * * * * * * * * * * * * * * * *

```

Figure 26. GCG Pileup alignment of the spermidine synthase protein products from three ascomycetes showing 193/240 or 80% conserved amino acids. The numbers and bars (|) indicate the five exons in *A. nidulans*. Exon 1 was determined by the alignment of the three orthologs of spermidine synthase. (*) indicate conserved amino acids.

the step of the pentose phosphate pathway that links this pathway to glycolysis and provides four carbon sugars. Two cDNA clones, y4a09a1 and m0d06a1, aligned with this cosmid region. The h4a05a1 ESTs also aligned with the ESTs from these two clones in the Unigene database. However, it was apparent that this was not an alignment of clones from the same gene but rather an alignment of clones from overlapping convergent transcription units because the 3' sequences overlapped but in the incorrect orientation and the 5' EST sequences did not align (Figure 27). Further examination revealed that there was a 23 basepair overlap between the two regions in the 3' untranslated regions of both genes (Table 19).

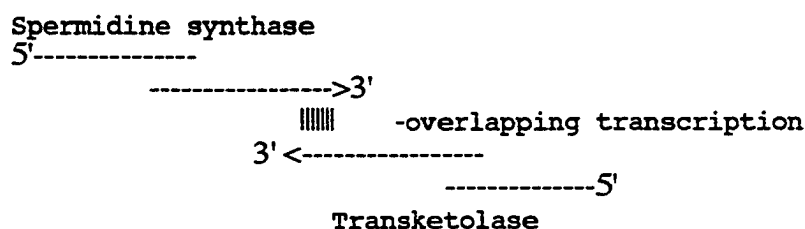


Figure 27. Alignment of the ESTs from clones y4a09a1 and m0d06a1 for transketolase and h4a05a1 from spermidine synthesis showing overlapping convergent transcription units.

Alignment of the Unigene consensus sequence containing the y4a09a1 and m0d06a1 ESTs with the *A. nidulans* cosmid sequences revealed three introns, three small exons, and one large exon (Table 19). The sequence alignment with the *Kluyveromyces* transketolase had 61% identity (Figure 28) but only in the region of the large 3' exon from cosmid nucleotide position 17,500 to 16,121. This region when analyzed with Blocks (Henikoff, 1991), revealed homology to three of six transketolase specific domains (block BL00801d-f). Since the remaining three domains were contained in the 5' end of the *Kluyveromyces* transketolase and the 5' region of the *A. nidulans* gene showed no homology to any GenBank entries and has no Blocks matches, it appeared


```

sp|Q12630|TKT1_KLULA TRANSKETOLASE (TK) >gi|1488336 (U65983)
transketolase [Kluyveromyces lactis] Length = 679

Score = 570 bits (1453), Expect = e-162
Identities = 281/460 (61%), Positives = 353/460 (76%), Gaps = 2/460
Frame = -1
Query=2h2-30b1 subject=transketolase

cosmid: 17500 GDNDLEGIRAAIKQAQAVKDKPSMIRLTTTIGFGSKLQGTGGVHGPNLKADDIEGVKKRF 17321
      G++DL+ I A+++A+ + D+P+++I+LTTTIGFGS G+ VHG PLKADD++ +K +F
Kluyv: 220 GNDLDAISKALEQAK-LSDRPTLIKLTMTTIGFGSLNAGSHSVHGAPLKADDVVKLVKVF 278

cosmid: 161320 GFDPAQSFFVVPQQVYDLYHKHA-EEGAAQEQEWNQLLQKYAGEYPNEHADLTRRLSGKLP 17144
      GF+P +SFVVPQ+VYDLY+K E G ++W+ LL Y G++P A++ RRL+G+ P
Kluyv: 279 GFNPKESFVVPQEVYDLYNKSTIEPGIEANKQWDALLDAYVGOFPPELGAEVKRRRLAGEFP 338

cosmid: 161143 EGWEKSLPVYKPSDPAIASRKLSEAVLEKIHVSVIPELLSGSADLTGSNNTRWKNVDFQP 16964
      EGWE LP Y P D A+ASRKLSE VL+ + +PELL GSADLT SN TR K AVDFQP
Kluyv: 339 EGWESKLPYTPEDSAVASRKLSEIVLDNVFDTLPELLGGSADLTSPNLTSTRKGAVDVDFQP 398

cosmid: 16963 PEYGIGEWSGRYLRYGVREHAMAAIMNGLAAYGTVI-PAAGTFLNFVSYAAGAVRLSALS 16787
      P G+G++SGRY+RYGVREH M AIMNG++A+G P GTFLNFVSYA+GAVRLSALS
Kluyv: 399 PITGLGDYSGRYIRYGVREHGMGAIMNGISAFGANYPYGGTFLNFVSYASGAVRLSALS 458

cosmid: 16786 RVRAIHVATHDSIGLGEDGPTHQPIETLAHFRALPNCMVWRPADGNETSAAYYSALTAKH 16607
      I VATHDSIGLGEDGPTHQPIETLAHFRA+PN VWRPADGNE +AAY ALT KH
Kluyv: 459 GHPVIWVATHDSIGLGEDGPTHQPIETLAHFRAIPNLQVWRPADGNEVTAAYKVALTNKH 518

cosmid: 16606 TPSILALTRQNLPOLENSIEAALKGAYPVFEAADAKVTIISTGSEVSI CIDAAYLQEK 16427
      TP+I+AL+RQNLPOLE+ SS+E A+KG Y + + + I+STGSEV I ++AAK L EK
Kluyv: 519 TPAIALSRQNLPOLENSIEAALKGAYPVFEAADAKVTIISTGSEVSI CIDAAYLQEK 578

cosmid: 16426 HGVVARVVSIPCFEVFDAQDKEYRLKVLDPDGIPILSVEVCSTMGWERYSHQFGLNRFGA 16247
      + + AR+VS+P F F Q KEY+L V PDG+PILSVEV +T GW +Y+H+ FGL+RFGA
Kluyv: 579 N-IKARIVSLPDFHSGQSQKEYQLSVFPDGVPIILSVEVLATSGWVSKYAHQSFGLDRFGA 637

cosmid: 16246 SGPYKEVYAKFEFTPEGISKRALATIDFYKGVFVRSPINRAF 16121
      SG VY KFEFTP+GI+ RA T++FYKG V SP+N AF
Kluyv: 638 SGKGPVYKFEFTPEGIATRAEKTVEFYKGVFVRSPINRAF 679

```

Figure 28. BlastX alignment of W30B01-W02H02 ORF7 with *Kluyveromyces* transketolase showing alignment only of the 3' end of the proteins beginning at amino acid 220 of this yeast homolog.

likely that the *A. nidulans* ORF7 did not code for a homolog of this transketolase but rather coded for a gene with a related function or another transketolase with a unique 5' domain.

The region designated ORF8 had significant homology to the *A. nidulans* acetate regulatory DNA binding protein (*facB*). *FacB* is a transcriptional activator involved in acetamide and acetate utilization (Todd, 1997). The nucleotide sequences of the cosmid ORF8 differed only slightly from the GenBank entry, showing two base substitutions, one deletion and three insertions, all in intronic regions. This may be due to strain differences as the *A. nidulans* strain which was the source of the U56097 sequence was not reported. Interestingly, this gene included the single base difference between cosmid W02H02 and W30B01 that was discussed above and which occurs in the 3' most exon of *facB*. The W02H02 cosmid sequence matches the GenBank entry at position 21,460. The lack of the G in cosmid W30B01 results in a frameshift within the last exon that affects a coding region for *facB* and suggests that the difference between the two cosmid sequences may have occurred during the growth and maintenance of the cosmids under conditions where there was no selective pressure to maintain this gene. Even so, this difference is curious, especially since both cosmids were constructed from the same *A. nidulans* strain and by the same researcher (Prade, 1997).

A single *facB* cDNA clone, z4e02a1, was present in the Unigene database which covered only the 3' most exon but did not include the deletion site. It was surprising to find a clone for *facB* in the cDNA library since the culture producing the library was grown on glucose as the sole carbon source. However, it is known that fungi often ferment sugars to ethanol, glycerol, acetate, butanediol and additional products (Carlile, 1994). Since the Unigene database contains a butanediol dehydrogenase, Contig 1693, involved in butanediol production from glucose, it may be that *A. nidulans* fermented at least part of the available glucose and thus this gene is expressed because its product is required for utilization of acetate.

ORF 9 had homology to a hypothetical *S. pombe* protein, spac3c7.01c. There appeared to be three exons in this *A. nidulans* gene that would encode for a protein with 458 amino acids. The two 3' most exons had homology with the spac3c7.01c ORF. Since no ESTs aligned with this ORF, additional studies would be needed to verify if this hypothetical gene is expressed.

ORF10 encodes a region with homology to *S. cerevisiae* thioredoxin-2, a reducing power carrier, which is involved in control of the cell cycle S phase (Muller, 1991). No ESTs were observed for this region but the alignment with the *S. cerevisiae* thioredoxin-2 homolog predicts two exons that would encode a 104 amino acid protein, which is the same size as that predicted for the *S. cerevisiae* gene.

ORF 11 was defined by eight cDNA clones and was matched with 100% identity by their Unigene consensus sequence. Since the Unigene consensus defined two exons and an apparent 5' end, a protein of 136 amino acids was predicted (Figure 29). In addition, GeneMark defined both exons with "good" coding potential. Although there are no homologs to this gene in the GenBank databases, analysis of the eight homologous cDNAs revealed that the entire coding region and the intron were defined by the Unigene consensus sequence aligning with this region that included an intron with consensus borders (Table 21). Even though this gene was a member of the abundantly expressed class of genes in the *A. nidulans* Unigene database there were no known homologs in the public databases and thus this represented a new gene encoding a 136 amino acid polypeptide with unknown function.

ORF 12 is similar to ORF11 in that GeneMark indicated a region with coding potential that aligned with two cDNA clones. Again, there were no homologs found by a BlastX search. The ORF12 region had a single open reading frame but only the 5' end was present in the Unigene consensus sequence. Thus, this represents a new gene that encodes a protein with 245 predicted amino acids defined by the alignment of the cDNA clones.

Query file(s): Contig2h2-30b1.fasta (ORF11)

Subject file(s): Unigene Contig19.fasta

minmatch: 14, minscore: 30

Exon 1

```
CONTIG30B1 23197 GTACCAACAAACCATTAACTAACACCACAACCTCTATCTCTGCATTCAAC 23246
Contig19    1 GTACCAACAAACCATTAACTAACACCACAACCTCTATCTCTGCATTCAAC 50
                    5'-> M S D S T F H T T I
CONTIG30B1 23247 GAAACATATCTCTATCACAATGTCTGACAGCACCTTCCACACCACCATTTC 23296
Contig19    51 GAAACATATCTCTATCACAATGTCTGACAGCACCTTCCACACCACCATTTC 100
                    Q D I R K P E S H A S H A A K G N
CONTIG30B1 23297 AGGACATTTCGCAAGCCAGAGTCTCAGCCTTCCCATGCTGCTAAGGGCAAC 23346
Contig19    101 AGGACATTTCGCAAGCCAGAGTCTCAGCCTTCCCATGCTGCTAAGGGCAAC 150
                    T P K D S N V S A M K
CONTIG30B1 23347 ACTCCTAAGGATTCTAATGTCTCCGCAATGAAG 23379
Contig19    151 ACTCCTAAGGATTCTAATGTCTCCGCAATGAAG 183
```

Exon 2

```
                    S I I D Q N T D K Q A D I E K T
CONTIG30B1 23430 TCCATTATCGACCAGAACACAGACAAGCAAGCCGACATCGAAAAGACC 23477
Contig19    184 TCCATTATCGACCAGAACACAGACAAGCAAGCCGACATCGAAAAGACC 231
                    K A N L P L P D Q P P V A S D W N
CONTIG30B1 23478 AAGGCCAACCTGCCATTACCAGACCAGCCCCCTGTGCTAGTGACTGGAA 23527
Contig19    232 AAGGCCAACCTGCCATTACCAGACCAGCCCCCTGTGCTAGTGACTGGAA 281
                    S A D Q R A V N V G S G G I E G
CONTIG30B1 23528 TTCCGCCGATCAGCGAGCTGTCAATGTTGGTTCCGGCGGTATCGAAGGAC 23577
Contig19    282 TTCCGCCGATCAGCGAGCTGTCAATGTTGGTTCCGGCGGTATCGAAGGAC 331
                    P I S G E N N S A L R G P A T A S
CONTIG30B1 23578 CCATCTCAGGCGAGAACAACCTCTGCTCTCCGAGGTCCAGCCACAGCCTCA 23627
Contig19    332 CCATCTCAGGCGAGAACAACCTCTGCTCTCCGAGGTCCAGCCACAGCCTCA 381
                    S S A R E V G E E T H R N T Q P T
CONTIG30B1 23628 AGCAGTGCTCGCGAGGTTCGGAGAGGAGACGCACAGGAACACACAGCCAAC 23677
Contig19    382 AGCAGTGCTCGCGAGGTTCGGAGAGGAGACGCACAGGAACACACAGCCAAC 431
                    S N V G R G D L P A D A Q A R ->3'
CONTIG30B1 23678 TAGCAATGTTGGTCGGGGAGACCTCCCTGCCGATGCTCAGGCTCGGTAAC 23727
Contig19    432 TAGCAATGTTGGTCGGGGAGACCTCCCTGCCGATGCTCAGGCTCGGTAAC 481
                    S I I D Q N T D K Q A D I E K T
CONTIG30B1 23728 CCATGATTCTCATTGTTTGCAGCATAGCGATGTGATACGAACAAAACGAA 23777
Contig19    482 CCATGATTCTCATTGTTTGCAGCATAGCGATGTGATACGAACAAAACGAA 531
                    S I I D Q N T D K Q A D I E K T
CONTIG30B1 23778 GACATAATGATGATTTCTCCATG 23800
Contig19    532 GACATAATGATGATTTCTCCATG 554
```

Figure 29. Crossmatch comparison of the Unigene sequence Contig19 and a region of composite sequence of cosmids W30B01 and W02H02 showing 100% identity. This defines a new gene containing one exon with no homologs in the GenBank databases. The proposed translation start and stop are highlighted.

ORF13 had homology to a cDNA clone, c5f04a1, with a significant BlastX similarity to high mobility group-like nuclear protein2 (Kolodrubet, 1991). Although the function of this protein is unknown, it is essential in *S. cerevisiae* and had 62% identity with the *A. nidulans* ORF13. Comparison of the cDNA and genomic sequences revealed two exons that would encode a 213 amino acid protein.

ORF 14 is a homolog of the *S. cerevisiae* spindle assembly checkpoint gene MAD1 (Hardwick, 1995). The ESTs from clone c5g07a1 aligned with the large 3' exon but not with the smaller 5' exon. The BlastX alignment of ORF 14 with the *S. cerevisiae* MAD1 revealed that the *A. nidulans* homolog did not have the asparagine rich domain found in the smaller exon of the yeast homolog. In addition, the *A. nidulans* 5' exon defined by GeneMark does not align with the yeast homolog. The *A. nidulans* exons encoded a 594 amino acid protein, smaller in size than the 749 amino acid yeast homolog. This result was similar to that observed with the transketolase gene homolog reported above since the 5' end of the gene either coded for a domain unique to the *A. nidulans* homolog or defined a gene with a different function which shared a domain with MAD1.

The last two ORFs did not have any significant GenBank homologs as revealed by a BlastX search, had no homologs in the Unigene database, and had coding potential suggested only by GeneMark. Thus it was unclear whether these ORFs actually encode their predicted proteins.

3.25 Cosmid W02H02-W30B01 Conclusion.

Seventeen ORFs were assigned to this 45 Kbp region of *A. nidulans* chromosome VIII by a combination of GeneMark, BlastX homology to the GenBank databases and by homology to the *A. nidulans* EST Unigene database. GeneMark ORF analysis was for all six frames, using a coding potential matrix specifically designed for *A. nidulans* based on codon usage. A BlastX search of the nonredundant protein database with the entire 45 Kbp region was examined and a BlastN search of the *A. nidulans* entries in the dbEST

also was performed. The results from GeneMark and BlastX gave support for the presence of a gene but neither was sufficient to allow more than a predicted, putative gene assignment to a region. However, the alignment of one or more ESTs could confirm that a region contained a transcribed gene. Based on these assumptions, the 45 Kbp W02H02-W30B01 region had 10 of the 17 genes predicted by GeneMark and/or BlastX verified as expressed genes by homology with one or more ESTs.

The difference between the number of ESTs which align with this region and to the SW06E08 cosmid region where only one in 13 predicted genes was aligned with an EST, is striking and it presumably was fortuitous that the W02H02-W30B01 region contained genes which were transcriptionally active at the time of library mRNA harvest.

3.26 The Sterigmatocystin Gene Cluster

Accession number U34740 represented a 60 Kbp region of Chromosome IV containing the 25 gene cluster for sterigmatocystin biosynthesis, a secondary metabolite produced by a complex pathway which includes a polyketide synthase (Figure 30). This fungal toxin is of extreme interest to agricultural scientists because of the enormous grain crop loss after infection by either *A. paraciticus* and *A. flavus* (Jimenez, 1991; Magnoli, 1998).

Polyketides, a structurally diverse class of secondary metabolites produced by fungi, bacteria and plants, are synthesized by polyketide synthase through a complex series of condensation reactions of small carboxylic acids (Katz, 1993). Sterigmatocystin and aflatoxin are related polyketide toxins produced by members of the *Aspergillus* family. These two toxins are among the most highly toxic and carcinogenic naturally produced products. *A. nidulans* does not produce aflatoxin because it lacks the last two enzymes in the pathway but does produce sterigmatocystin from which aflatoxin is derived. As Figure 30 shows, understanding of the pathway is incomplete and the

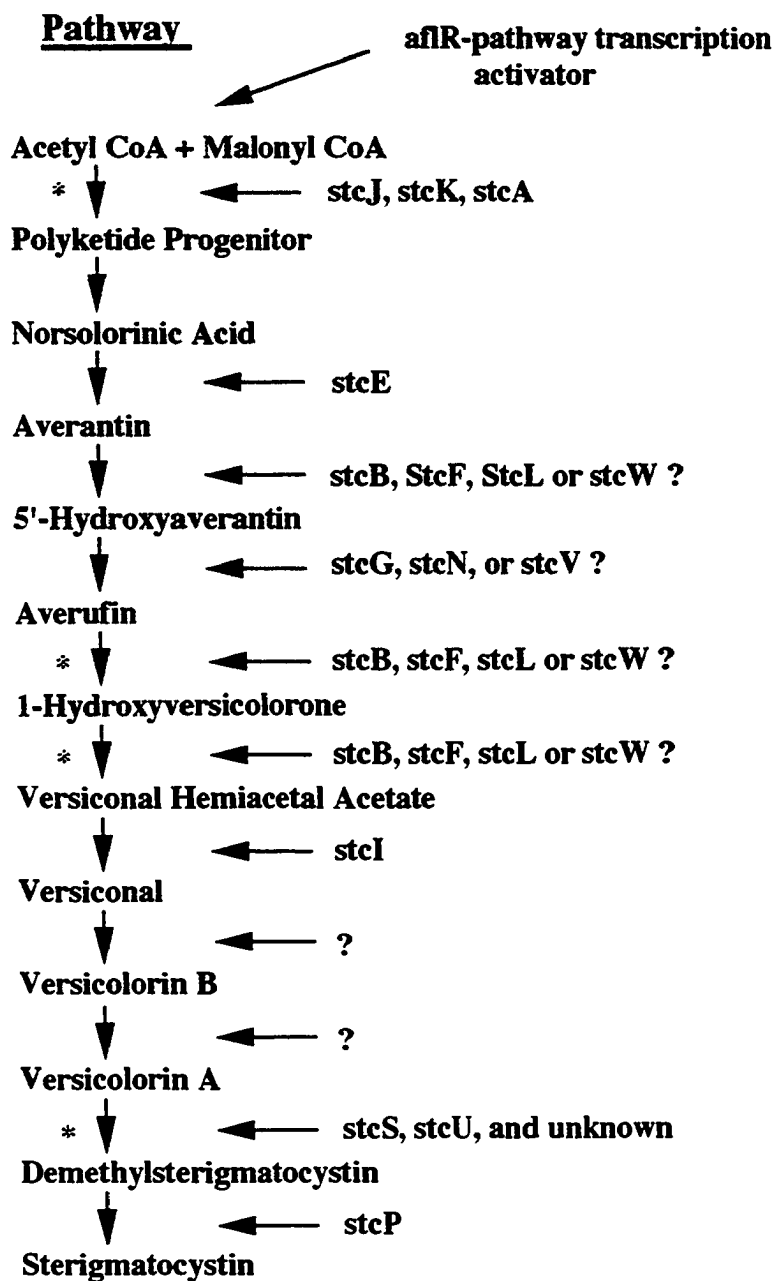


Figure 30. The proposed sterigmatocystin biosynthetic pathway. The cluster genes whose product is potentially responsible for particular steps are shown. * indicates a step for which no biochemical evidence has been found. ? indicates that the gene product involved in the step is not known or it is not clear which of several with similar activities are involved in a pathway step (adapted from Brown et al., 1996).

subject of current study (Chan, 1999; Fernandes, 1998; Guzman-de-Pena, 1998). The sterigmatocystin pathway of *A. nidulans* thus can serve as a model system to study regulation of aflatoxin production in other members of the *Aspergillus* family, including *A. parasiticus*, *A. flavus*, and *A. fumigatus* which do not have well studied genetic systems (Adams, 1998; Guzman-de-Pena, 1998) but have been shown to contain at least part of the aflatoxin gene cluster (Keller, 1997). Infection of crops such as corn by these organisms leads to large crop losses annually. For example, in Texas during the 1990 and 1995 growing seasons it was estimated that 50% and 30% respectively of the \$400 million crop was lost due to unacceptable levels of aflatoxin contamination (Breaux, 1995). In addition, in low income nations, including regions of China and sub-Saharan Africa, consumption of infected grain leads to high incidents of liver cancer marked by a specific G-T transversion in the p53 tumor-suppressor gene (Bressac, 1991; Hsu, 1991). Finally, both *A. fumigatus* and *A. flavus* increasingly were reported as the causative agents of systemic fungal infections of immunocompromised patients (Iwen, 1997; Nenoff, 1996) where the possibility of low level exposure to aflatoxin increases the already serious nature of the disease.

In fungi, pathway genes for secondary metabolites are often clustered within a gene dense region as occurred for the genes belonging to the sterigmatocystin pathway. Recently, the region of *A. nidulans* chromosome IV containing the sterigmatocystin gene cluster was sequenced and revealed many of the members of the sterigmatocystin/aflatoxin pathway (Brown, 1996). Since this pathway was of considerable interest to health and agricultural researchers, the EST database was examined to determine if the cDNA library contained members derived from this cluster. Although, the RNA for this cDNA library was harvested at 24 hours into development, a time earlier than *st/afl* RNA had been detected by hybridization (Brown, 1996), EST sequences for members of this cluster were present in the database at relatively high levels. This allowed the comparison of those homologous ESTs from the Unigene

database with the *A. nidulans* sterigmatocystin gene cluster genomic sequence and resulted in several annotation corrections and the identification of a new cluster gene.

The genes in this cluster are named *stcA* through *stcX* in physical order arbitrarily from left to right for this region of the genome, except for the sixth gene in the cluster, *aflR*, the pathway transcriptional regulator (Brown, 1996). A number of the sterigmatocystin genes were represented by multiple cDNA clones in the EST database. Although three of the proposed ORFs in the cluster, *stcD*, *stcO*, and *stcX*, lacked EST homologs the remaining 21 proposed genes in the cluster were represented by between one and 22 cDNA clones. Interestingly, ORFs *stcH*, *stcM*, and *stcR* were assigned but the function or role of their gene products in the sterigmatocystin pathway were unknown and it was unclear if these genes were expressed. The presence of ESTs for these ORFs supported the idea that they were functional genes. The identification of homologous cDNAs indicated that *stcH*, *M*, *Q* and *R* were genes located in the cluster region with no identified function in the sterigmatocystin biosynthetic pathway but which were expressed at the same time as many of the cluster genes. This observation suggested that they indeed may be members of the sterig/afla pathway as originally proposed (Brown, 1996). A detailed discussion of the alignments of the ESTs and EST consensus sequences with homology to the sterigmatocystin region GenBank accession number U34740 (Brown, 1996) is included below where all references to genomic sequence were to the region described in this GenBank entry. Table 20 at the end of section 3.26 gives a summary of the finding of the Unigene database comparison to the *stc* cluster sequence.

stcA, the first gene in the cluster, encodes the polyketide synthase responsible for assembling the sterigmatocystin carbon skeleton (Katz, 1993). The consensus sequence obtained by the alignment of the three matching clones covered the 3' end of the gene. Although only two introns had been predicted, the EST consensus sequence reveals a third with conserved intron-exon borders, lengthening the predicted protein by 16 amino acids at the C-terminal.

stcB is a p450 monooxygenase homolog, which along with stcF, stcL, and stcW did not have an assigned role in the cluster. Two cDNAs with homology to stcB confirmed the presence of one of three predicted introns. A small region 14 Kbp to the right of stcB, between stcI and stcK at position 24,274-24368, also aligned with 93% identity to the stcB cDNAs and the stcB ORF. This indicated that at least a portion of the stcB gene might have been duplicated during the evolution of this gene cluster.

stcC, a predicted peroxidase, had homology to one cDNA. When aligned with the genomic sequence revealed a single large ORF as reported earlier (Brown, 1996).

stcE, a putative ketoreductase had homology to two EST clone pairs in the Unigene database. The Unigene consensus differed slightly from the genomic sequence by the insertion of an A residue at 14,899, a T residue at 14,189 and the deletion of a G residue at 14,893. These changes result in an additional intron and a product that differed by 5 amino acids and added 3 additional amino acids to the originally predicted protein (figure 31). The final predicted protein would have 263 amino acids.

Additional investigation of the EST sequences which aligned with the stcE region showed an unexpected result. p0b05a1.r1, a 3' EST, could not be assembled with the three other ESTs aligning in this region even though it aligned with 99% identity to the stcE genomic region. When p0b05a1.r1 was compared with the EST consensus, the alignment revealed that p0b05a1.r1 contained the first intron and probably represented an incompletely spliced mRNA.

Another interesting observation was that the ESTs from cDNA, r1a06a1, aligned with the stcE Unigene consensus sequence but in the opposite orientation and was found instead to represent the 3' end of the adjacent aflR gene. The position of r1a06a1 showed that for these convergently transcribed genes, the 3' exon of aflR overlapped the 3'exon of stcE for a 36 basepair region (Figure 32). Overlapping 3' ends of convergently transcribed genes were also seen in the spermidine synthase and transketolase regions of W02H02-W30B01.

Percent Similarity: 96.923 Percent Identity: 96.923
 | = IDENTITY
 : = 2
 . = 1

stcE.EST x stcE.cosmid

```

EST      6 MPSAAVSVPEVPSSDRKTVVYLVTGASRGLGRGLVQAFLLRPNSIVIAGLR 55
          |||
Cosmid 1 1 MPSAAVSVPEVPSSDRKTVVYLVNRCQOG...GLVQAFLLRPNSIVIAGLR 47
          |||
          56 NRTSQAGALDALPRGENSSLIAVQLDSGSKSDPADAVSILQRDYGITHLD 105
          |||
          48 NRTSQAGALDALPRGENSSLIAVQLDSGSKSDPADAVSILQRDYGITHLD 97
          |||
          106 VVIANAATAANYGPASTMPLEYLETHMQINAYAALLLFQATRVLLQAAKS 155
          |||
          98 VVIANAATAANYGPASTMPLEYLETHMQINAYAALLLFQATRVLLQAAKS 147
          |||
          156 PQFICVGAPISTITEMESCARAPLTNYALSKLAACYLVRKIHFKWLVVA 205
          |||
          148 PQFICVGAPISTITEMESCARAPVTNYALSKLAACYLVRKIHFKWLVVA 197
          |||
          206 YIVDPGHIQSDMGAQAARLFGRKEAPTTIEESVAGICARMTEADKNTTSG 255
          |||
          198 YIVDPGHIQSDMGARSARLFGRKEAPTTIEESVAGICARMTEADKNTTSG 247
          |||
          256 RFILFSDGSDVPW 268
          |||
          248 RFILFSDGSDVPW 260
  
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Figure 31. A Bestfit alignment of the translation product of the stcE Unigene consensus (stcE.EST) with the U34740 stcE translation product (stcE.cosmid) showing the protein alignments.

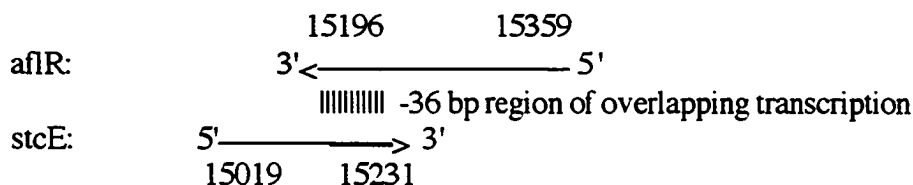


Figure 32. Region of overlapping convergent transcription between the aflR and stcE genes of the sterigmatocystin gene cluster. Overlap was defined by the homologous ESTs for these two regions, see text. Vertical lines indicate the overlap region. Numbers indicate exon endpoint positions in U34740.

Four cDNAs matched stcF, a predicted p450 monooxygenase, and confirmed the presence of its single intron but revealed its correct position at nucleotides 19946-19865, rather than 19996-19915 as reported (Brown, 1996). The entire coding region of stcF also was represented by the Unigene consensus sequence.

The EST analysis of stcG, a predicted dehydrogenase, revealed the position of its introns which were not determined by the earlier work and allowed prediction of a complete protein sequence. This 305 amino acid protein shared significant identity with the adhA gene from the *A. paraciticus* aflatoxin gene cluster that appeared to be regulated by nitrate and nitrite assimilation (Ehrlich, 1999). The Unigene consensus sequence differed from the cosmid sequence as two A residues occurred in the cosmid sequence at positions 21326-27 which introduced a stop codon, while the EST consensus had only one A residue which allowed the prediction of a larger open reading frame and the 305 amino acid protein with homology to the *A. paraciticus* homolog.

The ORF for stcH was not presented in Brown et al. (Brown, 1996). They originally mapped a 600 bp transcript to this region but could find no definitive ORF or any predicted protein with a homolog in GenBank (Brown, personal communication). The cDNA clone, h4b11a1, which mapped to stcH, was 480 bp in length and appeared to contain the complete coding region of stcH from U34740 position 22750-22372. The Unigene EST sequence differed from the genomic sequences by G deletions at position 22746 and at position 22632. Based on the EST sequence information, two introns with conserved intron-exon borders and an 83 amino acid protein which has no homolog in the databases was predicted.

ESTs from seven cDNA clones mapped to the stcI region and revealed a new intron and a new C-terminal exon which was not predicted previously. The translation of stcI thus was extended by the addition of eleven 5' amino acids from this additional 5' exon. The Unigene consensus allowed determination of the correct protein sequence as

well as indicating that this was an abundant transcript in the library.

ESTs from two cDNAs (Table 20) aligned with *stcJ*, a predicted alpha subunit of fatty acid synthase. The 3' third of the gene had 100% homology with the consensus sequence.

The g9b02a1 EST sequences had an exact match with the genomic *stcK* sequence which coded for a fatty acid synthase beta subunit. Although, the complete 5' end was not contained in the cDNA clone, the presence of two introns is confirmed.

The p450 monooxygenase coded for by *stcL* is represented by eight homologous cDNA clones which placed its representation in the abundant class of transcripts in the Unigene database. Their consensus sequence confirmed the one predicted intron.

stcM also was represented in the abundantly expressed class of the Unigene database by 15 clones whose ESTs showed homology to *stcM*. The consensus provided the entire coding region for this gene. This was another region that was not discussed in Brown et al. since they had no knowledge of the direction of transcription and the predicted ORF had no GenBank homolog. The presence of homologous ESTs predicted a gene with one intron that would encode a 148 amino acid protein that was transcribed in the same direction as is *stcL*. The Unigene consensus also differed from the published sequence in two positions and an intron was predicted in the 5' UTR.

stcN also was very highly represented in the EST database with 22 cDNA clones. The consensus sequence predicted two introns and had a 97 bp 3' overlap with the 3' end of the *stcO* gene. Interestingly, region 895-2024 of the *stcN* EST consensus sequence showed an exact match to a region between genes *stcO* and *stcP*, positions 41601-42612 of the genome cluster region, an apparently duplicated region within the sterigmatocystin cluster.

One cDNA showed homology to *stcO*. As mentioned above, this gene shared a 97 bp 3' overlap with *stcN* as predicted by Brown et al. and the *stcN* EST consensus. The 3' end of the *stcO* gene also showed an extensive perfect match with the same region

as stcN. No gene had been characterized in this duplication region that began at position 41601, one base from the 5' end of the stcO gene and extended to position 43449. The 5' end of the stcO EST did not align to the genomic stcO sequence but instead aligned, with one G-C transversion, to the second region. This suggested that the stcO region was not the gene defined by the matching cDNA but that the duplicated region containing an alternative ORF was more likely the transcribed region. This new region, stcO*, had a three prime end sequence that was identical to the stcO gene but had a unique 5' end (Figure 33). Neither StcO or stcO* had any homologs in the GenBank databases.

The consensus sequence for stcP revealed an intron in the 5' UTR. U34740 position 44901-44937 which defines exon 1, showed a significant sequence difference from the Unigene consensus sequence of 14 insertions not present in the ESTs (Figure 34). This region was immediately upstream of and included the predicted AUG start site. Therefore, the correct start codon occurred 36 bases downstream at position 44872 which shortened the predicted protein by 12 amino acids from that described by Brown, et al.

There is only one Unigene cDNA clone with homology to stcQ. The EST sequences agreed well with the cosmid sequence and revealed no introns. The direction of transcription is the same for both stcP and stcQ. 161 bases of the 5' end of stcP was overlapped by the 3' end of stcQ from positions 45071-44911. Since the overlap for both genes was in their respective 5' and 3' untranslated regions, the coding sequence would be unaffected.

StcR was represented by nine cDNA clones but had no sequence similarity to any GenBank entries. The Unigene consensus sequence covered the coding region and confirms the two predicted introns, one of which was in the 5' UTR region.

StcS was present as a large, 1560 bp, ORF and was represented by two cDNA clones with homology to the stcS p450 monooxygenase. This was one of the few genes in the sterigmatocystin gene cluster without an intron.

D. Brown (personal communication) predicted a single intron containing

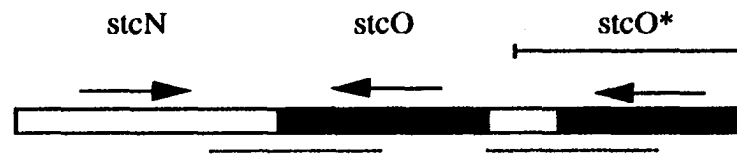


Figure 33. Sterigmatocystin gene cluster duplication region. Yellow indicates the sequences either coding for the *stcN* product or part of the the incomplete duplication. Green represents the 97 bp overlap between the *stcN* and *stcO* genes, also found in the duplicated region. *stcO** is shown as a composite region. Red is the region of gene *stcO** which is not homologous to *stcO* and contains a region homologous to clone *z7a04a1* not present in *stcO*. Red lines indicate the region of duplication, black line the *stcO** ORF, arrows indicate the direction of transcription.

```

C U34740      45071 GTTTCATTTTGGCCCGTGGACTTACCGGTTTCTTGGTGAATATAAGTTGC 45022
stcP          41 GTTTCATTTTGGCCCGTGGACTTACCGGTTTCTTGGTGAATATAAGTTGC 90

C U34740      45021 AGTCGAGAAAGGAGAGGGACAAGCAAGCTTAGGCAGACTCCATATTTTCAC 44972
stcP          91 AGTCGAGAAAGGAGAGGGACAAGCAAGCTTAGGCAGACTCCATATTTTCAC 140

C U34740      44971 CGTTCCTCCGTACTTATTACTTCCCATTTTCGGAGGTGAACTATTTACTC 44922
stcP          141 CGTTCCTCCGTACTTATTACTTCCCATTTTCGGAGC---ATCCAT--ACAT 185
                v--- v i -- vi

C U34740      44921 TTGTCTGATCTATCTATGCATTTGGTTGTTGACTGCACGTTTATCAATAA 4487
stcP          186 TTATC-G--CTGG---TG---TTGGTTGTTGACTGCACGTTTATCAATAA 226
                i - - - iv--- ---
                ***

C U34740      44871 TATGGACGCCATCTTCAAGCAAATCAAAGATGAGTACGCCCGTGCCGACG 4482
stcP          227 TATGGACGCCATCTTCAAGCAAATCAAAGATGAGTACGCCCGTGCCGACG 276

C U34740      44821 AGCATGGCAAGCGAGAGATTCAAGGCTATATCCGCGAGTTGCAGGTTGGC 4477
stcP          277 AGCATGGCAAGCGAAAGATTCAAGGCTATATCCGCGAGTTGCAGGTTGGC 326

C U34740      44771 TTCTATTCGGATGGGATGTGGTGTGATGCGGTTGAGCAGTGGT 44730
stcP          327 TTCTATTCGGATGGGATGTGGTGTGATGCGGTTGAGCAGTGGT 368

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Transitions / transversions = 1.25 (5 / 4)
Gap_init rate = 0.02 (6 / 328), avg. gap size = 2.33 (14 / 6)

Figure 34. Exon 1 of the stcP gene sequence from U34740 aligned with the consensus Unigene stcP sequence showing the sequence differences (underlined). The U34740 predicted AUG start site is highlighted and shown matching the corresponding EST consensus region. The new predicted start site based on the EST consensus sequence is also highlighted and marked with a (*).

conserved intron/exon borders for stcT. This was confirmed by the matching EST sequences from the single cDNA with homology to stcT but the positions of the ORF changed from the previously reported 48798-48863 to 50348-50401.

Five cDNA clones had homology to stcU. The consensus sequence extended to the 5' end of the gene and the two predicted introns were verified.

The Unigene consensus sequence generated from the three stcV homolog cDNA clones spanned the entire gene and confirmed that stcV had two predicted introns.

Brown et al. (Brown, 1996) stated that the stcW gene had homology to an FAD monooxygenase. However, no annotation of this region was given in the U34740 accession since the position of the introns was not clear (D. Brown, personal communication). Eight cDNA clones matched the stcW gene region and the Unigene consensus sequences revealed the positions of four introns (Table 21) and a final 424 amino acid protein.

3.27 Sterigmatocystin gene cluster relative expression level

It was possible to compare relative expression levels by examining the number of ESTs which were homologs to the predicted genes in the sterigmatocystin gene cluster. The various levels of individual gene expression ranged from no ESTs, to one and up to 22 ESTs (Figure 35). Although the direction of transcription for the genes in this cluster was determined, see Figure 35, it does not appear to correlate with the observed expression levels or their assigned role in the proposed biosynthetic pathway (Figure 30). There were four genes whose expression was exceptionally high, stcL, stcM, stcN and stcR. Since three of the genes were adjacent to each other and it has been suggested that selected enzymes in the sterigmatocystin biosynthetic pathway may have multiple functions or may be involved in another pathway (D. Brown, unpublished results), these four genes, two of which have no known function, would be candidates for investigation as a microcluster within the sterigmatocystin biosynthetic cluster.

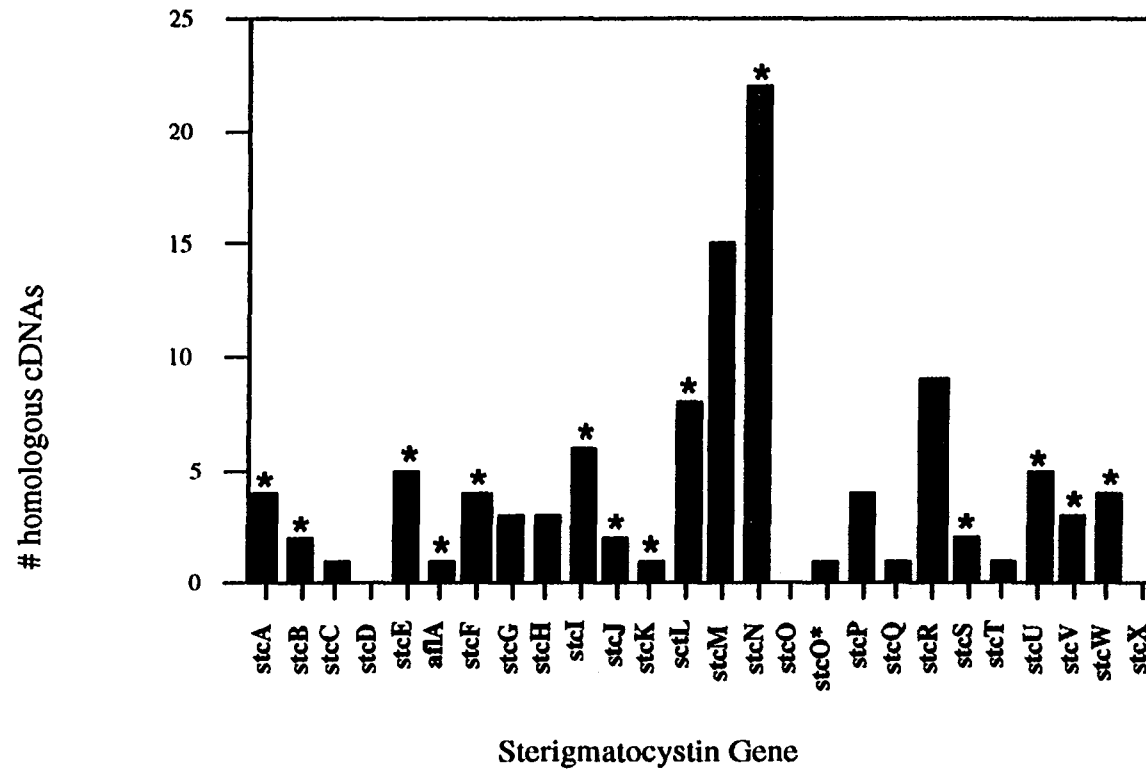


Figure 35. Histogram showing cDNA representation for each member of the sterigmatocystin gene cluster. Red bars represent genes transcribed to an arbitrary right, blue are genes are transcribed to the left. Genes are listed in order as they appear in the cluster. Stars indicate those genes assigned specific function in the sterigmatocystin biosynthetic pathway.

3.28 Sterigmatocystin summary.

Previously it had been thought that expression of the sterigmatocystin pathway did not begin until after 24 hours of development when the sterigmatocystin toxin could be detected (Brown, 1996). The presence of expressed pathway members therefore was surprising since this library was constructed from RNA harvested at 24 hours into asexual development. A total of 107 cDNA clones or 1.2% of the total genes in the EST database represented sterigmatocystin pathway members.

Table 20 gives a summary of the cDNA clones which had homology to the sterigmatocystin gene cluster. The genes are listed in the order in which they are present in the cluster. A brief summary of the result of the Unigene comparison to the specific genes in the cluster described in detail in section 3.26 is also given in Table 20 indicating whether a change to the cluster sequence was suggested by the comparison. The sterigmatocystin EST sequences were confirmed by resequencing the cDNA templates. In ten cases, sequences of the ESTs differed from the previously reported sequence (accession number U34740). The corresponding intron-exon borders sites were determined which allowed predicting correct amino acid sequences. This resulted in demonstrating overlapping transcription units and gene duplications. In one instance, the stcO* gene was found in a duplicated region that was shown to encode a transcribed gene which had not been predicted previously. Since the previously predicted stcO gene did not have any matching ESTs but the stcO* gene did, it remains to be determined whether only one or both of the regions contain actively transcribed genes.

3.29 A Consensus Intron-Exon Splice Site for *Aspergillus nidulans*.

No consensus intron-exon splice site sequence has been published for *A. nidulans*. However, now that a large number of ESTs have been obtained from this present study along with the genomic sequences of cosmids W02H02, W30B01, W06E08, and the sterigmatocystin biosynthetic gene cluster, it was possible to generate

Table 20. cDNAs with homology to *A. nidulans* sterigmatocystin biosynthetic gene cluster, accession number U34740. The gene is listed with its proposed function assignment. The homologous cDNA for each are listed. The corrections or confirmations of the reported annotation are given.

Pathway gene, proposed function	Homologous cDNAs in Unigene consensus	Alignment results
StcA, polyketide synthase	c8f01, g5g07, g9h10, c0d01	*third exon found, corrected protein product
StcB, p450 monooxygenase	d2f11, y8c09	one intron confirmed, duplication shown
stcC, peroxidase	h4e06	Confirmed reported sequence
stcD, ORF	-	not detected
StcE, ketoreductase	r4c09, p0b05, z5d08, r3b09, b0g10	*corrected sequencing error, additional intron found, corrected protein product
af1A, transcription factor	r1a06	convergent transcription with stcE shown to have 44 bp overlap of 3' ends
StcF, p450 monooxygenase	c9f01, h0f02, n5a05, a1f01	*corrected reported intron position
StcG, ORF	o6d04, o3d02, c7h01	*sequencing error corrected
StcH, ORF	i3d06, h4b11, y6h07	*sequencing error corrected, predicted two intron, 83 amino acid protein
StcI, lipase/esterase	g1d11, d4h03, r4g11, z6a11, e4a01, i7g11	*additional intron found, addition of 11 amino acids at 5' end

StcJ, fatty acid synthase-alpha subunit	n2e04, n2e10	Confirmed reported sequence
StcK, fatty acid synthase-beta subunit	g9b02	Confirmed reported sequence
StcL, p450 monooxygenase	w6h04, u4e02, m8f09, g2d05, h8g08, z5d12, f1b07, h0a05	Confirmed reported sequence
StcM, ORF	a1f04, z2f12, e6h09, s8h07, k8a06, u4e12, m9c04, p0f09, f1f08, h0b10, c4h10, y1e12, i0h09, y9e07, n3g07	*Sequencing error corrected, and 5' UTR intron shown.
StcN, GMC Oxydoreductase	u4g01, o0d06, h4g08, g9b11, x5h05, m6d05, y4b10, c9g02, c0g01, k5d04, m7f06, z3f08, h4a10, i8c11, v3b12, r8c03, g0f08, b0a06, n3c04, r8c03, c9c04, o9g06	Duplication found between stcO and stcP, includes 97 bp 3' overlap with 3' end of stcO
StcO, ORF	-	Not detected
StcO*, ORF	z7a04	*New gene identified, in partially duplicated region
StcP, O- methyl transferase	y4f12, c5e01, g6g10, c0c04	*Possible sequence error, correction shortens protein by 12 amino acids
StcQ, ORF	z4c07	3' end of stcQ overlaps 5' end of stcP
StcR, ORF	g4e02, r4g01, v1g04, x7g01, g2c03, j9b06, y6g09, h4f02, f1b10	Confirmed reported sequence

StcS, p450 monooxygenase	y1h01, x7g01	Confirmed reported sequence
StcT, Elongation factor 1 gamma	h4g09	*Corrected reported intron position
StcU, ketoreductase (verA)	d1e06, z4b06, f2c06, j7f11, w8e11	Confirmed reported sequence
StcV, dehydrogenase	o4e08, p0d10, b0f03	Confirmed reported sequence
StcW, FAD-containing monooxygenase	w6h01, g5a07, m8f12, u4b09	*4 introns found and 424 amino acid protein predicted.
StcX, ORF	-	not detected

* Unigene homolgs suggested a correction or addition to the sterigmatocystin cluster sequence U34740

the consensus splice sites for *A. nidulans* as presented in Table 22. The consensus derived from the examination of 49 introns was very similar to the eucaryotic consensus determined earlier by Mount (Mount, 1982) with the exception of position eight where a T residue was more prevalent in *A. nidulans* but no preference was noted in the Mount consensus and at position nine, where either a T residue or an A occurs in *A. nidulans* differs from the eucaryotic consensus.

3.30 Gene Density and Total Gene Number.

The prediction of gene density by combined EST and genomic sequencing allows a more accurate estimation of the number of genes contained in an organism's genome (Kupfer, 1997). The *A. nidulans* genome size and gene number have been independently estimated (Kupfer, 1997; Timberlake, 1978; Timberlake, 1980). Timberlake hybridized excess mRNA from varying growth conditions to single copy DNA to estimate that 13% of the genome was transcribed suggesting that *A. nidulans* contained about 6000 transcribed genes. More recently, Kupfer, et al. estimated the number of genes in the genome by applying the gene density determined for cosmid W06E08 (1 ORF/2.9 Kbp) to the 31 Mbp *A. nidulans* genome for an estimate of 10,000 genes. The sequence of the two overlapping cosmids W30b01 and W02H02 from a 45Kbp genomic region and the availability of the reannotated 60 Kbp genomic region of the sterigmatocystin cluster gave two additional genomic regions to further refine the estimate of average gene density and total gene number. The gene density for W30B01-W02H02 with its 17 predicted ORFs, was 1 gene/2.6 Kbp and the sterigmatocystin cluster region with 26 ORFs, including stcO*, was 1 gene/2.3 Kbp. An average of all three genomic regions gave a total of 56 genes in 144 Kbp or 1 gene/2.57 Kbp, suggesting that there are approximately 12,000 genes in the 31 Mbp genome. If this is a reasonable estimate, then the 3198 genes which were detected by this EST sequencing project represent approximately one third of the gene complement of *A. nidulans*.

Table 21. *A. nidulans* intron-exon splice sites for three chromosome VIII cosmids and the sterigmatocystin gene cluster of chromosome IV showing a consensus derived from 26 genes and 49 introns.

Gene Homolog	Left Position	Left Exon	Left Splice Junction.....	Intron Size(bp).....	Right Splice Junction	Right Exon	Right Position
Cosmid W06e08							
brlA	11303	β	CCCTCAGTCAGT	.1011..	TCGAACACATGT	α	12314
ribonuclease T2	24521	1	CCTCTGGTGCGC	...50..	GGCCAGATAACT	2	24570
	24779	2	AGCACGGTACTT	...34..	TAACAGGAACTT	3	24814
	25136	3	AGAAGCGTTAGT	...67..	CTCTAGTGACCC	4	25202
dihydroorotase	33166	1	TCCACGGTGCGT	..172..	AAATAGTCCACC	2	32993
	33088	2	GTTATGGTAAGG	...90..	TGATAGCCAAAT	3	33177
	33634	3	GGAACAGTAAAT	...52..	TTGTAGTTCCG	4	33682
Cosmids W02H02 and W30B01							
prohibitin	12073	1	TATCAGGTTCGT	...60..	TTTTAGTCCTAC	2	12014
spermidine synthase	14151	1	TCAAGGGTATGC	..159..	ATCCAGATGGCT	2	14309
	14491	2	GTTCTCGTATGC	...86..	TTCCAGCTACCA	3	14575
	14725	3	GATGAGGTAACA	..147..	TTGCAGGCCGTC	4	14831
	15052	4	ATGGAGGTGTCA	..274..	TTACAGCCGAAA	5	15325
Transketolase	18548	1	CTTGCGGTATGC	..262..	TTCCAGGTTGAT	2	18286
	18142	2	CCTCTCGTATGA	...54..	CTATAGCAACGG	3	18089
	18003	3	TTCCGTGTAAGC	...69..	TTATAGCAACTC	4	17935
Acetate reg. DNA binding	22654	1	TTGCTGGTATGG	..482..	GTCAAGCTGTCT	2	22596
	21942	2	GCTATTGTAAGT	..312..	CTGTAGTCGAGA	3	22228

protein (facB)	21536	3	CGGTTTCGTATCC...283..TCTTAGTCTTTG	4	21895
	20323	4	CTTTTGGTAAGT...50..TTAAAGTCTGTG	5	21487
Spac3c7.01c	26870	1	GGGAAGGTATGG...95..ATCCAGATCTCG	2	26777
	26305	2	GAGGAAGTACT...76..GGTCAGGATAAA	3	26230
Exp. gene 1	31066	1	ATGAAGGTAAGC...50..GTGCAGTCCATT	2	31115
HMG-like protein 2	34051	1	AGAAGGGTAAGC...128..CTACAGCTGCGG	2	33889

Sterigmatocystin Gene Cluster

stcA	1907	1	CTGATGGTATGT...62..TCACAGACGAAG	2	1846
stcB	10187	1	AGACACGTAAGT...72..GGCTAGGTTCGA	2	10258
StcF	19946	1	GGAGGGGTAAGT...83..TTACAGACGACC	2	19865
StcG	22062	1	TACATGGTACAT...69..CTACAGGACGTG	2	22130
StcH	22730	1	ACGTCGGTATGC...53..CGACAGCCCTAT	2	22678
	22457	2	TGACCAGTAAGT...58..CGACAGAGTTCA	3	22400
StcI	22975	1	CAGCAGGTACAG...48..TACCAGTTCATA	2	23022
StcK	35585	1	GATCAGGTATCT...50..TCGTAGACTTCA	2	35634
	35751	2	GGCCATGTAGTA...67..GAACAGGTCCGC	3	35817
StcL	36770	1	GCTGAGGTGTGT...66..CTCTAGACCCGC	2	36705
StcM	38588	1	TGTCCGGTAATG...46..CACCAGGTGCAA	2	38543
StcN	39281	1	CAGCCAGTATGC...47..TTTCAGGGGCTG	2	39327
	39411	2	TCATCGGTATCT...44..AGACAGGGGCTC	3	39454

StcP	44729	1	AGTGGTGTATGT...47..GTGTAGCCCTTG	2	44683
	44555	2	TGCCTTGTAAGT...51..CAACAGGGCGCA	3	44505
	44380	3	ACAGCTGTATGT...50..TCTCAGATTCGA	4	44331
stcR	46167	1	TCCATAGTAAGT...45..GTACAGATCACA	2	46211
	46517	2	ACTCTGGTAGGT...46..AGGCAGGTTTTC	3	46562
stcT	50348	1	TCTATGGTACCA...54..GTATAGTAACAT	2	50401
stcU	51601	1	CCAGATGTATGC...51..CAGCAGGAATTC	2	51551
	51226	2	GATGAGGTACGT...46..GGACAGTGTGCC	3	51181
STCV	52738	1	CCGAGAGTATGT...48..TACCAGCCTATC	2	
STCW	53875	1	GCACTTGTTTGT...55..TCGAAGTACCCC	2	53929
	54179	2	CCAAAGGTGCGC...46..CTGCAGTGGCCA	3	54224
	55174	3	GCAGCTGTACGT...95..CACCAGGGTACA	4	55268
	55495	4	AGTTTGTAAGT...46..CAATAGGTTCAA	5	55540

Summary for 26 genes and 49 introns:
Average intron size-114 bp, range 45-1011 bp

Eucaryotic consensus: AG-G T A ^C A G T...T N C C A G-N
[Mount, 1982 #140] T G G

A. nidulans
Consensus motif: NG-G T A ^C A G T...T T N C A G-N
 T G

percent bases at	1	2	3	4	5	6	7	8	9	10	11	12	
position for 49 introns:	q-	G	T	A	C\A\T	G	T....C/T/G	T	N	T\C	A	G-	
	56	100	100	82	95.5	75	58	95.5	53	-	64	100	100

Chapter IV

Summary and Conclusion

During this work the sequence of over 12,000 ESTs were generated from an *A. nidulans* cDNA library that contained mRNAs from both vegetative and asexually expressed genes. To process the data in a rapid and orderly fashion, an automated screening method for handling a large number of sequences was developed. The sequences of each EST were released on the ACGT website and deposited in the GenBank dbEST.

The individual ESTs were aligned into a Unigene database with the Phred/Phrap sequence assembly programs (Green, copyright 1993-1996; Green, copyright 1994-1996) at various stages of the data collection. This allowed following the sampling progress of the cDNA library by estimating the efficiency of the sampling rate for unique 3' sequences and determining the endpoint of sampling by measuring how frequently new sequences were seen as the project progressed. In addition, the Unigene database aligned the respective 3' and 5' EST sequences into gene families yielding a consensus sequence with a greater accuracy than was obtained for each EST individually because of multiple overlapping EST sequences.

The Unigene consensus sequences also were examined for homologs via BlastX against the GenBank nonredundant protein database. These studies helped organize the data based on biological function. The resulting metabolic outline revealed a glimpse into the biochemistry of *A. nidulans*. These results were placed on the ACGT website where they are available for keyword searches and a summary of this data is presented in the appendices. Developing a Unigene database and the Biological Function outline allowed both estimating the numbers of genes expressed in the *A. nidulans* cell during vegetative and asexual development, as well as their role in the cell and revealed several examples of paralogous genes.

The heat shock protein 30 (HSP30) represented such a paralogous gene group in *A. nidulans* that contained at least six genes. Comparative analysis of the *A. nidulans* HSP30 paralogs revealed that the *N. crassa* and *A. nidulans* HSP30s are as closely related to *S. cerevisiae* as they are to bacteria or archae HSP30s.

Examination of the biological function outline shown in Appendix II also reveals that a number of other gene families including chitinases, serine/threonine kinases, and phosphatases as well as other classes of HSPs are present in this Unigene database. Thus, as with the HSP30 genes, new predictions based on sequencing, which otherwise were unavailable, now can be tested with more traditional genetic and molecular methods.

Examination of the very abundantly expressed Unigene members with no GenBank homologs suggested that additional studies will be required to identify conserved domains or motifs which may suggest the function or cellular location of EST sequences with no homologs in GenBank.

An important application of ESTs also has been demonstrated for genomic sequence annotation. The three cosmids which were sequenced and annotated supplied 83 Kbp of Chromosome VIII data for comparison to the EST database. These studies elucidated the structure of new genomic regions such as those for a transposon belonging to a class not found previously in fungus as well as the consensus sequence for fungal intron/exon borders and sequences of several new genes. The Chromosome IV region encoding the sterigmatocystin pathway had a large number of homologous ESTs. This EST information allowed reexamination of the sterigmatocystin cluster transcription units and revealed several new exons, a new gene and corrected previous sequencing errors in this region. The presence of the sterigmatocystin ESTs also suggested that the time of appearance of the toxin, sterigmatocystin, occurs more than 12 hours after the mRNAs in the pathway are synthesized (Brown, 1996; N. Keller, personal communication).

Intron/exon borders identified in the cosmids and sterigmatocystin cluster region were confirmed by the EST sequences and a more representative set of 3' and 5' splice

site consensus sequences was developed that was determined to be quite similar to those seen in mammalian systems. Finally, the present EST and genomic sequencing study provided the data needed to give a more accurate estimate (~12,000) of the number of genes in the *A. nidulans* genome.

The EST database produced during this dissertation research and the genomic regions sequenced provided a valuable tool for determining both gene content of this eucaryotic organism and for its genomic annotation. The study presented here will serve as a foundation for dissecting the molecular biology of *A. nidulans*. With the growing number of sequences available in the public databases, fruitful homology comparisons will continue to be made. As the fungal community looks toward the future when the entire sequence of *A. nidulans* and other fungal genomes will be completed, these *A. nidulans* ESTs will be an extremely useful resource for annotating future genomic sequences and beginning to understand gene expression in this and related fungi.

Chapter V

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Appendix I. *Aspergillus nidulans* categories of cellular functions with keywords. In bold are headings, in standard type are the keywords. & indicates a variation of the keyword in the line above, all entries matching the & keyword will be placed under the higher keyword. Some keywords are abbreviated or capitalized as needed for use in searching the BlastX reports.

Categories of Cellular Functions

I. Bioenergetics and Metabolism

A. Metabolism of Carbohydrates(for glucose see energy)

1. Chitin metabolism

chitin
&chitinase

2. Cellulose degradation

beta glucosidase-breakdown of cellulose
&beta glucosidase
&beta-glucosidase
&glucoside glucohydrolase
glucanase
&GNS1 PROTEIN
&beta glucanase
cellulase
cellobiohydrolase

3. Pectin degradation

pectin
pectate
2-deoxy-D-gluconate 3-dehydrogenase

4. Cutin metabolism

cutin

5. Polysaccharide synthesis

UDP-glucose dehydrogenase

6. Energy reserve synthesis-see also energy reserve metabolism

GLYCOGEN (STARCH) SYNTHASE
&glycogen synthase
1,4-ALPHA-GLUCAN BRANCHING ENZYME
starch branching enzyme
starch synthase
trehalose synthase

7. Arabinose metabolism

arabin

8. Glucosamine

GLUCOSAMINE--FRUCTOSE-6-PHOSPHATE AMINOTRANSFERASE
GLUCOSAMINE-6-PHOSPHATE ISOMERASE-GLUCOSAMINE UTILIZATION
PATHWAY
&GLUCOSAMINE-6-PHOSPHATE ISOMERASE

&GLUCOSAMINE-6-PHOSPHATE DEAMINASE

beta glucosamine

&glycoamidase

GLUCOSAMINIDASE-degradation of glycans

&GLUCOSAMINIDASE

GLUCOSAMINE--FRUCTOSE-6-PHOSPHATE AMINOTRANSFERASE

9. Aminosugar metabolism

PHOSPHOACETYLGLUCOSAMINE MUTASE

10. Sucrose metabolism

sucro

levanase-sucrose to glucose

&levanase

11. Galactose metabolism

galactose

GALACTOSE-1-PHOSPHATE URIDYLTRANSFERASE

12. Mannitol metabolism

manno

mannitol

13. Xylanose metabolism

xylosidase

xylanase

xylitol dehydrogenase

14. Quinate metabolism

quinate-utilization is in cluster

&quinate

15. Sorbitol metabolism

SORBITOL UTILIZATION PROTEIN

SORBITOL DEHYDROGENASE

16. Gluconate

Glucose Oxidase-first step of glucose + O₂ to gluconic acid

&Glucose Oxidase

17. Pyranose metabolism

pyranose oxidase

18. Ribitol metabolism

ribitol kinase

19. Calvin cycle

RIBULOSE-PHOSPHATE 3-EPIMERASE-ribulose-5 PO₄->xylulose-5 PO₄

&RIBULOSE-PHOSPHATE 3-EPIMERASE

B. Metabolism of Amino acids and Related Molecules

1. Arginine metabolism

a. Arginine anabolism-glutamine, CO₂ to arginine
ORNITHINE CARBAMOYLTRANSFERASE
ARGININOSUCCINATE SYNTHASE
ARGININOSUCCINATE LYASE
AGMATINASE

b. Arginine catabolism-arginine to proline
ARGINASE-also see urea cycle
&ARGINASE
ARG-6 PROTEIN
ACETYLORNITHINE AMINOTRANSFERASE
PYRROLINE-5-CARBOXYLATE REDUCTASE

2. Asparagine metabolism
ASPARAGINE SYNTHASE

3. Aspartic acid metabolism

aspartate anabolism-oxaloacetate, glutamate to aspartate
ASPARTATE AMINOTRANSFERASE
Aspartase

4. Cysteine metabolism
cysteine
O-ACETYLMOMOSERINESULFHYDRYLASE-also methionine biosyn
&O-ACETYLMOMOSERINESULFHYDRYLASE
&O-acetyl-L-homoserine sulfhydrylase
homocysteine synthase
CYSTATHIONINE GAMMA-LYASE

5. Glutamine metabolism
GLUTAMINE SYNTHETASE

6. Glycine metabolism

a. Serine to glycine
GLYCINE HYDROXYMETHYLTRANSFERASE

b. glycolate to glycine
GLYCERATE DEHYDROGENASE
SERINE--PYRUVATE AMINOTRANSFERASE

c. threonine to glycine
THREONINE ALDOLASE
PYRIDOXAMINE-PHOSPHATE AMINOTRANSFERASE

d. glycine catabolism

-glycine to serine
GLYCINE HYDROXYMETHYLTRANSFERASE

e. glycine decarboxylase complex-made up of P,T,L,H-removes amino group
GLYCINE CLEAVAGE SYSTEM H PROTEIN

&glycine cleavage system protein H
Glycine cleavage system T protein
&AMINOMETHYLTRANSFERASE

7.Histidine metabolism
HISTIDINE BIOSYNTHESIS

8.Isoleucine metabolism
2,3-DIHYDROXYACID HYDROLYASE-4th step in iso & val biosyn
&2,3-DIHYDROXYACID HYDROLYASE
&DIHYDROXY-ACIDDEHYDRATASE

-catabolism
propionyl-CoA carboxylase-also leucine and valine degradation
&propionyl-CoA carboxylase
methylcrotonyl-CoA carboxylase

9.Leucine metabolism
hydroxy-3-methylglutaryl-CoA lyase-FINAL STEP OF KETOGENESIS AND
LEUCINE CATABOL
ISM
&hydroxy-3-methylglutaryl-CoA lyase
&HYDROXYMETHYLGLUTARYL-COALYASE

10.Lysine metabolism
HOMOCITRATEDEHYDRATASE
SACCHAROPINE DEHYDROGENASE

11.Methionine metabolism
methionine synthase
ORNITHINE AMINOTRANSFERASE
HOMOSERINE O-ACETYLTRANSFERASE
cystathionine beta-lyase-3rd step
&cystathionine beta-lyase
methionine synthase-last step in met biosynthesis
&methionine synthase
&5-METHYLTETRAHYDROPTEROYLTRIGLUTAMATE--
HOMOCYSTEINEMETHYLTRANSFERASE

12.Phenylalanine metabolism
phenylalanine hydroxylase

13.Proline metabolism
proline dehydrogenase

14.Serine metabolism
PHOSPHOSERINE AMINOTRANSFERASE

15.Tryptophan metabolism
anthranilate phosphoribosyltransferase-2nd step in tryp biosyn
&anthranilate phosphoribosyltransferase
CATECHOL 1,2-DIOXYGENASE-tryp & lysine catabolism in KETOADIPATE
PATHWAY
&CATECHOL 1,2-DIOXYGENASE

&hydroxyquinol-1, 2-dioxygenase

16. Tyrosine metabolism

prephenate dehydrogenase

17. Valine metabolism

hydroxyisobutyrate dehydrogenase

valine synthetase

METHYLMALONATE-SEMIALDEHYDEDEHYD

18. Aromatic amino acid metabolism

PHOSPHO-2-DEHYDRO-3-DEOXYHEPTONATEALDOLASE

19. Polyamine biosynthesis

polyamine

&spermidine

C. Metabolism of Nucleotides and Nucleic Acids, Purines, Pyrimidines

1. Nucleotide metabolism

NUCLEOSIDE DIPHOSPHATE KINASE

ribose-phosphate pyrophosphokinase-purine, pyrimidine biosyn, also his and trypto
phan biosyn

&ribose-phosphate pyrophosphokinase

2. Purine metabolism

a. inosine mono phosphate de novo biosynthesis

amidophosphoribosyl transferase

phosphoribosylamine-glycine ligase

phosphoribosylglycinamide formyltransferase

PHOSPHORIBOSYLFORMYLGLYCINAMIDINE SYNTHASE

PHOSPHORIBOSYLFORMYLGLYCINAMIDINE CYCLOLIGASE

amidophosphoribosyltransferase

ADENYLOSUCCINATELYASE

ADENYLOSUCCINATE SYNTHETASE-first committed step to AMP biosyn

&ADENYLOSUCCINATE SYNTHETASE

&IMP--ASPARTATELIGASE

PHOSPHORIBOSYLAMINOIMIDAZOLECARBOXAMIDE

FORMYLTRANSFERASE

&IMP cyclohydrolase

b. other purine metabolic enzymes

INOSINE-5'-MONOPHOSPHATE DEHYDROGENASE

glutamine phosphoribosylpyro

xanthine dehydrogenase

amidophosphoribosyl transferase

phosphoribosyl synthase

Purine Nucleoside Phosphorylase

AMPDEAMINASE

&MYOADENYLATEDEAMINASE

adenosine kinase-phosphorylates purine nucleoside

&adenosine kinase

3. Pyrimidine metabolism

a. de novo pyrimidine biosynthesis

PYRIMIDINE BIOSYNTHESIS ENZYME Thi5-thiamine biosyn
&PYRIMIDINE BIOSYNTHESIS ENZYME Thi5
carbamoyl-phosphate synthase
ASPARTATE CARBAMOYLTRANSFERASE
dihydroorotase
dihydroorotate
dihydroorotate dehydrogenase-4th step in pyr biosyn
&dihydroorotate dehydrogenase
orotate reductase
OROTATE PHOSPHORIBOSYLTRANSFERASE
orotidine

b. other pyrimidine metabolic enzymes

PHOSPHORIBOSYLPYROPHOSPHATE SYNTHETASE
DEOXYCYTIDYLATE DEAMINASE-degradation to dUMP
&DEOXYCYTIDYLATE DEAMINASE
&DCMP DEAMINASE

D. Metabolism of Lipids, Fatty Acids, Sterols-See also fatty acid degradation

1. Fatty acid biosynthesis

a. ACETYL-COA CARBOXYLASE-yields malonylcoA, committed step to FA biosyn.
&ACETYL-COA CARBOXYLASE

b. ACYL-CARRIER PROTEINS

ACYL-CARRIER-PROTEIN

c. FATTY ACID SYNTHASE

&FATTY ACID SYNTHASE
3-OXOACYL-[ACYL-CARRIER-PROTEIN] SYNTHASE
[ACYL-CARRIER PROTEIN] S-MALONYLTRANSFERASE
CROTONOYL-[ACYL-CARRIER PROTEIN] HYDRATASE
ENOYL-[ACYL-CARRIER PROTEIN] REDUCTASE (NADH)
3-ketoacyl-acyl carrier protein reductase
3-HYDROXYPALMITOYL-[ACYL-CARRIER PROTEIN] DEHYDRATASE
3-HYDROXYDECANOYL-[ACYL-CARRIER PROTEIN] DEHYDRATASE

d. BRANCHED-CHAIN ALPHA-KETO ACID DEHYDROGENASE-keto acids->short branch-chain fatty acids

&BRANCHED-CHAIN ALPHA-KETO ACID DEHYDROGENASE

e. Other

stearoyl-CoA desaturase-adds double bonds to fatty acyl coA
&stearoyl-CoA desaturase

2. Sterols

a. sterol

sterol
steroid monooxygenase

LANOSTEROL SYNTHASE
& LANOSTEROL CYCLASE

glucuronidase

HYDROXY-3-METHYLGLUTARYL-COENZYME A REDUCTASE-also mevalonate
biosyn->isoprenoids

& HMG-CoA-reductase

& HYDROXY-3-METHYLGLUTARYL-COENZYME A REDUCTASE

C-5 STEROL DESATURASE

sterol demethylase

& eburicol C14-alpha-demethylase

b. Farnesol biosynthesis

ISOPENTENYL-DIPHOSPHATE ISOMERASE

GERANYLGERANYL PYROPHOSPHATE SYNTHETASE

& dimethylallyltransferase

GERANYLGERANYL TRANSFERASE

hydroxysteroid dehydrogenase

c. cholesterol metabolism

C-4 METHYL STEROL OXIDASE-cholesterol biosynthesis

& C-4 METHYL STEROL OXIDASE

STEROL O-ACYLTRANSFERASE-esterification of cholesterol

& STEROL O-ACYLTRANSFERASE

3. Lipids

a. phospholipid biosynthesis

LYSOPHOSPHOLIPASE PRECURSOR

phospholipid biosynthesis

phosphatidyl synthase

PHOSPHATIDYLSERINE SYNTHASE

PHOSPHATIDYLSERINE DECARBOXYLASE

myo-inositol phosphate synthase-biosynthesis of inositol containing phospholipids

& myo-inositol phosphate synthase

& myo-inositol 1-phosphate synthase

& myo-inositol-3-phosphate synthase

b. SPHINGOLIPIDS

serine palmitoyltransferase

c. Lipopolysaccharide biosyn-biomembrane precursors

UDP-glucose:sterol glucosyltransferase

UDP-GLUCOSE PYROPHOSPHORYLASE

& UTP--GLUCOSE-1-PHOSPHATE URIDYLYLTRANSFERASE

E. Aromatic compound metabolism

4-coumarate--CoA ligase-thioester substrates for phenylpropanoid biosyn

& 4-coumarate--CoA ligase

& coumarate:CoA ligase

& COUMARATE--COA LIGASE

chorismate

aminobutyrate aminotransferase

CARBOXYMUCONOLACTONE DECARBOXYLASE-aromatic hydrocarbon cat.

&CARBOXYMUCONOLACTONEDECARBOXYLASE

F. Sulfur Metabolism

SULFATE ADENYLYLTRANSFERASE

ADENYLYLSULFATEKINASE

sulphur metabolite repression-4 genes, met down, no S up

& sulphur metabolite repression

& sconC

-sulfate assimilation

sulfate adenylyltransferase-leads to biosynthesis of cys&met

& sulfate adenylyltransferase

& ATP-SULFURYLASE

G. Phosphate Metabolism

INORGANIC PYROPHOSPHATASE

& PYROPHOSPHATEPHOSPHO-HYDROLASE

H. Nitrogen Metabolism (see also amino acid metabolism)

nitrate reductase

nitrite reductase

NITROGEN METABOLIC REGULATION PROTEIN -NEGATIVE REGULATORY
PROTEIN IN THE NITROG

EN CONTROL CIRCUIT

& NITROGEN METABOLIC REGULATION PROTEIN

& nitrogen metabolite repression regulator

cyanate lyase-cyanate, bicarbonate substrates

& cyanate lyase

-urea cycle

urea cycle

glutamate dehydrogenase

CARBAMOYL-PHOSPHATE SYNTHASE-also arginine and pyrimidine biosynthesis

& CARBAMOYL-PHOSPHATE SYNTHASE

I. Metabolism of Cofactors, prosthetic groups

1. Nicotinamide coenzymes

nicotinamide adenine dinucleotidephosphate

NICOTINATE-NUCLEOTIDE PYROPHOSPHORYLASE-DE NOVO

BIOSYNTHESIS OF NAD AND NADP

& NICOTINATE-NUCLEOTIDE PYROPHOSPHORYLASE

kynureninase-biosyn of NAD cofactors

& kynureninase

& alpha-aminoadipate aminotransferase

2. Biocytin (biotin)

biotin carboxylase

3. Thiamine

thiamine

THIAMIN BIOSYNTHESIS

& THIAMIN-PHOSPHATE PYROPHOSPHORYLASE

& THIAMIN BIOSYNTHETIC BIFUNCTIONAL ENZYME

&NMT1 PROTEIN

4. Coenzyme A

acetyl-coenzyme A synthetase
&acetyl coenzyme A acetyltransferase

5.f Favins

riboflavin synthase
GTP cyclohydrolase II-riboflavin biosyn
>P cyclohydrolase II

6. Folate-methyl donor

folate

7. Heme

heme
siroheme synthase
##-iron uptake
FERRIC REDUCTASE TRANSMEMBRANE COMPONENT 2

8. PANTOTHENATE

PANTOTHENATE SYNTHETASE
&PANTOATE--BETA-ALANINE LIGASE

9. Molybdopterin

molybdopterin biosynth
&molybdopterin converting factor

J. Energy

1. Glycolysis

a. hexokinase

&hexokinase

b. glucose-6-phosphate isomerase

&glucose-6-phosphate isomerase

c. fructose-6-phosphate 2-kinase

&fructose-6-phosphate 2-kinase
&PHOSPHOFRUCTOKINASE
&phosphofructo-2-kinase
&fructose-2,6-bisphosphate 2-phosphatase

d. fructose-bisphosphate aldolase-also gluconeogenesis, PP cycle, carbon fixation, fructose and mannose metab

&fructose-bisphosphate aldolase

e. triose-phosphate isomerase

&triose-phosphate isomerase

f. glyceraldehyde-3-phosphate dehydrogenase

&glyceraldehyde-3-phosphate dehydrogenase

g.phosphoglycerate kinase
&phosphoglycerate kinase

h.phosphoglycerate mutase
&phosphoglycerate mutase
&PHOSPHOGLYCERATEDEHYDRATASE

i.phosphopyruvate hydratase
&phosphopyruvate hydratase

j.pyruvate kinase
&pyruvate kinase

2. Gluconeogenesis
a.LACTATE DEHYDROGENASE
&LACTATEDEHYDROGENASE

b.pyruvate carboxylase
&pyruvate carboxylase

c.phosphoenolpyruvate carboxykinase
&phosphoenolpyruvate carboxykinase

d.FRUCTOSE-1,6-BISPHOSPHATASE
&FRUCTOSE-1,6-BISPHOSPHATASE
&FRUCTOSE-1,6-BISPHOSPHATAS
&fructose-bisphosphatase

3. Pentose-phosphate pathway
a.glucose-6-phosphate dehydrogenase
&glucose-6-phosphate dehydrogenase
&GLUCOSE-6-PHOSPHATE 1-DEHYDROGENASE

b. gluconeolactonase
&gluconeolactonase

c.phosphogluconate dehydrogenase
&phosphogluconate dehydrogenase

d.ribose 5-phosphate isomerase-nonoxidative PO4
&ribose 5-phosphate isomerase

e.ribulose-phosphate 3-epimerase
&ribulose-5-phosphate 3-epimerase

f.transketolase
&transketolase

g.transaldolase
&transaldolase

4. Pyruvate dehydrogenase
pyruvate dehydrogenase
pyruvate decarboxylase

dihydrolipoyl transacetylase
&LIPOAMIDEACYLTRANSFERASE
dihydrolipoamide dehydrogenase
pyruvate dehydrogenase kinase-inhibits pyruvate dehyd by phos of E1 alpha subunit
&pyruvate dehydrogenase kinase

5. Tricarboxylic acid pathway

a.citrate synthase
&citrate synthase

b.aconitate hydratase
&aconitate hydratase

c.isocitrate dehydrogenase
&isocitrate dehydrogenase

d.alpha-ketoglutarate dehydrogenase
&alpha-ketoglutarate dehydrogenase

e.SUCCINYL-COA LIGASE
&SUCCINYL-COA LIGASE

f.succinate dehydrogenase
&succinate dehydrogenase

g.FUMARATEHYDRATASE
&FUMARATEHYDRATASE

h.malate dehydrogenase
&malate dehydrogenase

6. Related reactions

citrate lyase-citrate to oxaloacetate+acetylcoA
&citrate lyase
&citrate-lyase
&CITRATE CLEAVAGE ENZYME
&CITRATE (PRO-S-)-LYASE
carbonic anhydride
&carbonic

-related pathway

homoprotocatechuate degradation-degraded to krebs intermediates
&homoprotocatechuate degradation

7. Glyoxylate cycle

malate synthase
isocitrate lyase

8. Fermentation, alcoholic

a.pyruvate decarboxylase
&pyruvate decarboxylase

b.alcohol dehydrogenase
&alcohol dehydrogenase

9. Fermentation, other

LACTATE DEHYDROGENASE-pyruvate to lactate
& LACTATE DEHYDROGENASE

butanediol dehydrogenase

10. Monocarbon metabolism

formate dehydrogenase

-C1 Metabolism

ALCOHOL OXIDASE-first step-methanol utilization-> FORMALDEHYDE
& ALCOHOL OXIDASE

11. Metabolism of energy reserves (glycogen, starch, trehalose)

a. Glycogen degradation

glycogen phosphorylase
PHOSPHOGLUCOMUTASE-glycogen deg, glu1PO4 to glu6PO4
& PHOSPHOGLUCOMUTASE
& GLUCOSE PHOSPHOMUTASE

b. Starch degradation

alpha glucosidase
& alpha-glucosidase
& MALTASE
glucoamylase
& alpha-amylase

c. Trehalose degradation

trehalose-6-phosphate synthase
trehalase

12. Fatty acid degradation

a. lipase-triacylglycerols->glycerol+FA
& lipase

b. beta-oxidation of fatty acids
beta-oxidation

i. fatty acid activation-thiokinase
long-chain-fatty-acid-CoA ligase
medium-chain acyl-CoA ligase
LONG-CHAIN ACYL-COASYNTHETASE

ii. carnitine acetyl transferase
& carnitine acetyl transferase
carnitine racemase-d to l form
& carnitine racemase

iii. acyl-CoA dehydrogenase
& acyl-CoA dehydrogenase

iv. enoyl-CoA hydratase
& enoyl-CoA hydratase

v. 3-hydroxyacyl-CoA dehydrogenase
& 3-hydroxyacyl-CoA dehydrogenase

vi. HYDROXYBUTYRYL-COA DEHYDROGENASE
& HYDROXYBUTYRYL-COA DEHYDROGENASE

vii. 3-ketoacyl-CoA thiolase
& 3-ketoacyl-CoA thiolase
& 3-keto-acyl-CoA thiolase

c. odd chain fatty acids
methylmalonyl carboxylase-also ile, thr, met, val degradation
& methylmalonyl carboxylase

d. Unsaturated fatty acid degradation
fatty acyl-CoA reductase

e. Branch chain fatty acid degradation
branched-chain enoyl CoA reductase
& branched-chain enoyl CoA reductase
& 2-methyl branched-chain enoyl CoA reductase

f. Ketone body metabolism
3-OXOACID COA-TRANSFERASE
hydroxybutyrate dehydrogenase
ACETOACETYL-COA THIOLASE-acetyl-coA->acetoacyl-coA
& ACETOACETYL-COA THIOLASE
SUCCINYL-COA:3-KETOACID-COENZYME A TRANSFERASE-acetoacetate->acetoacyl-coA
& SUCCINYL-COA:3-KETOACID-COENZYME A TRANSFERASE

13. Metabolism of other energy sources

a. alcohol dehydrogenases
FORMALDEHYDE DEHYDROGENASE-long chain primary alcohol->aldehyde or ketone
& FORMALDEHYDE DEHYDROGENASE
aldehyde reductase
& aldehyde reductase

b. GLYCEROL
glycerol
glycerol-3-phosphatase
glycerol-3-phosphate dehydrogenase

c. propionate
PRPD PROTEIN

d. Other

diaminobutyrate decarboxylase-dia-butyrate to dia-propane

&diaminobutyrate decarboxylase

ACETAMIDASE-allows acetamide and formamide as sole C or N source

&ACETAMIDASE

&amidase

formamidase

acetate

ALDEHYDE DEHYDROGENASE-broad substrate specificity

&ALDEHYDE DEHYDROGENASE

14. Electron transport

a. Complex I-NADH-ubiquinone

NADH dehydrogenase

&ubiquinone

b. Complex II-Succinate-ubiquinone

succinate dehydrogenase

c. Complex III-Ubiquinone to cytochrome C

cytochrome b

cytochrome c

cytochrome oxidase

CBP4 PROTEIN-cytoC, ubiquinol assembly

&CBP4 PROTEIN

d. Other electron transport pathways

NADH OXIDASE

NADPH DEHYDROGENASE

f. Electron carriers

flavoprotein

FLAVOHEMOPROTEIN

QUINONE

g. Component enzymes and molecules

FLAVIN OXIDOREDUCTASE

&HYDROXYPHENYLPYRUVATE DIOXYGENASE

RESPIRATORY complex assembly

MITOCHONDRIAL FAD CARRIER PROTEIN

h. ATP synthase

ATP SYNTHASE

PLASMA MEMBRANE ATPASE (PROTON PUMP)

&H⁺-transporting ATPase

&H⁺-transporting ATPase

&V-ATPase

&p-ATPase

i. Alternative respiratory path

ALTERNATIVE OXIDASE

15.Reducing carriers

a.glutaredoxin
&glutaredoxin

b.gluathione
gamma-glutamyl transpeptidase-synthesis and deg of glutathione
&gamma-glutamyl transpeptidase
glutathione S-transferase-reduces peroxides, reducing agent
&glutathione S-transferase

c.thioredoxin
&thioredoxin

II. Cell Growth, Cell Division

A. Cell walls

septin
RODLET PROTEIN-spore-wall fungal hydrophobin
&RODLET PROTEIN
SPORE-WALL FUNGAL HYDROPHOBIN-not rodlet
&SPORE-WALL FUNGAL HYDROPHOBIN
glucanoyltransferase-elongation of cell wall beta(1-3)glucan
&glucanoyltransferase
INTEGRIN ALPHA CHAIN-LIKE PROTEIN-cell adhesion
&INTEGRIN ALPHA CHAIN-LIKE PROTEIN

B. Biomembranes

phosphatidylethanolamine methyltransferase
ACETYLMURAMIDASE-6-PHOSPHATE DEACETYLASE
DIACETYLMURAMIDASE
Glycosyltransferase-glycopeptidolipid biosyn
&Glycosyltransferase

C. Cytoskeleton, organelle biogenesis

peroxisom
KINESIN
tubulin
ankyrin
VACUOLAR ASSEMBLY PROTEIN VPS39

1. Actin-see also mitosis

#actin
PROFILIN-assembly of actin monomers
&PROFILIN
ARP2/3 COMPLEX-actin polymerization
&ARP2/3 COMPLEX
&p21-Arc
fimbrin-actin bundling
&fimbrin
COFILIN-actin binding
&cofilin
ACTIN-BINDING PROTEIN

2.Choline

choline dehydrogenase
choline kinase
chitin synthase

3. Other

phosphatidylethanolamine methyltransferase
ACETYLGLUCOSAMINE-6-PHOSPHATE DEACETYLASE
DIACETYLMURAMIDASE
Glycosyltransferase-glycopeptidolipid biosyn
&Glycosyltransferase

D. cell cycle control

cell division protein
CELL DIVISION CONTROL PROTEIN
cell cycle protein
SCH9 protein-cell progress through G1
&SCH9 protein
G1/S-SPECIFIC CYCLIN-essential for movement from g1-S
&G1/S-SPECIFIC CYCLIN
cullin-neg regulator of cell cycle
&cullin
&cell cycle control
SENESCENCE
apoptosis
&DEFENDER AGAINST CELL DEATH
&DAD-1

E. Mitosis/cytokinesis

1. MITOSIS

MITOSIS
DNA DAMAGE CHECKPOINT PROTEIN-allows entry into Mitosis
&DNA DAMAGE CHECKPOINT PROTEIN
centromere
CHROMOSOME SEGREGATION PROTEIN-with microtubule, migration of
chromosomes
&CHROMOSOME SEGREGATION PROTEIN
dynamin-molecular motor, associated with microtubule, endocytosis
&dynamin
dynein-molecular motor
&dynein
nuclear positioning
&apsB
DMR-N9 PROTEIN-regulation of mitosis
&DMR-N9 PROTEIN
CALTRACTIN-ASSOCIATED WITH THE POLES OF THE MITOTIC SPINDLES
&CALTRACTIN

2. Cytokinesis

cytokinesis
f-actin-contractile ring during cytokinesis
&f-actin
TROPOMYOSIN-component of contractile ring

&TROPOMYOSIN

F. Meiosis

Rad9-required for meiotic chromosome condensation and synapsis

&Rad9

condensin-chromosome condensation protein

&condensin

III. DNA synthesis

A. DNA replication

DNA POLYMERASE

replication factor

Single-stranded DNA-binding protein

&SSB

&SINGLE-STRANDED DNA-BINDING PROTEIN

DnaJ protein

B. DNA modification and repair

DNA LYASE

endonuclease IV

DNA REPAIR PROTEIN

DNA methyltransferase

cytosine C5-DNA methyltransferase

C. DNA packaging

1. Histone

Histones, class H1 (or I, or f1)

Histones, class H2a (or IIb1, or f2a2)

Histones, class H2b (or IIb2, or f2b)

Histones, class H3 (or III, or f3)

Histones, class H4 (or IV, or f2a1)

histone

2. DNA-binding

DNA-binding protein amdA

IV. Gene Expression

A. Transcription

1. RNA Polymerase

RNA POLYMERASE I, rRNA

RNA POLYMERASE II, mRNA

RNA POLYMERASE III, tRNA

RNA POLYMERASE

2. Regulation

transcription factor

SUPPRESSOR OF STEM-LOOP PROTEIN-transcription initiation, pol binding

&SUPPRESSOR OF STEM-LOOP PROTEIN

&SSL1

TRANSCRIPTION INITIATION FACTOR TFIID

TRANSCRIPTION INITIATION PROTEIN SPT6

RNA helicase

HELICASE MOT1-essential
&HELICASE MOT1
regulatory protein creA-carbon catabolite repression
&creA
HAC1-unfolded protein response pathway, transcrip activation
&HAC1
TRANSCRIPTIONAL REPRESSOR
QutR protein-repressor protein in the quinic acid utilization pathway, cluster
&QutR protein
QUINATE REPRESSOR
PacC-factor regulates acid and base expressed genes
&PacC

-Asexual development-Central regulatory pathway

regulatory protein brlA-transcription factor
®ulatory protein brlA
REGULATORY PROTEIN WETA
STUA transcription factor
CELL PATTERN FORMATION-ASSOCIATED PROTEIN: SPATIAL
LOCALIZATION OF ABAA AND BRLA
&CELL PATTERN FORMATION-ASSOCIATED PROTEIN

3. Processing

a. SPLICEOSOME

SPLICEOSOME ASSOCIATED PROTEIN
splicing factor
small nuclear ribonucleoprotein
U4/U6 SNRNA-ASSOCIATED SPLICING FACTOR PRP24
SPLICING FACTOR U2AF 65 KD SUBUNIT
MITOCHONDRIAL RNA SPLICING PROTEIN
&MRS2 protein
tRNA splicing endonuclease
HETEROGENEOUS NUCLEAR RIBONUCLEOPROTEIN K

b. polyA addition

POLYA

c. 5' capping

MRNA CAPPING ENZYME

d. Other

nucleolin-rRNA processing
&nucleolin
3'-TERMINAL PHOSPHATE CYCLASE-RNA processing?
&3'-TERMINAL PHOSPHATE CYCLASE
FIBRILLARIN
MINOR CAPSID PROTEIN C
QUEUINE TRNA-RIBOSYLTRANSFERASE-tRNA, guanine modification
&QUEUINE TRNA-RIBOSYLTRANSFERASE
&Trna-Guanine Transglycosylase

4. tRNA synthetase

&trna synthetase
&tRNA ligase

5. Degradation

ribonuclease

NONSENSE-MEDIATED MRNA DECA Y PROTEIN-DECA Y OF MRNAS
CONTAINING PREMATURE STOP CO

DONS

&NONSENSE-MEDIATED MRNA DECA Y PROTEIN

&NMD3 protein

B. Protein Biosynthesis

1. Initiation

EUKARYOTIC TRANSLATION INITIATION
INITIATION FACTOR

2. Elongation

ELONGATION FACTOR, eucaryotic and archaeal
&ELONGATION FACTOR

EF-TU

elongation factor 2

TRANSLATION FACTOR

3. Termination

PEPTIDE CHAIN RELEASE FACTOR

4. Ribosomal proteins

a. 40S ribosomal protein

&40S ribosomal protein

b. 60S ribosomal protein

&60S ribosomal protein

ribosomal protein

5. Post-translational modifications

a. methylation

SERINE HYDROXY METHYLTRANSFERASE

SERINE METHYLASE

b. glycosylation

glycosylation

GPI-ANCHOR TRANSMIDASE

MNN9 PROTEIN

GUANOSINE-DIPHOSPHATASE-glycosylation

&GUANOSINE-DIPHOSPHATASE

UDP-GLUCOSE:GLYCOPROTEIN GLUCOSYLTRANSFERASE

c. myristoylization

PEPTIDE N-MYRISTOYLTRANSFERASE

&GLYCYLPEPTIDEN-TETRADECANOYLTRANSFERAS

d. other

protein disulfide-isomerase

cyclophilin

6. Folding and Targeting

a. folding

CALNEXIN HOMOLOG-folding of glycoproteins
&CALNEXIN HOMOLOG

b.chaperones

chaperone

&chaperonin

prefoldin-chaperone which delivers unfolded proteins to another chaperonin

&prefoldin

heat shock protein Hsp88

heat-shock protein30

&30 KD HEAT SHOCK PROTEIN

&HSP30

heat shock protein 70

&DNAK Protein

&HEAT SHOCK 70

HEAT SHOCK PROTEIN HSP1

HEAT SHOCK PROTEIN 104

T-COMPLEX PROTEIN-chaperone of actin, tubulin

&T-COMPLEX PROTEIN

complex I intermediate associated protein-chaperone in assembly of NADH: Ubiquinone Oxidoreductase

&complex I intermediate associated protein

c.protein sorting

protein sorting

CARBOXYPEPTIDASE Y-sorting of vacuolar protein

&CARBOXYPEPTIDASE Y

MVP1 PROTEIN-vacuolar protein sorting

&MVP1 PROTEIN

vacuolar protein sorting homolog h-vps45

clathrin

RIBOSYLATION FACTOR

SIGNAL RECOGNITION PARTICLE

MITOCHONDRIAL IMPORT RECEPTOR

synaptobrevin-protein trafficking

&synaptobrevin

VESICLE TRANSPORT V-SNARE PROTEIN

COATOMER ALPHA SUBUNIT-trafficking to golgi, nonclathrin vesicles

&COATOMER ALPHA SUBUNIT

COATOMER BETA SUBUNIT-trafficking to golgi, nonclathrin vesicles

&COATOMER BETA SUBUNIT

&beta COP

&BETA-COAT PROTEIN

&beta prime coatomer protein

COATOMER ZETA SUBUNIT-trafficking to golgi, nonclathrin vesicles

&COATOMER ZETA SUBUNIT

NPL6 PROTEIN-nuclear protein localization

&NPL6 PROTEIN

7.Turnover-protein degradation-including vacuolar

protein-L-isoaspartate O-methyltransferase-esterification for degradation
 &protein-L-isoaspartate O-methyltransferase
 &PROTEIN-BETA-ASPARTATE METHYLTRANSFERASE
 PROTEASE REGULATORY SUBUNIT
 proteasome
 ubiquitin
 UBIQUITIN-CONJUGATING ENZYME
 &ubiquitin-protein ligase
 pepsinogen
 aspartic protease
 &aspartyl protease
 &ASPARTIC PROTEINASE
 proline peptidase
 aminopeptidase
 iminopeptidase
 serine protease
 ASPARTATE PROTEASE
 metallopeptidase
 &metalloproteinase
 NEUTRAL PROTEASE II
 ca dependent protease
 alkaline protease
 &ALKALINE PROTEINASE
 ACID PROTEASE A
 &ASPERGILLOPEPSIN II
 CAAX PRENYL PROTEASE-cleavage of alpha factor for activation
 &CAAX PRENYL PROTEASE
 insulinase-peptidase M16 family
 &insulinase
 &INSULIN-DEGRADING ENZYME
 &ZINC-PROTEASE C2E11.12C
 #Lon serine protease
 &MITOCHONDRIAL ATP-DEPENDENT PROTEASE

V. Processes

A. Cell rescue, defense, osmotic adaptation, starvation response, development

(asexual, sexual)(includes antibiotics, toxins)see also B.cell signalling, signal transduction and C. transmembrane transport

1. Development

-asexual-conidiation

velvet A

CONIDIATION-SPECIFIC PROTEIN

SpoC1-C1C protein

-asexual-pigment production

GREEN PIGMENT SYNTHASE

PORPHYRIN

porphyrinogen oxidase

polyketide synthase

LACCASE

&LACCASE
&diphenol oxidase

2. Defense

clavulanate-inhibits beta-lactamases

&clavulanate

VEGETABLE INCOMPATIBILITY PROTEIN-vegetative incompatibility,intra-species

&VEGETABLE INCOMPATIBILITY PROTEIN

&VEGETABLE INCOMPATIBILITY PROTEIN HET-E-1

pisatin demethylase-inactivates plant compound pisatin

&pisatin demethylase

D-AMINO ACID OXIDASE-oxidation of cephalosporin C

&D-AMINO ACID OXIDASE

-Secondary metabolites

penicill

-sterigmatocystin biosynthesis

sterigmatocystin

&sterigmatocystin

&norsolorinic acid reductase

&(U34740)

3. Detoxification

oxygen resistance

SHO1 OSMOSENSOR

catalase

super oxide dismutase

&superoxide dismutase

peroxidase

epoxide hydrolase+ water=glycol

&epoxide hydrolase

4. Salt tolerance

HALOTOLERANCE PROTEIN HAL2

&3'(2'),5-diphosphonucleoside 3'(2') phosphohydrolase

5. Starvation response

MAK16 PROTEIN-moves cytoplasmic proteins to vacuole-autophagocytosis

&MAK16 PROTEIN

B. Cell signalling, signal transduction

1. Kinases and second messengers

a. PHOSPHATASES

PROTEIN PHOSPHATASE

ca dependent protein phosphatase

PROTEIN-TYROSINE PHOSPHATASE

serine/threonine phosphatase

phosphatase regulator

b. Kinases

protein kinase

protein kinase C

cAMP-dependent protein kinase
MAP kinase
MAP KINASE KINASE 1
MAP KINASE HOG1
CALMODULIN-DEPENDENT PROTEIN KINASE
CAMP-DEPENDENT PROTEIN KINASE
SERINE/THREONINE-PROTEIN KINASE, casein kinase acts on acidic proteins
&SERINE/THREONINE-PROTEIN KINASE
&serine/threonine protein kinase
&casein kinase
serine/threonine-specific protein kinase KIN2
&PROTEIN KINASE KIN2
histidine kinase
&histidine kinase
nitrogen permease reactivator-on for nitrogenous transport systems/s-t kinase
&nitrogen permease reactivator

c. cAMP

adenyl cyclase
ADENYLYL CYCLASE-ASSOCIATED PROTEIN-CAP protein, binds cAMP to allow activation
&ADENYLYL CYCLASE-ASSOCIATED PROTEIN

2. G proteins

GTP-binding protein
GUANINE NUCLEOTIDE-BINDING PROTEIN
RHO1 GDP-GTP EXCHANGE PROTEIN:GUANINE-NUCLEOTIDE RELEASING FACTOR
&RHO1 GDP-GTP EXCHANGE PROTEIN
rho-gdp dissociation inhibitor-prevents cycling of GDP with GTP of rho protein family
&rho-gdp dissociation inhibitor
GTPASE-ACTIVATING PROTEIN-neg regulator of Ras1, play antagonistic role with rho
-gdp dissociation inhibitor
>PASE-ACTIVATING PROTEIN

C. Transmembrane transport

1. Secretion

secretion
&SEC10
KEXIN-proteinase secretion
&KEXIN

2. Exoenzymes

exoenzyme
dipeptidyl peptidase-exoenzyme
&dipeptidyl peptidase

3. Transport

a. sugar transport

sugar transport
GLUCOSE TRANSPORTER

GALACTOSE TRANSPORTER
inositol transport

b. multidrug resistance

multidrug resistance
multidrug transporter
oxytetracycline exporter
cycloheximide resistance protein

c. nuclear membrane

nuclear pore membrane protein
NUCLEAR TRANSPORT FACTOR 2

d. cation transport-ATPase, or major facilitator superfamily

cation transport
CATION-TRANSPORTING ATPASE
&cation transport-ATPase
CALCIUM-TRANSPORTING ATPASE
&Ca²⁺-transporting ATPase
sodium transport
sulfate transporter
cobalt transporter
COPPER TRANSPORT
zinc cadmium resistance
manganese resistance

e. Anion transport

arsenite translocating ATPase-anion transport, resistance to arsenite, antimonite, arsenate
&arsenite translocating ATPase
phosphate transporter
&phosphate transport
tartrate transport
CHOLINE TRANSPORT
allantoate transport
&ALLANTOATE PERMEASE

f. Protein and amino acid

PROTEIN TRANSPORT PROTEIN
PEPTIDE TRANSPORTER
&PEPTIDE PERMEASE
AMINO ACID TRANSPORTER
AMINO-ACID PERMEASE
ARGININE PERMEASE

g. mitochondrial transport

MITOCHONDRIAL PROTEIN IMPORT PROTEIN 2
MITOCHONDRIAL IMPORT RECEPTOR SUBUNIT TOM22-translocation of
cytosolic proteins
into mitochondria
&MITOCHONDRIAL IMPORT RECEPTOR SUBUNIT TOM22
mitochondrial transport protein amc-1
MITOCHONDRIAL 2-OXOGLUTARATE/MALATE CARRIER
&2-oxoglutarate/malate translocator
benzodiazepine receptor-TRANSPORT OF PORPHYRINS AND HEME, mitochondria

&benzodiazepine receptor
ADP,ATP CARRIER PROTEIN

h. ABC transporter family
ATP-DEPENDENT PERMEASE
ABC transporter
&ABC1 transporter
&ABC-type ATPase
&ATP-DEPENDENT TRANSPORTER

i. Other
transport protein
aquaporin

D. Classes of Enzymes-not in defined pathways (Reily,1997)

1. Oxidoreductases
OXIDOREDUCTASE
MONOOXYGENASE
cytochrome P450
&cytochrome p450
CR(VI) REDUCTASE-FLAVIN OXIDOREDUCTASE family
&CR(VI) REDUCTASE
AMINE OXIDASE-OXIDATIVE DEAMINATION of AMINES
&AMINE OXIDASE
&COPPER AMINE OXIDASE
SEXUAL DIFFERENTIATION PROCESS PROTEIN-expressed during sexual diff in
S. pombe
&SEXUAL DIFFERENTIATION PROCESS PROTEIN
fructosyl amine:oxygen oxidoreductase
&fructosyl amino acid oxidase
chlorocatechol 1,2-dioxygenase-degradation
&chlorocatechol 1,2-dioxygenase
cytochrome-P450
&trichothecene biosynthesis
&trichodiene oxygenase 4
&isotrichodermin C-15 hydroxylase
HYDROXY ACID DEHYDROGENASE

2. Transferases
ORNITHINE AMINOTRANSFERASE

3. Hydrolases
alkaline phosphatase

4. Lyases

5. Isomerases

6. Ligases

7. Synthetases
S-adenosylmethionine synthetase

E. Non-enzymatic classes (not in defined pathways)

1. Zinc finger motif-DNA binding

zinc finger protein
&zinc-finger protein
KIN17 protein-crossreacts w recA antibody
&KIN17 protein
&protein KIN17

2. Leucine zipper motif

leucine zipper

VI. Unclassified (significant homolog but function uncertain in *Aspergillus nidulans*)

uncertain function
&unclassified
bacteriorhodopsin
LEUKOTRIENE biosynthesis
&LEUKOTRIENE A-4HYDROLASE
bleomycin hydrolase
&BLMHYDROLASE
transposase
&transposon
prohibitin
DIPHTHINE SYNTHASE
&morphine dehydrogenase
STRESS PROTEIN
PEPTIDASE
regulatory protein
short chain dehydrogenase

VII. Unidentified (includes significant match with ORFs)

unknown function
&unclear function
&hypothetical protein

VIII. No significant homolog

NONE
-Contigs:949
-Singlets:1419

Appendix II. *Aspergillus nidulans* Unigene database in the categories of cellular functions. Members in each category are listed in order of homology significance. Numbers in parentheses indicate the number of Unigene members belonging to a particular category, a gene represented by both 3' and 5' Unigene members is counted only once. Words in < > are the keywords used to identify functions from the Unigene database BlastX reports and can also be category headings. There are several categories with no members, these were left to indicate that the category was considered in preparation of the outline. Contigs beginning with a * indicate that placement in the categories was made on the basis of a homology that was not the highest match, generally done when the best homolog was an ORF with no assigned function. The first number is the homology score followed by the probability of error. The next two numbers are the Unigene member sequence homology endpoints. The next column contains the code indicating into which database the original sequence deposit was made with accession numbers specifically for that database (ex: sp|P48825|BGL1_ASPAC, in the SwissProt database with ac# BGL1_ASPAC and GenBank ac# P48825). The final column contains the description of the homolog, organism if available, and may also list the GenBank accession number. The category, Classes of Enzymes are for those enzymes not in a defined pathway and are grouped in general functions suggested by Riley (Riley, 1997). Also refer to Figure 14 and Table 12.

I. Bioenergetics and Metabolism

A. Metabolism of Carbohydrates (for glucose see energy)

1. Chitin metabolism (6)

<chitin>

Contig1860_a5f03a1.f1	2005	1.2e-206	175 1329	gnl PID d1013927	(D87063) chitinase [Emericella nidulans]
Contig33_v7h11a1.r1	635	1.3e-89	184 612	gnl PID d1013927	(D87063) chitinase [Emericella nidulans]
Contig558_o5d02a1.r1	662	2.5e-64	471 836	gnl PID d1013927	(D87063) chitinase [Emericella nidulans]
Contig40_i8f07a1.r1	400	1.5e-36	300 530	gnl PID d1013927	(D87063) chitinase [Emericella nidulans]
Contig1503_c3g10a1.r1	221	6.8e-15	27 536	gnl PID e220269	(Z68924) Chitinase [Clostridium thermocellum]
Contig685_u4c02a1.f1	127	8.8e-07	161 232	gnl PID d1013927	(D87063) chitinase [Emericella nidulans]

2. Cellulose degradation (17)

<beta glucosidase-breakdown of cellulose>

m7d01a1.r1	977	1.1e-97	7 678	sp P48825 BGL1_ASPAC	BETA-GLUCOSIDASE 1 PRECURSOR (GENTIOBIASE) (CELLOBIASE) (BETA-D-GLUCOSIDE GLUCOHYDROLASE) >
Contig1829_d5d08a1.f1	952	5e-95	5 1951	gi 3004863	(AF029354) exo-beta-1,3-glucanase [Amelomyces quisqualis]
m7d01a1.f1	700	2.4e-68	105 659	sp P48825 BGL1_ASPAC	BETA-GLUCOSIDASE 1 PRECURSOR (GENTIOBIASE) (CELLOBIASE) (BETA-D-GLUCOSIDE GLUCOHYDROLASE) >
Contig1715_e9e09a1.f1	651	3.8e-63	197 850	prf 1713235A	extracellular beta glucosidase [Trichoderma reesei]
*Contig1707_b0e09a1.f1	573	7.1e-55	375 1220	sp P15703 BGL2_Y	GLUCAN 1,3-BETA-GLUCOSIDASE PRECURSOR (EXO-1,3-BETA-GLUCANASE) (GP29) >pir A33499 beta-1,3-exoglucana
Contig7_e9e09a1.r1	274	1.9e-40	178 507	gi 493580	(U09580) beta-D-glucoside glucohydrolase [Trichoderma

				reesei]	
o5g11a1.r1	369	3.4e-33	8 436	gnl PID e218254	(X94986) beta glucosidase [Manihot esculenta]
t2h09a1.r1	317	5e-27	8 439	gi 534844	(U13672) beta-glucosidase [Candida wickerhamii]
t2h09a1.f1	307	6e-26	177 455	>prf 2107160Abeta-glucosidase [Candida wi gi 534844	(U13672) beta-glucosidase [Candida wickerhamii]
Contig608_c3g03a1.f1	288	1.1e-24	127 612	>prf 2107160Abeta-glucosidase [Candida wi sp P15703 BGL2_Y	GLUCAN 1,3-BETA-GLUCOSIDASE
Contig1738_c9c10a1.f1	123	6e-06	402 545	PRECURSOR(EXO-1,3-BETA-GLUCANASE) (GP29) >pir A33499 beta-1,3-exoglucana sp P49426 EXG1_C	GLUCAN 1,3-BETA-GLUCOSIDASE PRECURSOR (EXO-BETA 1,3GLUCANASE) (1,3-BETA-D-GLUCANOHYDROLASE) >gi 10665
<glucanase>					
Contig1263_m7b06a1.f1	316	1.2e-26	249 620	gi 3004863	(AF029354) exo-beta-1,3-glucanase [Ampelomyces quisqualis]
Contig974_r2f09a1.f1	277	1.6e-23	169 687	gi 2326188	(U81606) mixed-linked glucanase precursor [Cochliobolus carbonum]
Contig1017_c9g12a1.r1	255	3.6e-21	145 519	gi 2326188	(U81606) mixed-linked glucanase precursor [Cochliobolus carbonum]
Contig432_e0d11a1.r1	227	1.1e-17	310 606	gi 3152652	(AF064870) endo-1,3(4)-beta-glucanase [Phaffia rhodozyma]
Contig869_z2d08a1.f1	152	6.6e-08	579 863	sp P25358 GNS1_Y	GNS1 PROTEIN >pir 812916 probable membrane proteinYCR034w - yeast (Saccharomyces cerevisiae) >gi 449
<cellobiohydrolase>					
alg02f2.f1	387	3.6e-35	250 693	gi 912494	(U25129) cellobiohydrolase [Cochliobolus carbonum]
3. Pectin degradation					
<pectate>					
4. Cutin metabolism (3)					
<cutin>					
Contig1651_j9c08a1.f1	633	3e-61	127 711	sp P52956 CUTI_A	CUTINASE PRECURSOR (L1) >gnl PID d1008007 (D38311)Cutinase [Aspergillus oryzae]
Contig291_g7h01a1.r1	402	9.1e-37	6 443	gi 1438949	(U61841) cutinase G-box binding protein [Fusarium solani f. sp.pisi]
h1c07a1.r1	268	2.3e-21	28 390	sp P52959 CT1B_FUSSO	CUTINASE TRANSCRIPTION FACTOR 1 BETA >gi 1262914 (U51672)cutinase transcription factor 1 [
5. Polysaccharide synthesis (1)					
<UDP-glucose dehydrogenase>					
*Contig602_c3h05a1.r1	188	2.2e-11	41 730	gi 3127129	(AF061017) UDP-glucose dehydrogenase [Mus musculus]
6. Energy reserve synthesis-see also energy reserve metabolism (10)					
<GLYCOGEN (STARCH) SYNTHASE>					
Contig1376_c6c08a1.r1	925	3.4e-92	23 871	sp P23337 UGS1_Y	GLYCOGEN (STARCH) SYNTHASE, ISOFORM 1

r5h03a1.r1	734	6.1e-72	168 794	>pir A38326UDPglucose--starch glucosyltransferase (EC 2.4.1.11 sp P49841 KG3B_HUMAN GLYCOGEN SYNTHASE KINASE-3 BETA (GSK-3 BETA) >pir S53324protein kinase - human >gi 529237,serine/threonine protein kinase
r5h03a1.f1	405	4.5e-37	381 776	sp P49841 KG3B_HUMAN GLYCOGEN SYNTHASE KINASE-3 BETA (GSK-3 BETA) >pir S53324protein kinase - human >gi 529237,serine/threonine protein kinase
Contig1713_c6c08a1.f1	273	4.4e-22	250 552	sp P27472 UGS2_Y GLYCOGEN (STARCH) SYNTHASE, ISOFORM 2 >pir S51396UDPglucose--starch glucosyltransferase (EC 2.4.1.11
w5c03a1.r1	213	1.2e-15	329 475	sp P23337 UGS1_YEAST GLYCOGEN (STARCH) SYNTHASE, ISOFORM 1 >pir A38326UDPglucose--starch glucosyltransferase (
<1,4-ALPHA-GLUCAN BRANCHING ENZYME>				
Contig1640_c5a02a1.f1	1633	3.3e-167	201 1571	sp P32775 QLGB_Y 1,4-ALPHA-GLUCAN BRANCHING ENZYME (GLYCOGEN BRANCHINGENZYME) >pir S50448 1,4-alpha-glucan branching
<starch branching enzyme>				
Contig1191_e9a06a1.r1	430	7.3e-39	119 640	gnl PID e1228556 (AJ000004) starch branching enzyme II, SBE-II [Solanumtuberosum]
<trehalose synthase>				
*Contig1396_l3f08a1.f1	831	3.1e-82	103 1026	gnl PID d1032303 (AB010104) trehalose synthase [Grifola frondosa]>gnl PID d1032304 (AB010105) trehalose synthase [Grif
l3f08a1.r1	394	3.9e-35	22 594	gnl PID d1032303 (AB010104) trehalose synthase [Grifola frondosa]>gnl PID d1032304 (AB010105) trehalose syn
Contig1266_m7e05a1.r1	158	7.9e-08	418 768	gnl PID d1032303 (AB010104) trehalose synthase [Grifola frondosa]>gnl PID d1032304 (AB010105) trehalose synthase [Grif
7.Arabinose metabolism (5)				
<arabin>				
Contig1764_e9h03a1.r1	805	1.6e-79	366 1277	sp P42256 ABNA_A ARABINAN ENDO-1,5-ALPHA-L-ARABINOSIDASE A PRECURSOR(ENDO-1,5-ALPHA-L-ARABINANASE A) (ABN A) >gi 44106
n8b03a1.f1	486	1.1e-45	144 521	gnl PID e257620 (Z78010) (1,4)-beta-D-arabinoxylan arabinofuranohydrolase[Aspergillus tubingensis]
Contig1676_c5f01a1.f1	264	6.5e-22	634 990	sp P50166 ARDH_C D-ARABINITOL 2-DEHYDROGENASE (RIBULOSE FORMING) (ARDH)>pir JC4041 D-arabinitol dehydrogenase (EC 1.1
r7f11a1.f1	254	4.4e-21	225 749	sp P43066 ARDH_CANAL D-ARABINITOL 2-DEHYDROGENASE (RIBULOSE FORMING) (ARDH)>gi 295568 (L16227) D-arabinitol deh
n8b03a1.r1	137	8.7e-06	413 568	gnl PID e257619 (Z78011) (1,4)-beta-D-arabinoxylan arabinofuranohydrolase[Aspergillus niger]
8.Glucosamine (4)				
<GLUCOSAMINE-6-PHOSPHATE ISOMERASE-GLUCOSAMINE UTILIZATION PATHWAY>				
i7c04a1.r1	365	7.5e-33	100 474	sp P09375 NAGB_ECOLI GLUCOSAMINE-6-PHOSPHATE ISOMERASE(GLUCOSAMINE-6-PHOSPHATE DEAMINASE) >pir MUECNG probable
<beta glucosamine>				

g0a07a1.f1 184 1.8e-11 29 244 gi|2731443 (U96923) [prot= cDNA of the glycoamidase gene
[Aspergillus niger]

<GLUCOSAMINIDASE-degradation of glycans>
Contig1463_n2f07a1.r1 716 4.4e-70 13 780 sp|P43077|HEX1_C BETA-HEXOSAMINIDASE
PRECURSOR(N-ACETYL-BETA-GLUCOSAMINIDASE)
(BETA-GLCNACASE) (BETA-N-ACETYLHEXOSAMINI

<GLUCOSAMINE--FRUCTOSE-6-PHOSPHATE AMINOTRANSFERASE>
Contig1420_c5a03a1.r1 1107 1.8e-111 25 1008 sp|P53704|GFA1_C GLUCOSAMINE--FRUCTOSE-6-PHOSPHATE AMINOTRANSFERASE
(ISOMERIZING) (HEXOSEPHOSPHATE AMINOTRANSFERASE)(D-

9.Aminosugar metabolism (1)

<PHOSPHOACETYLGLUCOSAMINE MUTASE>

o8f08a1.f1 353 1.6e-31 212 655 sp|Q09687|PCM1_SCHPO PUTATIVE PHOSPHOACETYLGLUCOSAMINE
MUTASE(ACETYLGLUCOSAMINE PHOSPHOMUTASE)=S. pombe

10.Sucrose metabolism (1)

<sucro>

<levanase-sucrose to glucose>

Contig581_c4h06a1.r1 323 8.8e-58 619 1014 prf||1404371A levanase [Bacillus subtilis]
Contig586_c4f06a1.f1 335 2.9e-29 87 431 gnl|PID|e1254710 (AJ000493) Sucrose:Sucrose
1-Fructosyltransferase[Aspergillus foetidus]

11.Galactose metabolism (2)

<galactose>

e0g08a1.f1 400 1.4e-36 109 483 sp|P08431|GAL7_YEAST GALACTOSE-1-PHOSPHATE URIDYLTRANSFERASE
>pir||XNBYUGUDPglucose--hexose-1-phosphate uridy

<alpha-1,4 polygalactosaminidase>

Contig387_f2f12a1.f1 160 8e-11 139 462 gnl|PID|d1004085 (D14846) endo alpha-1,4 polygalactosaminidase
precursor[Pseudomonas sp.]

12.Mannitol metabolism (14)

<manno>

Contig1218_f2g08a1.r1 1143 2.6e-115 40 801 gi|2407176 (AF016850) alpha-mannosidase [Emericella nidulans]
Contig1260_a5h12a1.f1 596 2.5e-57 81 635 gnl|PID|d1009247 (D49827) alpha-mannosidase [Aspergillus phoenicis]
r2h02a1.r1 518 4.6e-49 186 677 gnl|PID|e1287777 (AL022600) putative mannose-1-phosphate gaunyl
transferase[Schizosaccharomyces pombe]
Contig453_d4h02a1.f1 445 2.6e-41 66 587 gnl|PID|e1287777 (AL022600) putative mannose-1-phosphate gaunyl
transferase[Schizosaccharomyces pombe]
Contig243_h4a11a1.r1 436 1.2e-39 9 464 sp|P31382|PMT2_Y DOLICHYL-PHOSPHATE-MANNOSE--PROTEIN
MANNOSYLTRANSFERASE 2>pir||S36711 hypothetical protein YAL023 - y
Contig115_m5e09a1.r1 369 2.7e-33 46 783 gnl|PID|e1263907 (AL022103) mannose-6-phosphate isomerase
[Schizosaccharomyces pombe]
Contig1306_c9e06a1.f1 336 5.8e-29 99 683 gi|2245570 (AF005035) alpha 1,2-mannosidase [Spodoptera

e0f01a1.f1	289	3.3e-24	65 490	frugiperda] sp P53966 KTR5_YEAST PROBABLE MANNOSYLTRANSFERASE KTR5 >pir s62941 probablemembrane protein YNL029c - yeast (S
Contig1098_ale06c9.r1	283	1.3e-23	218 862	sp P31723 MA12_P MANNOSYL-OLIGOSACCHARIDE ALPHA-1,2-MANNOSIDASE PRECURSOR(MAN(9)-ALPHA-MANNOSIDASE) >pir s58766 manno
Contig244_h4a11a1.f1	151	6.6e-20	165 380	sp P31382 PMT2_Y DOLICHYL-PHOSPHATE-MANNOSE--PROTEIN MANNOSYLTRANSFERASE 2>pir s36711 hypothetical protein YAL023 - y
Contig1340_m5e09a1.f1	146	9.8e-07	177 431	gnl PID e1263907 (AL022103) mannose-6-phosphate isomerase [Schizosaccharomycespombe]
<mannitol>				
Contig1790_f0h09a1.f1	642	3.3e-62	137 1237	sp Q45421 MTLD_B MANNITOL-1-PHOSPHATE 5-DEHYDROGENASE >gi 1480431 (U18943)mannitol-1-phosphate dehydrogenase [Bacillus
*Contig1086_i3e06a1.r1	242	3.3e-19	414 884	gi 3128349 (AF010496) mannitol 2-dehydrogenase [Rhodobacter capsulatus]
Contig1601_a5b09a1.f1	161	1.5e-10	567 809	sp Q02418 MTLD_S MANNITOL-1-PHOSPHATE 5-DEHYDROGENASE >pir C44798mannitol-phosphate dehydrogenase MtlD - Streptococcu
13.Xylanose metabolism (5)				
<xylosidase>				
l0h06a1.f1	497	7e-47	192 479	pir JC5034 xylan endo-1,3-beta-xyloisidase (EC 3.2.1.32) - Emericella nidulans>gi 1050888 (Z49894) xyl
<xylanase>				
i2e05a1.r1	850	2.7e-84	109 600	pir s57397 xylanase - Emericella nidulans
Contig1167_h1e02a1.f1	206	2.1e-13	289 768	sp P54865 XYND_C ENDO-1,4-BETA-XYLANASE D PRECURSOR (XYLANASE D) (XYLD)>pir I40712 endo-1,4-beta-xylanase (EC 3.2.1.8
<xylitol dehydrogenase>				
Contig310_g6d05a1.f1	620	7.1e-60	84 770	gi 3264834 (AF072541) xylitol dehydrogenase; XDH [Galactocandidamastotermitis]
u4g02a1.r1	372	3e-49	37 369	gi 3264834 (AF072541) xylitol dehydrogenase; XDH [Galactocandidamastotermitis]
14.Quinate metabolism (2)				
<quinate-utilization is in cluster>				
o4h02a1.r1	872	1.3e-86	22 522	pir s11944 QUTG protein - Emericella nidulans
Contig394_flg10a1.r1	235	1.8e-17	8 397	sp P07547 ARO1_E PENTAFUNCTIONAL AROM POLYPEPTIDE (CONTAINS:3-DEHYDROQUINATE SYNTHASE , 3-DEHYDROQUINATE DEHYDRATASE(3
15.Sorbitol metabolism (4)				
<SORBITOL UTILIZATION PROTEIN>				
h1e12a1.f1	492	2.7e-46	11 565	sp P87218 SOU2_CANAL SORBITOL UTILIZATION PROTEIN SOU2 >gi 2183242 (AF002134)Sou2p [Candida albicans]
w7c03a1.r1	331	3e-29	36 509	sp P87219 SOU1_CANAL SORBITOL UTILIZATION PROTEIN SOU1 >gi 2183243 (AF002134)Sou1p [Candida albicans]

a0e03a1.f1 275 2.7e-23 174 476 sp|P87218|SOU2_CANAL SORBITOL UTILIZATION PROTEIN SOU2 >gi|2183242
(AF002134)Sou2p [Candida albicans]
a0e03a1.r1 250 1.2e-20 35 463 sp|P87218|SOU2_CANAL SORBITOL UTILIZATION PROTEIN SOU2 >gi|2183242
(AF002134)Sou2p [Candida albicans]
<SORBITOL DEHYDROGENASE>
i7c03a1.f1 185 7.4e-12 148 666 sp|P35497|DHSO_YEAST SORBITOL DEHYDROGENASE (L-IDITOL
2-DEHYDROGENASE)>pir||S55941 sorbitol dehydrogenase - yea

16. Gluconate (1)

<Glucose Oxidase-first step of glucose + O2 to gluconic acid>

f2f08a1.r1 401 1.5e-36 23 367 sp|P81156|GOX_PENAG GLUCOSE OXIDASE (GLUCOSE OXYHYDRASE)
(GOD)(BETA-D-GLUCOSE:OXYGEN 1-OXIDO-REDUCTASE)=Penicillium

17. Pyranose metabolism (1)

<pyranose oxidase>

y6e01a1.r1 366 2.3e-32 88 630 gnl|PID|d1011780 (D73369) pyranose oxidase [Coriolus versicolor]

18. Ribitol metabolism (1)

<ribitol kinase>

Contig831_r2d11a1.f1 541 1.8e-51 240 1055 gi|2905643 (AF045244) ribitol kinase [Klebsiella pneumoniae]

19. Calvin cycle (1)

<RIBULOSE-PHOSPHATE 3-EPIMERASE-ribulose-5 PO4 to xylulose-5 PO4>

w8f11a1.r1 330 3.7e-29 198 620 sp|P46969|RPE_YEAST RIBULOSE-PHOSPHATE 3-EPIMERASE
(PENTOSE-5-PHOSPHATE3-EPIMERASE) (PPE) (RPE) >pir||S51587 P

B. Metabolism of Amino acids and Related Molecules

1. alanine metabolism (0)

2. arginine metabolism(8)

a. arginine anabolism-glutamine, CO2 to arginine

<ORNITHINE CARBAMOYLTRANSFERASE>

h1a01a1.f1 753 5.7e-74 106 558 sp|P11803|OTC_EMENI ORNITHINE CARBAMOYLTRANSFERASE PRECURSOR
(OTCASE)(ORNITHINE TRANSCARBAMYLASE) >gi|168017 (

h1a01a1.r1 355 8.8e-32 37 297 pir||OWASN ornithine carbamoyltransferase (EC 2.1.3.3) precursor
- Emericellanidulans

<AGMATINASE>

m0g05a1.r1 271 7.2e-23 184 465 sp|Q10088|SPEB_SCHPO PUTATIVE AGMATINASE PRECURSOR (AGMATINE
UREOHYDROLASE)(AUH) >gi|1107898 (Z68166) unknown {

m0g05a1.f1 225 1.4e-17 125 418 sp|Q10088|SPEB_SCHPO PUTATIVE AGMATINASE PRECURSOR (AGMATINE
UREOHYDROLASE)(AUH) >gi|1107898 (Z68166) unknown {

b. arginine catabolism-arginine to proline

<ARGINASE-also see urea cycle>

Contig1156_x7b02a1.r1 174 6e-12 17 265 gn1|PID|e1250600 (AL021815) arginase family protein
[Schizosaccharomycespombe]

<ARG-6 PROTEIN>

i2h03a1.r1 667 7.3e-65 34 597 sp|P54898|AR56_NEUCR ARG-6 PROTEIN PRECURSOR
(CONTAINS:N-ACETYL-GAMMA-GLUTAMYL-PHOSPHATE REDUCTASE (N-ACETYL-GL
Contig1036_d1f07a1.f1 372 1.5e-32 223 486 sp|P54898|AR56_N ARG-6 PROTEIN PRECURSOR
(CONTAINS:N-ACETYL-GAMMA-GLUTAMYL-PHOSPHATE REDUCTASE
(N-ACETYL-GLUTAMATESEMI

<PYRROLINE-5-CARBOXYLATE REDUCTASE>

Contig905_y7b01a1.r1 268 1.4e-22 55 522 sp|P22008|PROC_P PYRROLINE-5-CARBOXYLATE REDUCTASE (P5CR) (P5C
REDUCTASE)>pir||JQ0418 pyrroline-5-carboxylate reductas
y7b01a1.f1 227 3.3e-18 59 379 sp|P22008|PROC_PSEAE PYRROLINE-5-CARBOXYLATE REDUCTASE (P5CR) (P5C
REDUCTASE)>pir||JQ0418 pyrroline-5-carboxyla

3.asparagine metabolism (0)**4.aspartic acid metabolism (3)**

-aspartate anabolism-oxaloacetate, glutamate to aspartate

<ASPARTATE AMINOTRANSFERASE>

g6c09a1.r1 640 5.2e-62 152 769 gi|1049345 (U39645) similar to aspartate aminotransferase
[Caenorhabditiselegans]
g5e09a1.r1 512 1.9e-48 4 561 gn1|PID|e1202255 (AL009197) hypothetical aspartate
aminotransferase[Schizosaccharomyces pombe]
g5e09a1.f1 305 1.8e-26 309 599 gn1|PID|e1202255 (AL009197) hypothetical aspartate
aminotransferase[Schizosaccharomyces pombe]

<Aspartase>

5.cysteine metabolism (2)

<O-ACETYLHOMOSERINESULFHYDRYLASE-also methionine biosyn>

Contig1712_g8g09a1.r1 2041 1.8e-210 27 1337 gi|2605905 (AF029318) O-acetyl-L-homoserine sulphydrylase
[Emericellanidulans]

<CYSTATHIONINE GAMMA-LYASE>

Contig1760_c6e02a1.f1 1228 2.8e-124 246 1391 sp|P31373|CYS3_Y CYSTATHIONINE GAMMA-LYASE
(GAMMA-CYSTATHIONASE)>pir||S31228 cystathionine gamma-lyase (EC 4.4.1.1) -

6.glutamic acid metabolism (0)**7.glutamine metabolism (2)**

<GLUTAMINE SYNTHETASE>

Contig1740_c6a12a1.rl 1383 1e-140 302 1336 sp|Q12613|GLNA_C GLUTAMINE SYNTHETASE (GLUTAMATE--AMMONIA
LIGASE)>gi|1322275 (L78067) glutamine synthetase [Glomerella
Contig398_m7a03a1.rl 558 2.7e-53 133 588 sp|Q12613|GLNA_C GLUTAMINE SYNTHETASE (GLUTAMATE--AMMONIA
LIGASE)>gi|1322275 (L78067) glutamine synthetase [Glomerella

8.glycine metabolism (5)

a. glycolate to glycine

<GLYCERATE DEHYDROGENASE>

Contig1128_m5d12a1.rl 376 5.3e-34 133 540 sp|P40054|SERX_Y PUTATIVE D-3-PHOSPHOGLYCERATE DEHYDROGENASE
YER081W(PGDH) >pir||S50584 hypothetical protein YER081w -
d2e07a1.rl 125 2.3e-06 9 233 sp|O29445|SERA_ARCFU D-3-PHOSPHOGLYCERATE DEHYDROGENASE (PGDH)
>gi|2649798(AE001048) phosphoglycerate dehydroge

b. glycine catabolism

-glycine decarboxylase complex-made up of P,T,L,H-removes amino group

<GLYCINE CLEAVAGE SYSTEM H PROTEIN>

b0e07a1.f1 282 4.2e-24 112 438 gn1|PID|e1184358 (Z99120) similar to glycine cleavage system protein
H[Bacillus subtilis]

<Glycine cleavage system T protein>

f1d07a1.rl 234 1.4e-18 233 448 sp|P48015|GCST_YEAST AMINOMETHYLTRANSFERASE PRECURSOR (GLYCINE CLEAVAGE
SYSTEMT PROTEIN) >pir||S54642 glycine c
f1d07a1.f1 184 3e-11 200 460 gn1|PID|e339946 (Z98979) aminomethyltransferase precursor
[Schizosaccharomycespombe]

9.histidine metabolism (1)

<HISTIDINE BIOSYNTHESIS>

o5a04a1.rl 365 1.4e-32 73 525 sp|P33734|HIS5_YEAST HISTIDINE BIOSYNTHESIS BIFUNCTIONAL AMIDOTRANSFERASE
/CYCLASE >pir||S46125 amidotransferas

10.isoleucine metabolism (3)

<2,3-DIHYDROXYACID HYDROLYASE-4th step in iso & val biosyn>

Contig760_z5f11a1.f1 552 1.2e-52 76 603 sp|P39522|ILV3_Y DIHYDROXY-ACID DEHYDRATASE PRECURSOR (DAD)
(2,3-DIHYDROXYACID HYDROLYASE) >pir||S55205 dihydroxy-acid

-catabolism

<propionyl-CoA carboxylase-also leucine and valine degradation>

nlc10a1.rl 588 1.7e-56 25 552 gn1|PID|e290075 (Y07660) B subunit of propionyl-CoA carboxylase
[Mycobacteriumtuberculosis]

<methylcrotonyl-CoA carboxylase>

*Contig665_alh06c9.rl 273 4.4e-22 222 560 gi|533707 (U12536) 3-methylcrotonyl-CoA carboxylase precursor
[Arabidopsisthaliana]

11.leucine metabolism (2)

<hydroxy-3-methylglutaryl-CoA lyase-FINAL STEP OF KETOGENESIS AND LEUCINE CATABOLISM>
w8e12a1.r1 203 1.8e-25 162 422 sp|P13703|HMGL_PSEMV HYDROXYMETHYLGLUTARYL-COA LYASE (HMG-COA LYASE)
(HL)(3-HYDROXY-3-METHYLGLUTARATE-COA LYASE
w8e12a1.fl 275 2.7e-23 110 469 sp|P13703|HMGL_PSEMV HYDROXYMETHYLGLUTARYL-COA LYASE (HMG-COA LYASE)
(HL)(3-HYDROXY-3-METHYLGLUTARATE-COA LYASE

12. Lysine metabolism (2)

<HOMOCITRATE DEHYDRATASE>

z8g05a1.fl 215 5.9e-17 175 516 sp|P40202|LYS7_YEAST HOMOCITRATE DEHYDRATASE >pir||s50245 LYS7 protein -
yeast(Saccharomyces cerevisiae) >gi|59

<SACCHAROPINE DEHYDROGENASE>

w5f07a1.r1 306 1.4e-26 26 271 sp|P38997|LYS1_YARLI SACCHAROPINE DEHYDROGENASE (NAD+, L-LYSINE
FORMING)(LYSINE--2-OXOGLUTARATE REDUCTASE) (SDH

13. methionine metabolism(5)

<HOMOSERINE O-ACETYLTRANSFERASE>

Contig185_i7e07a1.fl 362 1.6e-32 400 783 sp|P12917|MET2_A HOMOSERINE O-ACETYLTRANSFERASE
(HOMOSERINEO-TRANS-ACETYLASE) >pir||XYIMHA homoserine O-acetyltransfer

<cystathionine beta-lyase-3rd step>

g5d05a1.r1 749 1.6e-73 166 702 gi|1399263 (U28383) cystathionine beta-lyase [Emericella
nidulans]

<methionine synthase-last step in met biosynthesis>

gla07a1.fl 680 2.9e-66 8 766 gnl|PID|d1014526 (D89167) similar to Saccharomyces cerevisiae
5-METHYLTETRAHYDROPTEROYLTRIGLUTAMATE--HOMOCYSTEINEMETHYLTRANSFERASE

g7c01a1.r1 487 2.8e-45 188 637 gi|609350 (U15099) methionine synthase [Saccharomyces
cerevisiae]

Contig712_t2d10a1.fl 417 1.3e-37 358 753 gi|2738248 (U97200) cobalamin-independent methionine synthase
[Arabidopsisthaliana]

14. phenylalanine metabolism (0)

15. proline metabolism (0)

16. serine metabolism (2)

<PHOSPHOSERINE AMINOTRANSFERASE>

Contig190_j9e07a1.fl 211 5.3e-16 137 442 sp|P33330|SERC_Y PHOSPHOSERINE AMINOTRANSFERASE >pir||s42680
phosphoserinetransaminase (EC 2.6.1.52) - yeast (Saccharo

Contig128_j9e07a1.r1 200 8.1e-15 21 308 sp|P33330|SERC_Y PHOSPHOSERINE AMINOTRANSFERASE >pir||s42680
phosphoserinetransaminase (EC 2.6.1.52) - yeast (Saccharo

17. threonine metabolism (0)

18. tryptophan metabolism (2)

<anthranilate phosphoribosyltransferase-2nd step in tryp biosyn>

f5b03a1.f1 133 5.3e-06 115 291 gnl|PID|e1292700 (AL023554) anthranilate
phosphoribosyltransferase[Schizosaccharomyces pombe]
<CATECHOL 1,2-DIOXYGENASE-tryp & lysine catabolism in KETOADIPATE PATHWAY>
u4d08a1.f1 268 1.4e-22 316 612 gnl|PID|d1013794 (D86544) hydroxyquinol-1, 2-dioxygenase [Ralstonia
pickettii]

19.tyrosine metabolism (2)
<prephenate dehydrogenase>

Contig920_c5c08a1.r1 525 7.5e-87 466 987 gnl|PID|e1295792 (AL023776) prephenate dehydrogenase
[Schizosaccharomycespombe]
Contig1454_w8d03a1.f1 200 1.2e-14 310 642 gnl|PID|e1295792 (AL023776) prephenate dehydrogenase
[Schizosaccharomycespombe]

20.valine metabolism (3)

<hydroxyisobutyrate dehydrogenase>

Contig1468_r5h09a1.f1 474 2.2e-44 356 1117 sp|P29266|D3HI_R 3-HYDROXYISOBUTYRATE DEHYDROGENASE PRECURSOR
(HIBADH)>pir|A32867 3-hydroxyisobutyrate dehydrogenase
*Contig16_e4a03a1.r1 130 2.4e-05 88 717 sp|P28811|MMSB_P 3-HYDROXYISOBUTYRATE DEHYDROGENASE (HIBADH)
>pir|C429023-hydroxyisobutyrate dehydrogenase (EC 1.1.1.

<METHYLMALONATE-SEMIALDEHYDE DEHYD>

*Contig100_k9d06a1.f1 249 4.1e-20 224 469 sp|Q02252|MMSA_H METHYLMALONATE-SEMIALDEHYDE DEHYDROGENASE
(ACYLATING)(MMSDH) >gi|188696 (M93405) methylmalonate semia

21.aromatic amino acid metabolism (2)

<PHOSPHO-2-DEHYDRO-3-DEOXYHEPTONATE ALDOLASE>

w5e04a1.r1 618 1.1e-59 8 481 sp|P79023|AROG_CANAL PHOSPHO-2-DEHYDRO-3-DEOXYHEPTONATE
ALDOLASE, TYROSINE-INHIBITED (PHOSPHO-2-KETO-3-DEOXYHEPT
f0b04a1.r1 435 2.9e-40 155 592 sp|P34725|AROF_CANAL PHOSPHO-2-DEHYDRO-3-DEOXYHEPTONATE
ALDOLASE, PHENYLALANINE-INHIBITED (PHOSPHO-2-KETO-3-DEOX

22.polyamine biosynthesis(3)

<polyamine>

Contig1761_clh02a1.f1 574 4.9e-55 275 1102 gnl|PID|e1286476 (AJ002204) polyamine oxidase [Zea mays]
h4a05a1.r1 458 1e-42 13 450 sp|Q12074|SPEE_YEAST SPERMIDINE SYNTHASE (PUTRESCINE
AMINOPROPYLTRANSFERASE)(SPDSY) >pir|s54090 SPE3 protein -
hlh03a1.r1 125 0.00047 135 401 gnl|PID|e1286476 (AJ002204) polyamine oxidase [Zea mays]

C. Metabolism of Nucleotides and Nucleic Acids, Purines, Pyrimidines

1. Nucleotide metabolism (5)

<NUCLEOSIDE DIPHOSPHATE KINASE>

Contig1402_m3d02a1.r1 557 2.8e-53 122 565 sp|P19804|NDKB_R NUCLEOSIDE DIPHOSPHATE KINASE B (NDK B) (NDP KINASE

B)(P18) >pir||A38369 nucleoside-diphosphate kinas
 <ribose-phosphate pyrophosphokinase-purine, pyrimidine biosyn, also his and tryptophan biosyn>
 t2f05a1.f1 469 7.4e-44 83 478 sp|P32895|KPR1_YEAST RIBOSE-PHOSPHATE PYROPHOSPHOKINASE 1
 (PHOSPHORIBOSYLPYROPHOSPHATE SYNTHETASE 1) >pir||s305
 z8e10a1.f1 419 1.5e-38 149 532 pir||s60393 ribose-phosphate pyrophosphokinase (EC 2.7.6.1) PRS1 -
 yeast(Candida albicans)
 f0e10a1.r1 401 1.2e-36 13 618 pir||s61716 ribose-phosphate pyrophosphokinase PRPS3 homolog
 YOL061w - yeast(Saccharomyces cerevisiae)
 t2f05a1.r1 154 1.1e-09 206 433 sp|P32895|KPR1_YEAST RIBOSE-PHOSPHATE PYROPHOSPHOKINASE 1
 (PHOSPHORIBOSYLPYROPHOSPHATE SYNTHETASE 1) >pir||s305

2. Purine metabolism (12)

a. inosine mono phosphate de novo biosynthesis

<amidophosphoribosyl transferase-1st step>

Contig382_f5e07a1.r1 378 3.1e-34 25 495 sp|Q12698|PUR1_S AMIDOPHOSPHORIBOSYLTRANSFERASE
 (GLUTAMINEPHOSPHORIBOSYLPYROPHOSPHATE AMIDOTRANSFERASE) (ATASE) >gi|98
 r5c10a1.r1 318 2.1e-27 448 795 sp|P41390|PUR1_SCHPO AMIDOPHOSPHORIBOSYLTRANSFERASE
 (GLUTAMINEPHOSPHORIBOSYLPYROPHOSPHATE AMIDOTRANSFERASE) (AT

<PHOSPHORIBOSYLAMINOIMIDAZOLECARBOXAMIDE FORMYLTRANSFERASE-9th step>

n1b11a1.r1 583 5.8e-56 49 450 sp|P38009|PU92_YEAST PHOSPHORIBOSYLAMINOIMIDAZOLECARBOXAMIDE
 FORMYLTRANSFERASE2 (AICAR TRANSFORMYLASE) / IMP CY
 Contig900_t2g09a1.f1 138 1.1e-07 139 276 sp|P38009|PU92_Y PHOSPHORIBOSYLAMINOIMIDAZOLECARBOXAMIDE
 FORMYLTRANSFERASE2 (AICAR TRANSFORMYLASE) / IMP CYCLOHYDROLAS

b. other purine metabolic enzymes

<INOSINE-5'-MONOPHOSPHATE DEHYDROGENASE>

Contig1754_c8d12a1.f1 1706 5.5e-175 244 1845 sp|P50094|IMH3_Y PROBABLE INOSINE-5'-MONOPHOSPHATE DEHYDROGENASE
 (IMPDEHYDROGENASE) (IMPDH) (IMPD) >pir||s50890 hypoth

<xanthine dehydrogenase>

m6e07a1.r1 1208 3.5e-122 74 832 sp|Q12553|XDH_EMENI XANTHINE DEHYDROGENASE (PURINE HYDROXYLASE I)
 >pir||A55875xanthine dehydrogenase (EC 1.1.1)
 m6e07a1.f1 915 1.4e-90 52 621 sp|Q12553|XDH_EMENI XANTHINE DEHYDROGENASE (PURINE HYDROXYLASE I)
 >pir||A55875xanthine dehydrogenase (EC 1.1.1)

<ADENYLOSUCCINATE SYNTHETASE-first committed step to AMP biosyn>

s9f08a1.r1 448 1e-41 37 516 sp|P80210|PURA_YEAST ADENYLOSUCCINATE SYNTHETASE (IMP--ASPARTATE
 LIGASE)>pir||s48515 adenylosuccinate synthase

<Purine Nucleoside Phosphorylase>

Contig1116_d5b06a1.f1 220 8.7e-36 553 840 sp|Q05788|PNPH_Y PROBABLE PURINE NUCLEOSIDE PHOSPHORYLASE
 (INOSINEPHOSPHORYLASE) (PNP) >pir||s48560 hypothetical prote

<AMP DEAMINASE>

Contig992_c3h03a1.f1 1551 1.6e-158 61 1197 sp|P15274|AMDM_Y AMP DEAMINASE (MYOADENYLATE DEAMINASE) >pir||s49744
 AMPdeaminase (EC 3.5.4.6) - yeast (Saccharomyces

<adenosine kinase-phosphorylates purine nucleoside>

*Contig838_y6e09a1.r1 405 4e-37 46 609 gnl|PID|e1198603 (Y15430) adenosine kinase [Physcomitrella patens]
y6e09a1.f1 145 7.1e-07 385 618 sp|P47143|ADK_YEAST PUTATIVE ADENOSINE KINASE >pir||S57126 ribokinase
homologyYJR105w - yeast (Saccharomyces ce

3. Pyrimidine metabolism (4)

a. de novo pyrimidine biosynthesis

<dihydroorotate dehydrogenase-4th step in pyr biosyn>

w6f03a1.r1 682 1.8e-66 81 551 gi|1181887 (U47318) dihydroorotate dehydrogenase [Emericella
nidulans]
Contig138_j9d03a1.f1 430 9.5e-40 103 348 gi|1181887 (U47318) dihydroorotate dehydrogenase [Emericella
nidulans]
w6f03a1.f1 232 5.8e-18 338 577 gi|1181887 (U47318) dihydroorotate dehydrogenase [Emericella
nidulans]

b. other pyrimidine metabolic enzymes

<DEOXYCYTIDYLATE DEAMINASE-degeradation to dUMP>

Contig885_r7c07a1.f1 556 3.8e-67 91 549 gnl|PID|e1263904 (AL022103) deoxycytidylate deaminase
[Schizosaccharomycespombe]

D. Metabolism of Lipids, Fatty Acids, Sterols-See also fatty acid degradation

1. Fatty acid biosynthesis (7)

<a. ACETYL-COA CARBOXYLASE-yields malonylcoA,comitted step to FA biosyn.>

<b. ACYL-CARRIER PROTEINS>

Contig449_d5b09a1.f1 285 2.2e-24 158 865 gnl|PID|e1185182 (Z99112) 3-ketoacyl-acyl carrier protein reductase
[Bacillussubtilis]
Contig140_j7h12a1.f1 271 6.9e-23 92 433 gi|2827320 (AF042860) 3-oxoacyl-[acyl-carrier-protein]-reductase
[Neurospora crassa]

<c. FATTY ACID SYNTHASE>

Contig950_r5g09a1.f1 785 1.3e-169 777 1226 gi|1805261 (U75347) fatty acid synthase, alpha subunit
[Emericella nidulans]
r7b10a1.r1 1012 3e-100 54 656 gi|1805261 (U75347) fatty acid synthase, alpha subunit
[Emericella nidulans]

<d. BRANCHED-CHAIN ALPHA-KETO ACID DEHYDROGENASE-keto acids to short branch-chain fatty acids>

Contig1741_f5h02a1.f1 824 1.6e-81 106 954 sp|P50136|ODBA_M 2-OXOISOVALERATE DEHYDROGENASE ALPHA SUBUNIT
PRECURSOR(BRANCHED-CHAIN ALPHA-KETO ACID DEHYDROGENASE C
Contig1560_i2a01a1.r1 343 1.6e-30 175 612 sp|P50136|ODBA_M 2-OXOISOVALERATE DEHYDROGENASE ALPHA SUBUNIT
PRECURSOR(BRANCHED-CHAIN ALPHA-KETO ACID DEHYDROGENASE C

e. other

<stearoyl-CoA desaturase-adds double bonds to fatty acyl coA>

j5f02a1.r1 485 1.4e-45 321 755 pir||852746 stearyl-CoA desaturase (EC 1.14.99.5) - *Ajellomyces capsulata*>gi|757862 (X85962) delta-9

2. sterols (19)

a. sterol metabolism

<sterol>

*Contig1334_g3e09a1.r1 182 2e-10 28 693 gnl|PID|e314043 (Y12693) oxysterol-binding protein [*Neurospora crassa*]

*Contig1364_c1g09a1.f1 133 5.4e-05 425 820 sp|Q02318|CP27_H STEROL 26-HYDROXYLASE MITOCHONDRIAL PRECURSOR (VITAMIND(3) 25-HYDROXYLASE)(5-BETA-CHOLESTANE-3-ALPHA,

<steroid monooxygenase>

d5g12a1.r1 246 1.9e-19 262 717 gnl|PID|d1025370 (AB010439) steroid monooxygenase [*Rhodococcus rhodochrous*]

<LANOSTEROL SYNTHASE>

Contig252_h1b11a1.f1 639 7.2e-62 129 749 sp|P38604|ERG7_Y LANOSTEROL SYNTHASE (OXIDOSQUALENE--LANOSTEROL CYCLASE)(2,3-EPOXYSQUALENE--LANOSTEROL CYCLASE) (OSC)

<glucuronidase>

t2f06a1.r1 175 1.1e-11 52 420 pir||843555 beta-glucuronidase - *Escherichia coli* >gi|475169

Contig1101_alb06c9.r1 126 1.6e-05 72 392 gi|529326 (U12638) beta-glucuronidase [Cloning vector pdeltagusBin19]>gi|529330 (U12639) beta-glucuronidase [Cl

<HYDROXY-3-METHYLGLUTARYL-COENZYME A REDUCTASE-also mevalonate biosyn to isoprenoids>

q0d11a1.r1 423 4.8e-39 27 365 gnl|PID|e233478 (X94308) HMG-CoA-reductase [*Sphaceloma manihoticola*]

<C-5 STEROL DESATURASE>

q0g11a1.f1 261 9e-42 290 523 sp|P32353|ERG3_YEAST C-5 STEROL DESATURASE >pir||JQ1146 C-5 sterol desaturase(EC 1.--.--) - yeast (*Saccharomyce*

q0g11a1.r1 173 1.6e-12 213 488 gnl|PID|d1019713 (D85181) fungal sterol-C5-desaturase homolog [*Homo sapiens*]

<sterol demethylase>

w9e04a1.r1 617 1.6e-59 34 636 gi|2406574 (U72657) eburicol C14-alpha-demethylase [*Uncinula necator*]>gi|2406576 (U72658) eburicol 14

w9e04a1.f1 275 1.2e-22 244 582 gi|2406574 (U72657) eburicol C14-alpha-demethylase [*Uncinula necator*]>gi|2406576 (U72658) eburicol 14

b.Farnesol biosynthesis

<GERANYLGERANYL PYROPHOSPHATE SYNTHETASE>

Contig1202_g8c10a1.r1 885 6.3e-88 61 831 sp|P24322|GGPP_N GERANYLGERANYL PYROPHOSPHATE SYNTHETASE (GGPP SYNTHETASE)(DIMETHYLALLYLTRANSFERASE / GERANYLTRANSTRAN

Contig727_w6a02a1.f1 217 1.4e-16 342 536 sp|P24322|GGPP_N GERANYLGERANYL PYROPHOSPHATE SYNTHETASE (GGPP SYNTHETASE)(DIMETHYLALLYLTRANSFERASE / GERANYLTRANSTRAN

<GERANYLGERANYL TRANSFERASE>

d5a07a1.f1 306 1.4e-26 340 594 sp|P20133|BET2_YEAST TYPE II PROTEINS GERANYLGERANYLTRANSFERASE BETA SUBUNIT(TYPE II PROTEIN GERANYL-GERANYLTRA
d5a07a1.r1 289 8.9e-25 231 641 sp|P53611|PGTB_HUMAN GERANYLGERANYL TRANSFERASE TYPE II BETA SUBUNIT (RABGERANYLGERANYLTRANSFERASE BETA SUBUNIT
<hydroxysteroid dehydrogenase>
m6f07a1.r1 305 1.6e-26 320 679 sp|P51652|PE2R_RAT 20-ALPHA-HYDROXYSTEROID DEHYDROGENASE (20-ALPHA-HSD) (HSD1)>gnl|PID|d1003827 (D14424) 20-a
g4f11a1.f1 186 1.2e-11 159 470 gnl|PID|e1254577 (AL022019) putative 3-beta-hydroxysteroid dehydrogenase[Schizosaccharomyces pombe]

c.cholesterol metabolism

<C-4 METHYL STEROL OXIDASE-cholesterol biosynthesis>

Contig1481_a0g03a1.f1 559 2.1e-53 260 649 gi|2970627 (AF051914) C-4 methyl sterol oxidase [Candida albicans]
i0b10a1.r1 427 2.1e-39 15 470 sp|P53045|ER25_YEAST C-4 METHYL STEROL OXIDASE >pir||s64354 ERG25 protein -yeast (Saccharomyces cerevisiae) >gi

<STEROL O-ACYLTRANSFERASE-esterification of cholesterol>

o0d02a1.r1 151 4.3e-09 16 180 sp|P53629|ARE2_YEAST STEROL O-ACYLTRANSFERASE 2 (STEROL-ESTER SYNTHASE 2)>pir||s63350 probable membrane protein

<ISOPENTENYL-DIPHOSPHATE DELTA-ISOMERASE-ISOPRENE and CHOLESTEROL BIOSYNTHESIS>

Contig1411_c7a08a1.f1 727 3.1e-71 133 783 sp|Q10132|IPPI_S ISOPENTENYL-DIPHOSPHATE DELTA-ISOMERASE (IPP ISOMERASE)>pir||A56442 isopentenyl-diphosphate Delta-iso

3. lipids (25)

a. phospholipid metabolism

<LYSOPHOSPHOLIPASE PRECURSOR>

o8d03a1.r1 636 1.5e-61 29 583 sp|P39457|PLB1_PENNO LYSOPHOSPHOLIPASE PRECURSOR (PHOSPHOLIPASE B)>pir||s29318 lysophospholipase (EC 3.1.1.5) -
Contig635_o8d03a1.f1 633 3e-61 155 706 sp|P39457|PLB1_P LYSOPHOSPHOLIPASE PRECURSOR (PHOSPHOLIPASE B)>pir||s29318 lysophospholipase (EC 3.1.1.5) - Penicilliu
g5d06a1.r1 611 6.6e-59 34 624 sp|P39457|PLB1_PENNO LYSOPHOSPHOLIPASE PRECURSOR (PHOSPHOLIPASE B)>pir||s29318 lysophospholipase (EC 3.1.1.5) -

<CDP-DIGLYCERIDE PYROPHOSPHORYLASE-PHOSPHOLIPID BIOSYNTHESIS>

Contig129_j9d09a1.f1 387 3.4e-35 399 797 sp|O04940|CDS1_S PHOSPHATIDATE CYTIDYLYLTRANSFERASE (CDP-DIGLYCERIDESYNTHETASE) (CDP-DIGLYCERIDE PYROPHOSPHORYLASE) (C
*Contig1059_d0f03a1.f1 222 1.5e-14 639 872 gnl|PID|d1033240 (AB010810) phospholipase D [Candida albicans]

<phosphatidyl synthase>

Contig1664_m3d12a1.r1 388 3.5e-35 8 691 gnl|PID|e349677 (Z99295) phosphatidyl synthase [Schizosaccharomyces pombe]
d4b08a1.f1 153 1.8e-07 207 566 gnl|PID|e349677 (Z99295) phosphatidyl synthase [Schizosaccharomyces pombe]

<PHOSPHATIDYLSERINE SYNTHASE>

Contig1674_a0a05a1.f1 402 4.7e-57 498 950 sp|P08456|PSS_YE CDP-DIACYLGLYCEROL--SERINE
O-PHOSPHATIDYLTRANSFERASE(PHOSPHATIDYLSERINE SYNTHASE) >pir||S00080CDPdiac
k0a08a1.f1 438 1.4e-40 17 550 sp|P79001|PEL1_SACPS PUTATIVE
CDP-DIACYLGLYCEROL--SERINEO-PHOSPHATIDYLTRANSFERASE (PHOSPHATIDYLSERINE
SYNTHASE)=*Saccaromyces pateurianus*
Contig1324_d4b03a1.r1 412 7.5e-38 164 616 sp|P08456|PSS_YE CDP-DIACYLGLYCEROL--SERINE
O-PHOSPHATIDYLTRANSFERASE(PHOSPHATIDYLSERINE SYNTHASE) >pir||S00080CDPdiac
k0a08a1.r1 200 1.6e-14 35 376 sp|P79001|PEL1_SACPS PUTATIVE
CDP-DIACYLGLYCEROL--SERINEO-PHOSPHATIDYLTRANSFERASE (PHOSPHATIDYLSERINE
SYNTHASE)

<PHOSPHATIDYLSERINE DECARBOXYLASE>
m0g03a1.f1 181 3.6e-13 223 483 gi|3329153 (AE001340) Phosphatidylserine Decarboxylase [*Chlamydia*
trachomatis]

<ETHANOLAMINE KINASE-PHOSPHATIDYLETHANOLAMINE SYNTHESIS>
Contig1288_j4e01a1.f1 282 1.6e-23 160 756 pir||A54980 easily shocked protein - fruit fly (*Drosophila*
melanogaster)>gi|532128 (L35604) ethanolamine kinase [
<myo-inositol phosphate synthase-biosynthesis of inositol containing phospholipids>
g5a11a1.f1 534 9.3e-51 102 749 sp|P42803|INO1_SPIPO MYO-INOSITOL-1-PHOSPHATE SYNTHASE (IPS)
>pir||S60302D-myo-inositol-3-phosphate synthase =*Spirodela*
polyrrhiza, duckweed
g5a11a1.r1 469 7.4e-44 123 638 gi|973313 (U30250) myo-inositol 1-phosphate synthase isozyme-2
[*Arabidopsis thaliana*]

b. SPHINGOLIPIDS
<serine palmitoyltransferase>
g4a07a1.r1 230 8.9e-18 245 589 gnl|PID|e1285366 (AL022299) putative serine
palmitoyltransferase[*Schizosaccharomyces pombe*]

c. lipopolysaccharide biosyn-biomembrane precursors
<UDP-glucose:sterol glucosyltransferase>
v7h12a1.r1 340 1.4e-29 5 439 gnl|PID|e1169031 (Z83832) UDP-glucose:sterol glucosyltransferase
[*Avenasativa*]

<UDP-GLUCOSE PYROPHOSPHORYLASE>
*Contig1575_c5f09a1.f1 719 2.1e-70 64 681 sp|P32861|UDPG_Y PROBABLE UTP--GLUCOSE-1-PHOSPHATE
URIDYLYLTRANSFERASE(UDP-GLUCOSE PYROPHOSPHORYLASE) (UDPGP) >pir||S3
x1e10a1.r1 407 2.6e-37 195 710 sp|P32861|UDPG_YEAST PROBABLE UTP--GLUCOSE-1-PHOSPHATE
URIDYLYLTRANSFERASE(UDP-GLUCOSE PYROPHOSPHORYLASE) (UDPG

E. Aromatic compound metabolism (6)
<4-coumarate--CoA ligase-thioester substrates for phenylpropanoid biosyn>
f5b09a1.r1 311 4.2e-27 40 585 sp|P31687|4CL2_SOYBN 4-COUMARATE--COA LIGASE 2 (4CL) (CLONE
4CL16)>pir||PQ0772 4-coumarate--CoA ligase soybean pathogen resistance
response(EC 6.2.1687

m0f05a1.r1 261 5e-21 26 469 gi|2911799 (AF008184) 4-coumarate:CoA ligase 1 [Populus balsamifera subsp.trichocarpa X Populus delto-a fatty acid coA?

<chorismate>
 Contig968_m5b08a1.f1 444 2.8e-41 10 600 sp|P32178|CHMU_Y CHORISMATE MUTASE (CM) >pir||A45921 chorismate mutase (EC5.4.99.5) - yeast (Saccharomyces cerevisiae)

n3f08a1.r1 166 6.2e-10 109 312 gi|2983461 (AE000715) chorismate mutase/prephenate dehydratase [Aquifexaelicus]

<aminobutyrate aminotransferase>
 Contig1708_c7h01a1.f1 875 6.9e-87 163 783 sp|P14010|GATA_E 4-AMINOBTYRATE AMINOTRANSFERASE (GAMMA-AMINO-N-BUTYRATETRANSAMINASE) (GABA TRANSAMINASE) (GABA AMINO

<CARBOXYMUONOLACTONE DECARBOXYLASE-aromatic hydrocarbon cat.>
 m7e09a1.f1 207 2.1e-15 229 477 gnl|PID|e1313496 (AL031155) 3-oxoadipate enol-lactonehydrolase/4-carboxymuconolactone decarboxylase [Strept

F. Sulfur Metabolism (3)

<ADENYLYLSULFATE KINASE>

<sulphur metabolite repression-4 genes, methionine-down, no S-up>

Contig776_z1c07a1.f1 260 9.6e-22 223 372 gi|1658298 (U75874) sconCp [Emericella nidulans]

-sulfate assimilation

<sulfate adenylyltransferase-leads to biosynthesis of cys&met>

g4d04a1.r1 811 4.1e-80 23 538 pir||S55034 sulfate adenylyltransferase (EC 2.7.7.4) - Emericella nidulans>gi|572513 (X82541) sulfate

g4d04a1.f1 471 4.4e-44 150 428 pir||S55034 sulfate adenylyltransferase (EC 2.7.7.4) - Emericella nidulans>gi|572513 (X82541) sulfate

G. Phosphate Metabolism (1)

<INORGANIC PYROPHOSPHATASE>

Contig903_x1d03a1.r1 618 1e-59 15 482 sp|O13505|IPYR_P INORGANIC PYROPHOSPHATASE (PYROPHOSPHATEPHOSPHO-HYDROLASE) (PPASE) >gnl|PID|e1180018 (AJ001000) inorg

H. Nitrogen Metabolism (see also amino acid metabolism) (6)

<nitrite reductase>

x1d07a1.r1 285 4.9e-23 8 166 sp|P22944|NIR_EMENI NITRITE REDUCTASE (NAD(P)H) >pir||JH0181 nitrite reductase(NADH) (EC 1.6.6.4), long form =E. nidulans

<NITROGEN METABOLIC REGULATION PROTEIN -NEGATIVE REGULATORY PROTEIN IN THE NITROGEN CONTROL CIRCUIT>

h1a08a1.r1 622 4.5e-60 30 518 gi|3015626 (AF041976) nitrogen metabolite repression regulator NmrA[Emericella nidulans

<cyanate lyase-cyanate, bicarbonate substrates>

m3f04a1.r1 111 3.4e-05 35 160 gi|2055402 (U90436) cyanate lyase; cyanase [synthetic construct]

-urea cycle

<glutamate dehydrogenase>

Contig1839_c1c09a1.f1 3021 2.8e-314 12 2426 bbs|138429 (S66039) NAD(+)-specific glutamate dehydrogenase,
NAD-GDH {EC1.4.1.2} [Neurospora crassa, Peptide, 10
Contig17_x1a06a1.r1 461 4.1e-74 244 588 bbs|138429 (S66039) NAD(+)-specific glutamate dehydrogenase,
NAD-GDH {EC1.4.1.2} [Neurospora crassa, Peptide, 10
<CARBAMOYL-PHOSPHATE SYNTHASE-also arginine and pyrimidine biosynthesis>
d3g03a1.r1 752 7.5e-74 32 604 sp|P87183|CARA_TRIVE CARBAMOYL-PHOSPHATE SYNTHASE, ARGININE-SPECIFIC,
SMALLCHAIN PRECURSOR (ARGININE-SPECIFIC C
d3g03a1.f1 288 2e-24 182 496 sp|P22572|CARA_NEUCR CARBAMOYL-PHOSPHATE SYNTHASE, ARGININE-SPECIFIC,
SMALLCHAIN PRECURSOR (ARGININE-SPECIFIC C

I. Metabolism of Cofactors, prosthetic groups

1.nicotinamide coenzymes (5)

<NICOTINATE-NUCLEOTIDE PYROPHOSPHORYLASE-DE NOVO BIOSYNTHESIS OF NAD AND NADP>

y8c08a1.f1 298 3.9e-36 87 389 sp|P43619|NADC_YEAST PUTATIVE NICOTINATE-NUCLEOTIDE
PYROPHOSPHORYLASE(CARBOXYLATING) (QUINOLINATE PHOSPHORIBOSY
y8c08a1.r1 182 2.6e-13 316 534 sp|P43619|NADC_YEAST PUTATIVE NICOTINATE-NUCLEOTIDE
PYROPHOSPHORYLASE(CARBOXYLATING) (QUINOLINATE PHOSPHORIBOSY

<kynureninase-biosyn of NAD cofactors>

Contig1362_c5h01a1.f1 525 8.4e-50 194 883 pir||S59898 kynureninase (EC 3.7.1.3) - rat >bbs|171864
kynureninase,L-kynurenine hydrolase {EC 3.7.1.3} [rats, 1
Contig539_c7e07a1.f1 516 7.5e-49 71 1006 gi|1050752 (Z50144) kynurenine/alpha-aminoadipate
aminotransferase [Rattusnorvegicus]
c5h01a1.r1 384 7.5e-35 138 767 pir||S59898 kynureninase (EC 3.7.1.3) - rat >bbs|171864
kynureninase,L-kynurenine hydrolase {EC 3.7.1.

2.biocytin (biotin) (0)

3.thiamine (4)

<thiamine>

Contig146_m6c09a1.f1 410 1.3e-37 321 728 gnl|PID|d1019649 (D45894) thiamine-4 [Neurospora crassa]
<THIAMIN BIOSYNTHESIS>
Contig1797_d4c08a1.f1 1666 9.5e-171 145 1167 sp|P42882|NMT1_A NMT1 PROTEIN HOMOLOG >pir||S53697 nmt1 protein
-Aspergillus parasiticus >gi|557050 (U15196) the expre
Contig318_g5h01a1.r1 231 7.3e-18 81 515 sp|P40386|THI4_S PROBABLE THIAMIN BIOSYNTHETIC BIFUNCTIONAL
ENZYME(CONTAINS: THIAMIN-PHOSPHATE PYROPHOSPHORYLASE (TMPP
Contig319_g5h01a1.f1 164 1.4e-10 125 463 sp|P40386|THI4_S PROBABLE THIAMIN BIOSYNTHETIC BIFUNCTIONAL
ENZYME(CONTAINS: THIAMIN-PHOSPHATE PYROPHOSPHORYLASE (TMPP

4.coenzyme A (2)

<acetyl-coenzyme A synthetase>

Contig1515_f2c11a1.f1 1316 1.3e-133 442 1209 sp|P16928|ACSA_E ACETYL-COENZYME A SYNTHETASE (ACETATE--COA

206

y4b06a1.r1 469 2.4e-69 8 292 LIGASE)(ACYL-ACTIVATING ENZYME) >pir||SYASAA acetate--CoA
sp|P16928|ACSA_EMENI ACETYL-COENZYME A SYNTHETASE (ACETATE--COA
LIGASE)(ACYL-ACTIVATING ENZYME) >pir||SYASAA ac
<acetyl coenzyme A acetyltransferase>

5. flavins (2)
<riboflavin synthase>
Contig102_k8h04a1.f1 269 1.1e-22 151 390 sp|P50861|RIB4_Y 6,7-DIMETHYL-8-RIBITYLLUMAZINE SYNTHASE (DMRL
SYNTHASE)(LUMAZINE SYNTHASE) (RIBOFLAVIN SYNTHASE BETA
<GTP cyclohydrolase II-riboflavin biosyn>
g6e06a1.r1 508 5.1e-48 137 736 gnl|PID|e1291629 (AL023287) GTP cyclohydrolase II [Schizosaccharomyces
pombe]

6. folate-methyl donor (5)
<folate>
Contig1825_z4c10a1.f1 306 1.4e-26 283 717 gi|2565196 (AF000381) non-functional folate binding protein [Homo
sapiens]
Contig1487_e9g09a1.f1 264 3.9e-22 254 508 gi|2565196 (AF000381) non-functional folate binding protein [Homo
sapiens]
Contig1786_a0h08a1.r1 235 4.7e-19 286 507 gi|2565196 (AF000381) non-functional folate binding protein [Homo
sapiens]
Contig148_j4g02a1.f1 234 9.9e-18 281 628 sp|P28037|FTDH_R 10-FORMYLTETRAHYDROFOLATE DEHYDROGENASE (FBP-CI)
>gi|908915(M59861) 10-formyltetrahydrofolate dehydro
Contig1124_i0c01a1.f1 182 1.3e-12 276 431 gi|2565196 (AF000381) non-functional folate binding protein [Homo
sapiens]

7. heme (3)
<heme>
Contig669_a1c01c9.r1 270 9.4e-23 263 517 gnl|PID|e1284430 (AL022245) ferrochelatase [Schizosaccharomyces
pombe]=(PROTOHEME FERRO-LYASE) (HEMESYNTHETASE)
<siroheme synthase>
c5c01a1.r1 263 4.9e-22 85 600 gi|2983676 (AE000730) siroheme synthase [Aquifex aeolicus]
-iron uptake
<FERRIC REDUCTASE TRANSMEMBRANE COMPONENT 2>
o5f06a1.r1 168 8.3e-11 17 505 sp|P36033|FRE2_YEAST FERRIC REDUCTASE TRANSMEMBRANE COMPONENT 2
PRECURSOR>pir||S38063 ferric reductase FRE2 pre

8. PANTOTHENATE (1)
<PANTOTHENATE SYNTHETASE>
g2d12a1.f1 279 9.8e-24 122 715 sp|P40459|PANC_YEAST PUTATIVE PANTOATE--BETA-ALANINE LIGASE
(PANTOTHENATESYNTHETASE) (PANTOATE ACTIVATING ENZYM
g2d12a1.r1 198 3.8e-15 232 420 sp|P56061|PANC_HELPY PANTOATE--BETA-ALANINE LIGASE (PANTOTHENATE
SYNTHETASE)(PANTOATE ACTIVATING ENZYME) >gi|23

9.Molybdopterin (3)

<molybdopterin biosynth>

c9e12a1.rl	217	2.1e-16	208 564	gnl PID e349592	(Z99258) molybdopterin biosynthesis
				[Schizosaccharomycespombe]	
Contig567_c5e06a1.f1	166	9.1e-12	131 433	gi 2984359	(AE000776) molybdopterin converting factor subunit 2
				[Aquifexaeolicus]	
Contig265_m8c12a1.f1	138	3e-06	410 745	sp P12281 MOEA_E	MOLYBDOPTERIN BIOSYNTHESIS MOEA PROTEIN
				>pir A32352	molybdopterin-converting factor chlE - Escherichi

J. Energy

1. Glycolysis (16)

<a.hexokinase>

m0c07a1.rl	139	1.5e-07	220 429	sp P27926 HXX3_RAT	HEXOKINASE TYPE III (HK III) >pir S13913 hexokinase
				(EC2.7.1.1) III - rat	>gi 1658068 (U7

<b.glucose-6-phosphate isomerase>

<c.fructose-6-phosphate2-kinase>

Contig1540_c3b10a1.rl	607	6.8e-103	513 974	sp P32604 F26_YE	FRUCTOSE-2,6-BISPHOSPHATASE
				>pir S56938	fructose-2,6-bisphosphate 2-phosphatase (EC 3.1.3.46) - yeast
Contig1737_m8e11a1.f1	679	4.1e-66	388 1233	sp P40433 6P21_Y	6-PHOSPHOFRUCTO-2-KINASE 1 (PHOSPHOFRUCTOKINASE 2
				I)(6PF-2-K 1)	>pir S48465 6-phosphofructo-2-kinase
Contig1005_c1e06a1.f1	620	7.5e-60	2 742	sp P32604 F26_YE	FRUCTOSE-2,6-BISPHOSPHATASE
				>pir S56938	fructose-2,6-bisphosphate 2-phosphatase (EC 3.1.3.46) - yeast
g0e07a1.f1	222	1.8e-16	219 449	gi 172136	(M80801) 6-phosphofructo-2-kinase [Saccharomyces
				cerevisiae]	

<d.fructose-bisphosphate aldolase-also gluconeogenesis, PP cycle, carbon fixation, fructose and mannose metab>

Contig455_d4g02a1.rl	600	8.3e-58	113 682	sp P14540 ALF_YE	FRUCTOSE-BISPHOSPHATE ALDOLASE
				>pir ADBY2	fructose-bisphosphate aldolase (EC 4.1.2.13) II - yeast(Sac
c5b07a1.rl	251	7.8e-21	112 381	sp P36580 ALF_SCHPO	FRUCTOSE-BISPHOSPHATE ALDOLASE >gnl PID d1004756
				(D17415)	fructose 1,6-bisphosphate aldolas

<e.triose-phosphate isomerase>

m7h07a1.f1	468	8.4e-44	217 492	sp P04828 TPIS_EMENI	TRIOSEPHOSPHATE ISOMERASE (TIM)
				>pir ISASTN	triose-phosphate isomerase (EC 5.3.1.1) - Emer

<f.glyceraldehyde-3-phosphate dehydrogenase>

Contig1588_c5c07a1.rl	987	9e-99	44 622	pir DEASG3	glyceraldehyde-3-phosphate dehydrogenase (EC 1.2.1.12)
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Contig1637_a5b02a1.f1 569 1.8e-54 212 739 -*Emericella nidulans* >gi|168049 (M19694) glyce
 pir||DEASG3 glyceraldehyde-3-phosphate dehydrogenase (EC 1.2.1.12)
 Contig570_c5d10a1.f1 380 1.9e-34 118 537 -*Emericella nidulans* >gi|168049 (M19694) glyce
 gi|551682 (L07497) glyceraldehyde-3-phosphate dehydrogenase
 [*Anabaenavariabilis*]

<g.phosphoglycerate kinase>

<h.phosphoglycerate mutase>

z3b09a1.r1 626 1.6e-60 11 754 gi|2773203 (AF039713) Similar to phosphoglycerate mutase; coded
 for by *C.elegans* cDNA yk357d11.5; cod
 Contig1282_hlh12a1.f1 341 2.5e-30 222 443 sp|Q12560|ENO_AS ENOLASE (2-PHOSPHOGLYCERATE
 DEHYDRATASE)(2-PHOSPHO-D-GLYCERATE HYDRO-LYASE) >pir||JC45426beta-hydroxy

<i.phosphopyruvate hydratase>

<j.pyruvate kinase>

Contig176_i8e01a1.r1 850 3.1e-84 344 934 sp|P22360|KPYK_E PYRUVATE KINASE (PK) >pir||S27364 pyruvate kinase
 (EC2.7.1.40) - *Emericella nidulans* >gi|168074 (M369
 o5g10a1.r1 821 3.7e-81 11 499 sp|P22360|KPYK_EMENI PYRUVATE KINASE (PK) >pir||S27364 pyruvate kinase
 (EC2.7.1.40) - *Emericella nidulans* >gi|1
 Contig972_i8e01a1.f1 321 9.1e-28 379 555 sp|P22360|KPYK_E PYRUVATE KINASE (PK) >pir||S27364 pyruvate kinase
 (EC2.7.1.40) - *Emericella nidulans* >gi|168074 (M369

2. Gluconeogenesis (6)

<a.LACTATE DEHYDROGENASE>

<b.pyruvate carboxylase>

h1c05a1.r1 433 8.4e-39 24 350 sp|P32327|PYC2_YEAST PYRUVATE CARBOXYLASE 2 (PYRUVIC CARBOXYLASE 2) (PCB
 2)>pir||S46094 pyruvate carboxylase (E
 Contig1375_d1d10a1.f1 407 5.1e-36 326 712 gn1|PID|d1011901 (D78170) pyruvate carboxylase [*Schizosaccharomyces*
 pombe]

<c.phosphoenolpyruvate carboxykinase>

Contig1602_d2g02a1.r1 779 2.9e-101 164 808 gi|2738107 (U88575) phosphoenolpyruvate carboxykinase
 [*Kluyveromyces lactis*]
 Contig1620_d2g02a1.f1 659 7.9e-89 484 957 gi|2738107 (U88575) phosphoenolpyruvate carboxykinase
 [*Kluyveromyces lactis*]

<d.FRUCTOSE-1,6-BISPHOSPHATASE>

Contig292_g7g01a1.r1 544 7.2e-52 253 855 sp|P09201|F16P_Y FRUCTOSE-1,6-BISPHOSPHATASE
 (D-FRUCTOSE-1,6-BISPHOSPHATE1-PHOSPHOHYDROLASE) (FBPASE) >pir||PABY fruct
 x8e05a1.f1 387 3.6e-35 171 554 sp|P09201|F16P_YEAST FRUCTOSE-1,6-BISPHOSPHATASE

(D-FRUCTOSE-1,6-BISPHOSPHATE1-PHOSPHOHYDROLASE) (FBPASE) >pir|

3. Pentose-phosphate pathway (9)

<a.glucose-6-phosphate dehydrogenase>

n3f11a1.r1	1132	4.1e-114	31	681	sp P41764 G6PD_EMENI	GLUCOSE-6-PHOSPHATE 1-DEHYDROGENASE
					(G6PD)>gnl PID e99568	(X77830) glucose-6-phosphate 1-d
Contig619_c2a06a1.f1	737	2.7e-72	227	685	sp P41764 G6PD_E	GLUCOSE-6-PHOSPHATE 1-DEHYDROGENASE
					(G6PD)>gnl PID e99568	(X77830) glucose-6-phosphate 1-dehydrogenas

<b. gluconeolactonase>

<c.phosphogluconate dehydrogenase>

m2g12a1.f1	503	1.7e-47	190	597	sp P53319 6PG2_YEAST	6-PHOSPHOGLUCONATE DEHYDROGENASE, DECARBOXYLATING
					2>pir S64588	phosphogluconate dehydroge
n8f05a1.f1	213	5.8e-16	120	518	gi 3322609	(AE001213) phosphogluconate dehydrogenase (gnd)
					[Treponemapallidum]	

<d.ribose 5-phosphate isomerase-nonoxidative P04>

Contig1751_d4f10a1.f1	276	1.8e-23	121	570	gi 2983605	(AE000725) ribose 5-phosphate isomerase B [Aquifex
					aeolicus]	

<e.ribulose-phosphate 3-epimerase>

<f.transketolase>

y4a09a1.r1	549	2.4e-52	19	468	sp Q12630 TKT1_KLULA	TRANSKETOLASE (TK) >gi 1488336 (U65983)
					transketolase[Kluyveromyces lactis]	
m0d06a1.r1	482	2.6e-45	11	448	sp Q12630 TKT1_KLULA	TRANSKETOLASE (TK) >gi 1488336 (U65983)
					transketolase[Kluyveromyces lactis]	
Contig958_h4a05a1.f1	297	9.5e-23	998	1267	sp Q12630 TKT1_K	TRANSKETOLASE (TK) >gi 1488336 (U65983)
					transketolase[Kluyveromyces lactis]	

<g.transaldolase>

Contig1698_d4f04a1.f1	1097	1.6e-110	142	1107	gnl PID e1292580	(AL023518) Tallp transaldolase [Schizosaccharomyces
					pombe]	

4. Pyruvate dehydrogenase (5)

<pyruvate dehydrogenase>

o0a06a1.r1	609	1e-58	17	466	sp P16387 ODPA_YEAST	PYRUVATE DEHYDROGENASE E1 COMPONENT, ALPHA
					SUBUNITPRECURSOR (PDHE1-A)	>gi 298059 (X71664)
y3g01a1.f1	489	5e-46	177	680	gi 2623175	(AF030425) pyruvate dehydrogenase E1 component alpha
					subunit[Pichia stipitis]	

<pyruvate decarboxylase>

<dihydrolipoyl transacetylase>

Contig1423_f5f03a1.f1 549 2.4e-52 77 811 sp|P53395|ODB2_M LIPOAMIDE ACYLTRANSFERASE COMPONENT (E2) PRECURSOR
OFBRANCHED-CHAIN ALPHA-KETO ACID DEHYDROGENASE COM
<dihydrolipoamide dehydrogenase>
Contig1537_h8f04a1.r1 1470 5.6e-150 9 1370 gi|1911177 (L40360) dihydrolipoamide dehydrogenase
[Schizosaccharomyces pombe]
<pyruvate dehydrogenase kinase-inhibits pyruvate dehyd by phos of E1 alpha subunit>
vle08a1.r1 169 1.1e-09 47 598 gnl|PID|e1285215 (AJ001418) pyruvate dehydrogenase kinase-like protein
[Musmusculus]

5. Tricarboxylic acid pathway (18)

<a.citrate synthase>

d3a06a1.r1 952 4.5e-95 43 636 sp|O00098|CISY_EMENI CITRATE SYNTHASE, MITOCHONDRIAL PRECURSOR
>gi|2138332(U89675) citrate synthase [Emericella
Contig1245_d5a04a1.r1 607 1.7e-58 84 821 sp|P79024|CISY_C CITRATE SYNTHASE, MITOCHONDRIAL
PRECURSOR>gnl|PID|d1020163 (AB001565) citrate synthase [Candida trophi
d3a06a1.f1 428 1.6e-39 196 483 sp|O00098|CISY_EMENI CITRATE SYNTHASE, MITOCHONDRIAL PRECURSOR
>gi|2138332(U89675) citrate synthase [Emericella
Contig1092_d5a04a1.f1 358 3.7e-32 133 459 sp|P43635|CISX_Y CITRATE SYNTHASE 3 >pir||S52814 citrate (si)-synthase
(EC4.1.3.7) - yeast (Saccharomyces cerevisiae)

<b.aconitate hydratase>

Contig1726_clg04a1.f1 1464 2.5e-149 45 1178 sp|P19414|ACON_Y ACONITATE HYDRATASE, MITOCHONDRIAL PRECURSOR
(CITRATEHYDRO-LYASE) (ACONITASE) >pir||S50387 aconitate

<c.isocitrate dehydrogenase>

Contig119_k0c02a1.f1 528 3.4e-50 183 515 sp|P79089|IDHP_A ISOCITRATE DEHYDROGENASE (NADP), MITOCHONDRIAL
PRECURSOR(OXALOSUCCINATE DECARBOXYLASE) (IDH) (NADP+-S
k0c02a1.r1 469 6.5e-44 4 342 sp|P79089|IDHP_ASPNG ISOCITRATE DEHYDROGENASE (NADP), MITOCHONDRIAL
PRECURSOR(OXALOSUCCINATE DECARBOXYLASE) (ID
Contig1171_e7a09a1.f1 437 1.7e-40 125 736 gi|1182011 (X87172) NAD+-isocitrate dehydrogenase, alpha subunit
[Macacafascicularis]
Contig543_c7b05a1.f1 367 4.6e-33 234 464 gi|2266941 (AF009036) NAD(+)-isocitrate dehydrogenase subunit I
[Ajellomycescapsulatus]
n0h09a1.r1 327 8.1e-29 141 452 gi|2266941 (AF009036) NAD(+)-isocitrate dehydrogenase subunit I
[Ajellomycescapsulatus]

<d.alpha-ketoglutarate dehydrogenase>

<e.SUCCINYL-COA LIGASE>

c3f09a1.r1 771 7e-76 18 755 sp|P53587|SUCB_NEOFR SUCCINYL-COA LIGASE (GDP-FORMING), BETA-CHAIN
PRECURSOR(SUCCINYL-COA SYNTHETASE, BETA CHAI
i7e06a1.r1 452 4.5e-42 17 325 sp|O13750|SUCA_SCHPO PROBABLE SUCCINYL-COA LIGASE (GDP-FORMING),
ALPHA-CHAINPRECURSOR (SUCCINYL-COA SYNTHETASE,

l3e03a1.r1 334 1.5e-29 125 433 sp|O13750|SUCA_SCHPO PROBABLE SUCCINYL-COA LIGASE (GDP-FORMING),
ALPHA-CHAINPRECURSOR (SUCCINYL-COA SYNTHETASE,

<f.succinate dehydrogenase>

<g.FUMARATE HYDRATASE>

Contig1327_fih10a1.f1 575 1.4e-98 536 982 sp|P08417|FUMH_Y FUMARATE HYDRATASE, MITOCHONDRIAL PRECURSOR
(FUMARASE)>pir||UFBYM fumarate hydratase (EC 4.2.1.2) pre

<h.malate dehydrogenase>

Contig1670_e9b05a1.f1 870 2.2e-86 78 1046 sp|O02640|MDHM_C PROBABLE MALATE DEHYDROGENASE, MITOCHONDRIAL
PRECURSOR>gi|2076896 (AF002197) similar to malate dehydr
h8e02a1.r1 629 7.7e-61 31 492 bbs|179680 (S83228) beta-isopropylmalate dehydrogenase
[Aspergillus niger, strain A733, Peptide, 363 a
m8h04a1.r1 518 4e-49 127 612 sp|P17505|MDHM_YEAST MALATE DEHYDROGENASE, MITOCHONDRIAL
PRECURSOR>pir||DEBYMM malate dehydrogenase (EC 1.1.1.3
h8e02a1.f1 213 4.2e-28 139 306 bbs|179680 (S83228) beta-isopropylmalate dehydrogenase
[Aspergillus niger, strain A733, Peptide, 363 a

6. related reactions (6)

<citrate lyase-citrate to oxaloacetate+acetylcoa>

Contig1615_d5a08a1.r1 957 1.3e-95 242 1066 gnl|PID|e349683 (Z99295) citrate lyase [Schizosaccharomyces pombe]
k9f05a1.f1 562 9.8e-53 10 582 sp|P16638|ACLY_RAT ATP-CITRATE (PRO-S-)-LYASE (CITRATE CLEAVAGE
ENZYME)>pir||A35007 ATP citrate (pro-S)-lyase
x7b07a1.r1 424 7.3e-49 32 472 gnl|PID|e349683 (Z99295) citrate lyase [Schizosaccharomyces pombe]
j0h12a1.f1 353 1.3e-31 213 458 gnl|PID|d1014552 (D89194) similar to Rat ATP citrate-lyase,
SWISS-PROTAccession Number P16638 [Schizosaccha

<carbonic anhydride>

Contig1792_c3a07a1.r1 542 1.3e-51 165 788 gnl|PID|d1013665 (D86050) carbonic anhydrase [Porphyridium purpureum]
n0d03a1.f1 220 1.3e-16 108 452 gnl|PID|d1013666 (D86051) carbonic anhydrase [Porphyridium purpureum]

7. glyoxylate cycle (5)

<malate synthase>

o6e06a1.r1 931 6.9e-93 1 534 sp|P28344|MASY_EMENI MALATE SYNTHASE, GLYOXYSOMAL >pir||S17773 malate
synthase(EC 4.1.3.2) - Emericella nidulans
o6h07a1.r1 718 2.9e-70 82 552 sp|P28344|MASY_EMENI MALATE SYNTHASE, GLYOXYSOMAL >pir||S17773 malate
synthase(EC 4.1.3.2) - Emericella nidulans
Contig290_g8a02a1.f1 487 8.2e-46 190 480 sp|P28344|MASY_E MALATE SYNTHASE, GLYOXYSOMAL >pir||S17773 malate
synthase(EC 4.1.3.2) - Emericella nidulans >gi|2702

<isocitrate lyase>

Contig1259_h0h01a1.r1 1396 3.8e-142 6 866 pir||S26857 isocitrate lyase (EC 4.1.3.1) - Emericella nidulans
Contig1173_h0h03a1.f1 839 3.8e-83 109 585 sp|P28298|ACEA_E ISOCITRATE LYASE (ISOCITRASE) (ISOCITRATASE)

(ICL)>gi|2317 (X62696) isocitrate lyase [Emericella nidu

8. Fermentation, alcoholic (15)

<a.pyruvate decarboxylase>

Contig1526_d2a11a1.f1 731 4.2e-113 497 940 sp|P87208|DCPY_E PYRUVATE DECARBOXYLASE >gi|2160688 (U73194)
pyruvatedecarboxylase [Emericella nidulans]

<b.alcohol dehydrogenase>

Contig1832_a1a03c9.r1 1694 1e-173 60 1109 sp|P08843|ADH1_E ALCOHOL DEHYDROGENASE I >pir||A29054
alcoholdehydrogenase (EC 1.1.1.1) - Emericella nidulans >gi|1680
Contig848_z3d06a1.r1 783 4.2e-101 17 538 sp|P07754|ADH3_B ALCOHOL DEHYDROGENASE III >pir||A24648
alcoholdehydrogenase (EC 1.1.1.1) III - Emericella nidulans >g
Contig354_g3d03a1.f1 605 2.7e-58 73 810 gi|1790870 (U32622) toluenesulfonate zinc-independent alcohol
dehydrogenase[Comamonas testosteroni]
Contig1275_g2b06a1.f1 540 2.2e-51 144 875 gi|1790870 (U32622) toluenesulfonate zinc-independent alcohol
dehydrogenase[Comamonas testosteroni]
z5a05a1.f1 492 2.6e-46 133 444 sp|P07754|ADH3_EMENI ALCOHOL DEHYDROGENASE III >pir||A24648
alcoholdehydrogenase (EC 1.1.1.1) III - Emericella
j5d06a1.r1 423 5.2e-39 48 833 sp|P00332|ADH_SCHPO ALCOHOL DEHYDROGENASE >pir||DEZPA alcohol
dehydrogenase(EC 1.1.1.1) - fission yeast (Schiz
Contig1703_c4c04a1.r1 361 1.8e-32 50 997 gnl|PID|e209885 (X92868) NADP-dependent alcohol dehydrogenase
[Bacillus subtilis] >gnl|PID|e1183931 (Z99117) NADP-depe
Contig186_i7c03a1.r1 337 6.5e-30 28 627 sp|P39714|YAG0_Y HYPOTHETICAL ZINC-TYPE ALCOHOL DEHYDROGENASE-LIKE
PROTEININ GDH3-CNE1 INTERGENIC REGION >pir||S51962
*Contig411_e9g02a1.r1 239 1.6e-19 28 507 gi|1934622 (U93875) alcohol dehydrogenase [Bacillus subtilis]
c4b11a1.r1 230 3.3e-18 187 648 sp|P39714|YAG0_YEAST HYPOTHETICAL ZINC-TYPE ALCOHOL DEHYDROGENASE-LIKE
PROTEININ GDH3-CNE1 INTERGENIC REGION >p
Contig1508_d4d04a1.r1 170 1.5e-09 94 675 sp|P42328|ADH3_B ALCOHOL DEHYDROGENASE (ADH-HT) >pir||S45605
alcoholdehydrogenase (EC 1.1.1.1) - Bacillus stearothermo
Contig406_f0c08a1.f1 135 2.6e-06 384 665 gi|1118145 (U41749) strong similarity to the insect-type
alcoholdehydrogenase/ribitol dehydrogenase family [Caen
w9c01a1.r1 122 6.3e-05 348 566 gnl|PID|e1183501 (Z99113) similar to alcohol dehydrogenase [Bacillus
subtilis]>gnl|PID|e1185316 (Z99114) si
j7g12a1.f1 123 0.00013 127 417 sp|P42327|ADH2_BACST ALCOHOL DEHYDROGENASE (ADH) >pir||S47643
alcoholdehydrogenase (EC 1.1.1.1) - Bacillus stea

9.Fermentation, other (2)

<LACTATE DEHYDROGENASE-pyruvate to lactate>

r4g05a1.r1 386 4.4e-35 113 655 sp|P52643|LDHD_ECOLI D-LACTATE DEHYDROGENASE (D-LDH) >gi|1049265
(U36928)D-lactate dehydrogenase [Escherichia c

<butanediol dehydrogenase>

Contig1693_a5d12a1.f1 438 1.3e-40 145 945 gnl|PID|d1013772 (D86412) meso-2,3-butanediol dehydrogenase (D-acetoinforming) [Klebsiella pneumoniae]

10. Monocarbon metabolism (2)

<formate dehydrogenase>

Contig1117_d5c06a1.f1 917 2.2e-91 143 748 sp|Q07103|FDH_NE FORMATE DEHYDROGENASE (NAD-DEPENDENT FORMATEDEHYDROGENASE) (FDH) >pir||A47117 formate dehydrogenase (

C1 Metabolism

<ALCOHOL OXIDASE-first step-methanol utilization to FORMALDEHYDE>

x8a04a1.r1 174 1.6e-11 1 528 sp|P04841|ALOX_PICAN ALCOHOL OXIDASE (AOX) (METHANOL OXIDASE) (MOX)>pir||OXHQAP alcohol oxidase (EC 1.1.3.13) -

11. Metabolism of energy reserves (glycogen, starch, trehalose) (19)

a. Glycogen degradation

<glycogen phosphorylase>

Contig122_j9h10a1.r1 519 2.1e-48 37 594 pir||S61144 glycogen phosphorylase (EC 2.4.1.1) - yeast (Saccharomycescerevisiae) >gi|849168 (U28371) Glycogen ph

Contig1329_c4e04a1.r1 509 2.7e-47 28 504 pir||S61144 glycogen phosphorylase (EC 2.4.1.1) - yeast (Saccharomycescerevisiae) >gi|849168 (U28371) Glycogen ph

<PHOSPHOGLUCOMUTASE-glycogen deg, glu1PO4 to glu6PO4>

Contig519_c8e10a1.f1 431 7e-40 84 722 sp|P37012|PGM2_Y PHOSPHOGLUCOMUTASE 2 (GLUCOSE PHOSPHOMUTASE 2) (PGM 2)>pir||S41200 phosphoglucomutase (EC 5.4.2.2) PG

<GLYCOGEN DEBRANCHING ENZYME>

*Contig1382_c4a08a1.f1 867 5.6e-85 148 1257 sp|P35574|GDE_RA GLYCOGEN DEBRANCHING ENZYME (4-ALPHA-GLUCANOTRANSFERASE(OLIGO-1,4-1,4-GLUCANTRANSFERASE) / AMYLO-1,6-

b. Starch degradation

<alpha glucosidase>

l3h03a1.r1 621 2.3e-59 30 539 sp|Q12558|AGLU_ASPOR ALPHA-GLUCOSIDASE PRECURSOR (MALTASE) (AGL) >pir||JC4217alpha-glucosidase (EC 3.2.1.20) -

r5e04a1.r1 528 2.3e-49 5 652 sp|O04893|AGLU_SPIOL ALPHA-GLUCOSIDASE PRECURSOR (MALTASE)

o4g07a1.r1 278 6.6e-23 41 439 >gnl|PID|d1020713(D86624) alpha-glucosidase precours, glucoamylase pir||S44188 alpha-glucosidase (EC 3.2.1.20) - Staphylococcus xylosus>gi|474177 (X78853) alpha-D-1,4-gl

l0f02a1.f1 167 1.6e-10 85 480 sp|P38138|YB79_YEAST PUTATIVE FAMILY 31 GLUCOSIDASE IN PCS60-ABD1 INTERGENICREGION >pir||S46105 glucan 1,4-alpha=glucosidase

<glucoamylase>

Contig1841_ale02c9.r1 1195 5.2e-201 736 1914 dbj||AB008370_1 (AB008370) acid-stable alpha-amylase [Aspergillus kawachii]

y4b12a1.r1	750	1.3e-73	8 649	sp P36914 AMYG_ASPOR	GLUCOAMYLASE PRECURSOR (GLUCAN
Contig1310_h4b03a1.f1	544	8.2e-52	146 979	pir JN0588	1,4-ALPHA-GLUCOSIDASE) (1,4-ALPHA-D-GLUCAN GLUCOHYDROLASE) >
y4b12a1.f1	523	1.4e-49	101 661	sp P36914 AMYG_ASPOR	alpha-amylase (EC 3.2.1.1) precursor - <i>Aspergillus</i>
h4b03a1.r1	331	4.4e-29	83 448	sp Q02905 AMYA_ASPAW	oryzae
Contig213_i0g04a1.f1	268	2.4e-21	316 834	sp P22861 AMYG_D	GLUCOAMYLASE PRECURSOR (GLUCAN
z8c10a1.f1	154	6.8e-08	183 527	gi 3420947	1,4-ALPHA-GLUCOSIDASE) (1,4-ALPHA-D-GLUCAN GLUCOHYDROLASE) >
					pir A48305 alpha-amylas
					GLUCOAMYLASE 1 PRECURSOR (GLUCAN
					1,4-ALPHA-GLUCOSIDASE) (1,4-ALPHA-D-GLUCAN GLUCOHYDROLASE) >
					pir JN01
					(AF082188) glucoamylase [<i>Candida albicans</i>]

c. Trehalose degradation

<trehalose-6-phosphate synthase>

Contig1709_c5f08a1.f1	1999	7.2e-260	485 1696	gi 3170246	(AF043230) trehalose-6-phosphate synthase subunit 1
					[<i>Emericellanidulans</i>]
Contig1789_c7d11a1.f1	761	8.5e-75	704 1780	gnl PID e339278	(Z98850) hypothetical alpha-trehalose-phosphate
					synthase[<i>Schizosaccharomyces pombe</i>] >gnl PID e1314275

<trehalase>

Contig1448_f5f04a1.f1	998	6.3e-100	291 845	gi 2827392	(AF043229) neutral trehalase [<i>Emericella nidulans</i>]
r3b05a1.r1	298	1.4e-33	392 580	gi 2827392	(AF043229) neutral trehalase [<i>Emericella nidulans</i>]

12.fatty acid degradation (35)

a. lipase-triacylglycerols to glycerol+FA

r5h04a1.r1	203	2.4e-25	68 304	gnl PID d1033240	(AB010810) phospholipase D [<i>Candida albicans</i>]
c7c07a1.f1	224	1.2e-16	322 768	gi 2773042	(AF038440) phospholipase D2 [<i>Homo sapiens</i>]
*Contig1148_e9g05a1.r1	160	1.1e-10	348 608	gi 2853612	(AF034088) lipase [<i>Pseudomonas sp. B11-1</i>]

b. beta-oxydation of fatty acids

<beta-oxidation>

Contig1498_c8d05a1.f1	755	3.1e-74	128 841	pir S54786	multifunctional beta-oxidation protein - <i>Neurospora</i>
				crassa>gi 510867 (X80052)	multifunctional beta-ox
r5d03a1.r1	396	4.3e-35	23 406	pir S54786	multifunctional beta-oxidation protein - <i>Neurospora</i>
				crassa>gi 510867 (X80052)	multifunctio

i. fatty acid activation-thiokinase

<long-chain-fatty-acid-CoA ligase>

o9c08a1.r1	443	4.2e-41	12 527	gnl PID e316918	(Z95556) fadD35 [<i>Mycobacterium tuberculosis</i>]
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<medium-chain acyl-CoA ligase>

*Contig351_g3f05a1.f1	216	3.6e-16	105 416	gi 2650449	(AE001093) medium-chain acyl-CoA ligase (alkK-1)
					[<i>Archaeoglobusfulgidus</i>]

*Contig215_m7d10a1.f1 129 8.8e-07 3 143 gi|2648519 (AE000963) medium-chain acyl-CoA ligase (alkK-5) [Archaeoglobus fulgidus]

<LONG-CHAIN ACYL-COASYNTHETASE>

Contig1122_m1b06a1.r1 590 1.1e-56 13 741 gnl|PID|e1285372 (AL022304) fatty acid coa ligase [Schizosaccharomyces pombe]

Contig1087_o6c03a1.r1 518 4.7e-49 12 1079 sp|P41215|LCFA_H LONG-CHAIN-FATTY-ACID--COA LIGASE 1 (LONG-CHAIN ACYL-COASYNTHETASE 1) (LACS 1) (PALMITOYL-COA LIGASE)

g4g11a1.r1 496 9.7e-47 38 517 sp|P30624|LCF1_YEAST LONG-CHAIN-FATTY-ACID--COA LIGASE 1 (LONG-CHAIN ACYL-COASYNTHETASE 1) (FATTY ACID ACTIVATOR 1)

Contig1303_n2b07a1.r1 340 2.9e-29 203 805 sp|P39518|LCF2_Y LONG-CHAIN-FATTY-ACID--COA LIGASE 2 (LONG-CHAIN ACYL-COASYNTHETASE 2) (FATTY ACID ACTIVATOR 2) >pir||

Contig1701_c6g08a1.f1 179 5.1e-12 505 711 sp|P39002|LCF3_Y LONG-CHAIN-FATTY-ACID--COA LIGASE 3 (LONG-CHAIN ACYL-COASYNTHETASE 3) (FATTY ACID ACTIVATOR 3) >pir||

<fatty acid coa ligase>

Contig1120_m1c06a1.r1 353 1.4e-31 353 937 gi|2648302 (AE000952) 2-hydroxyhepta-2,4-diene-1,7-dioate isomerase (hpcE-2)[Archaeoglobus fulgidus]

<ii.carnitine acetyl transferase>

Contig1254_m6h06a1.r1 1647 1.1e-168 103 1185 gi|2511761 (AF023156) carnitine acetyl transferase Facc [Emericella nidulans]

Contig1138_m8c01a1.f1 427 1.9e-39 332 820 gi|2688966 (AF027979) carnitine acetyl transferase [Magnaporthe grisea]

m8c01a1.r1 268 1e-21 417 671 gi|2688966 (AF027979) carnitine acetyl transferase [Magnaporthe grisea]

<carnitine racemase-d to l form>

h0f12a1.f1 194 9.6e-15 224 649 gnl|PID|e326890 (Z97336) carnitine racemase homolog [Arabidopsis thaliana]

<iii.acyl-CoA dehydrogenase>

m8b09a1.r1 394 6.3e-36 168 719 gi|2736364 (AF039038) Similar to acyl-coA dehydrogenase; coded for by C.elegans cDNA yk335a7.3; coded

r2g12a1.f1 292 4e-25 192 617 gnl|PID|e306530 (Z92770) fadE2 [Mycobacterium tuberculosis]

Contig976_m8b09a1.f1 292 4.2e-25 314 724 gi|2736364 (AF039038) Similar to acyl-coA dehydrogenase; coded for by C.elegans cDNA yk335a7.3; coded for by C.

<iv.enoyl-CoA hydratase>

Contig1024_n0g10a1.r1 485 1.5e-45 76 783 gi|755067 (L39265) enoyl CoA hydratase [Rhizobium meliloti]

<v.3-hydroxyacyl-CoA dehydrogenase>

Contig747_q0a04a1.r1 205 8.4e-15 115 498 gnl|PID|e1294492 (AL023702) fatty acid oxidation complex alpha-subunit[Streptomyces coelicolor]

<vi.HYDROXYBUTYRYL-COA DEHYDROGENASE>

Contig70_m0d04a1.f1 405 4.4e-37 91 471 sp|P45856|MMGB_B PROBABLE 3-HYDROXYBUTYRYL-COA DEHYDROGENASE(BETA-HYDROXYBUTYRYL-COA DEHYDROGENASE) (BHBD) >gi|881605

<vii.3-ketoacyl-CoA thiolase>

c6b01a1.r1 517 5.8e-49 86 655 sp|Q05493|THIK_YARLI 3-KETOACYL-COA THIOLASE PEROXISOMAL PRECURSOR(BETA-KETOTHIOLASE) (ACETYL-COA ACYLTRANSFERASE)
 r2e10a1.f1 461 5.2e-43 128 610 pir|JT0551 acetyl-CoA C-acyltransferase (EC 2.3.1.16), peroxisomal - rat>gi|205049 (M32801) 3-ketoacyl-coa thiolase
 c6b01a1.f1 394 6e-36 130 483 sp|P07871|THI1_RAT 3-KETOACYL-COA THIOLASE PEROXISOMAL B PRECURSOR(BETA-KETOTHIOLASE B) (ACETYL-COA ACYLTRANS

c. odd chain fatty acids

<methylmalonyl carboxylase-also ile, thr, met, val degradation>

c9h06a1.f1 153 5.3e-10 151 351 gnl|PID|d1031330 (AP000005) 149aa long hypothetical methylmalonyl-CoA decarboxylase gamma chain [Pyrococcus

d. Unsaturated fatty acid degradation

<fatty acyl-CoA reductase>

c6g04a1.r1 184 5.4e-12 124 438 gi|1684886 (U77680) fatty acyl-CoA reductase [Acinetobacter calcoaceticus]

e. Branch chain fatty acid degradation

<branched-chain enoyl CoA reductase>

Contig556_c6b03a1.f1 169 8.9e-10 145 642 gi|2407655 (AF019136) 2-methyl branched-chain enoyl CoA reductase isoform I [Ascaris suum]

f. Ketone body metabolism

<3-OXOACID COA-TRANSFERASE>

Contig219_i0c10a1.r1 716 4.6e-70 5 748 sp|P55809|SCOT_H SUCCINYL-COA:3-KETOACID-COENZYME A TRANSFERASE PRECURSOR(SUCCINYL COA:3-OXOACID COA-TRANSFERASE) (OXC
 Contig750_q0b02a1.f1 218 1.9e-16 180 401 sp|Q09450|SCOT_C PROBABLE SUCCINYL-COA:3-KETOACID-COENZYME A TRANSFERASE PRECURSOR (3-OXOACID COA-TRANSFERASE) >gi|6656

<hydroxybutyrate dehydrogenase>

<ACETOACETYL-COA THIOLASE-acetyl-coA to acetoacyl-coA>

w8a06a1.r1 441 6.2e-41 21 503 sp|P17764|THIL_RAT ACETYL-COA ACETYLTRANSFERASE PRECURSOR, MITOCHONDRIAL(ACETOACETYL-COA THIOLASE) >pir||XXRT
 w8a06a1.f1 310 4.9e-27 97 411 sp|P17764|THIL_RAT ACETYL-COA ACETYLTRANSFERASE PRECURSOR, MITOCHONDRIAL(ACETOACETYL-COA THIOLASE) >pir||XXRT
 x3h11a1.r1 260 2.2e-20 198 569 sp|P17764|THIL_RAT ACETYL-COA ACETYLTRANSFERASE PRECURSOR, MITOCHONDRIAL(ACETOACETYL-COA THIOLASE) >pir||XXRT

<SUCCINYL-COA:3-KETOACID-COENZYME A TRANSFERASE-acetoacetate to acetoacyl-coA>

13. Metabolism of other energy sources (27)

a. alcohol dehydrogenases

<FORMALDEHYDE DEHYDROGENASE-long chain primary alcohol to aldehyde or ketone>

Contig1273_h4ci0a1.f1 661 3.2e-64 7 471 sp|Q06099|FADH_C GLUTATHIONE-DEPENDENT FORMALDEHYDE DEHYDROGENASE (FDH)(FALDH) >pir||JN0447 FDH1 protein - yeast (Cand
 j9h02a1.r1 539 2.5e-51 137 484 sp|Q06099|FADH_CANMA GLUTATHIONE-DEPENDENT FORMALDEHYDE DEHYDROGENASE (FDH)(FALDH) >pir||JN0447 FDH1 protein -
 Contig1690_e9a05a1.f1 484 1.9e-45 178 1086 sp|P47734|FADH_M GLUTATHIONE-DEPENDENT FORMALDEHYDE DEHYDROGENASE (FDH)(FALDH) >gi|496118 (L33464) alcohol dehydrogena
 Contig1186_e9a05a1.r1 198 1.7e-14 267 497 sp|P47734|FADH_M GLUTATHIONE-DEPENDENT FORMALDEHYDE DEHYDROGENASE (FDH)(FALDH) >gi|496118 (L33464) alcohol dehydrogena

<aldehyde reductase>

Contig879_r4e05a1.f1 507 6.9e-48 138 902 sp|P47137|YJ66_Y PROBABLE OXIDOREDUCTASE YJR096W >pir||S57117 aldehydereductase homolog YJR096w - yeast (Saccharomyces
 *Contig770_s9g09a1.f1 471 4.4e-44 118 831 sp|P23901|ALDR_H ALDOSE REDUCTASE (AR) (ALDEHYDE REDUCTASE) >pir||S15024aldose reductase-related protein - barley >gi|
 e4b11a1.r1 341 2.3e-30 25 564 gi|1142698 (U26463) NADPH-dependent aldehyde reductase [Sporidiobolussalmonicolor]
 Contig599_c4a06a1.r1 161 1.3e-20 195 422 sp|P47137|YJ66_Y PROBABLE OXIDOREDUCTASE YJR096W >pir||S57117 aldehydereductase homolog YJR096w - yeast (Saccharomyces
 Contig1561_h1h04a1.f1 175 2.5e-12 295 615 gi|1142698 (U26463) NADPH-dependent aldehyde reductase [Sporidiobolussalmonicolor]

b. GLYCEROL

<glycerol>

*Contig48_w8h05a1.f1 119 9.3e-06 201 410 gnl|PID|d1029223 (AB004569) glycerol kinase [Thermus aquaticus flavus]
 <glycerol-3-phosphatase>
 Contig1355_alb02c9.r1 294 2.4e-25 185 610 sp|P41277|GPP1_Y (DL)-GLYCEROL-3-PHOSPHATASE 1 >gnl|PID|d1009695 (D50471)unknown [Saccharomyces cerevisiae]
 Contig1_e0h11a1.r1 208 3.4e-16 120 578 sp|P41277|GPP1_Y (DL)-GLYCEROL-3-PHOSPHATASE 1 >gnl|PID|d1009695 (D50471)unknown [Saccharomyces cerevisiae]

<glycerol-3-phosphate dehydrogenase>

Contig1076_r5a09a1.r1 384 6e-45 288 806 sp|P41911|GPD2_Y GLYCEROL-3-PHOSPHATE DEHYDROGENASE (NAD+) 2 >pir||S61719glycerol-3-phosphate dehydrogenase (NAD+) (EC
 v3f01a1.r1 145 8.8e-09 9 116 sp|P21696|GPDA_SCHPO GLYCEROL-3-PHOSPHATE DEHYDROGENASE (NAD+), CYTOPLASMIC(GPD-C) (GPDH-C) >gi|4952 (X56162) g

c. proprionate

<PRPD PROTEIN>

i0h10a1.f1 309 8.8e-27 108 506 sp|P77243|PRPD_ECOLI PRPD PROTEIN >gi|1657530 (U73857) similar to yqiP of B.subtilis [Escherichia coli] >gi|178

d. other

<diaminobutyrate decarboxylase-dia-butyrate to dia-propane>
 Contig933_o6g06a1.f1 135 1.5e-05 602 835 gi|1573971 (U32776) L-2,4-diaminobutyrate decarboxylase
 [Haemophilus influenzae Rd]

<ACETAMIDASE-allows acetamide and formamide as sole C or N source>
 Contig1638_c1h08a1.f1 170 3.2e-09 15 443 pir||JS0633 amidase (EC 3.5.1.4) - Aspergillus oryzae
 >gnl|PID|d1001845(D10492) acetamidase [Aspergillus oryzae]
 p0e07a1.r1 146 1.3e-08 171 449 sp|P08158|AMDS_EMENI ACETAMIDASE >pir||A26511 amds protein -
 Emericellanidulans >gi|168015 (M16371) acetamidase

<formamidase>
 Contig978_c8h05a1.f1 606 9.6e-104 430 1011 gnl|PID|e256826 (X99632) formamidase [Methylophilus methylotrophus]
 <acetate>
 z4e02a1.r1 909 1.7e-90 13 528 gi|2262191 (U56097) acetate regulatory DNA binding protein FacB
 [Emericellanidulans]
 w9h04a1.r1 875 7e-87 9 509 gi|1130507 (L41670) fumarylacetoacetate hydrolase [Emericella
 nidulans]
 w9h04a1.f1 631 5.2e-61 65 457 gi|1130507 (L41670) fumarylacetoacetate hydrolase [Emericella
 nidulans]
 l5g04a1.f1 355 3.5e-43 209 439 gnl|PID|e1249838 (AJ001836) maleylacetoacetate isomerase [Emericella
 nidulans]>gnl|PID|e1249842 (AJ001837)
 n0c08a1.r1 301 4.1e-26 24 455 gnl|PID|e1285330 (AJ004870) acetate kinase
 [Thermoanaerobacteriumthermosaccharolyticum]

<ALDEHYDE DEHYDROGENASE-broad substrate specificity>
 Contig1813_alh01f2.f1 2420 1.1e-250 152 1639 sp|P08157|DHAL_E ALDEHYDE DEHYDROGENASE (ALDDH) >pir||A29055
 aldehydedehydrogenase (NAD+) (EC 1.2.1.3) - Emericella ni
 Contig147_j4g02a1.r1 663 2e-64 41 859 sp|P38067|YBL6_Y HYPOTHETICAL ALDEHYDE-DEHYDROGENASE LIKE PROTEIN
 INCOQ1-HHF1 INTERGENIC REGION >pir||s45858 probable
 Contig521_c8e01a1.r1 569 1.7e-54 176 1141 sp|P33008|DHAL_P PROBABLE ALDEHYDE DEHYDROGENASE >pir||s27652
 aldehydedehydrogenase - Pseudomonas sp >gi|151586 (M9144
 c4h09a1.f1 423 5.5e-39 46 540 sp|P54885|PROA_YEAST GAMMA-GLUTAMYL PHOSPHATE REDUCTASE
 (GPR)(GLUTAMATE-5-SEMIALDEHYDE DEHYDROGENASE)(GLUTAMYL-
 c4h09a1.r1 349 3.9e-31 136 642 sp|P54885|PROA_YEAST GAMMA-GLUTAMYL PHOSPHATE REDUCTASE
 (GPR)(GLUTAMATE-5-SEMIALDEHYDE DEHYDROGENASE)(GLUTAMYL-
 n1c01a1.r1 276 1.8e-23 91 426 gi|1399099 (U44901) aspartate semialdehyde dehydrogenase
 [Ustilago maydis]
 z1g07a1.r1 150 4e-09 311 490 sp|P25526|GABD_ECOLI SUCCINATE-SEMIALDEHYDE DEHYDROGENASE (NADP+)
 (SSDH)>gi|147901 (M88334) succinic semialdehy

14. Electron transport (78)

a. Complex I-NADH-ubiquinone

<NADH dehydrogenase>

f0b07a1.r1 619 9.1e-60 49 615 gnl|PID|e1227831 (AJ001520) 19.3kD iron-sulfur subunit of
 mitochondrialcomplex I [Neurospora crassa]

g8f09a1.r1	529	3.2e-50	65 601	sp Q02854 NUXM_NEUCR NADH-UBIQUINONE OXIDOREDUCTASE 21 KD SUBUNIT (COMPLEX I-21KD) (CI-21KD) >pir s27171 NADH d
o8h07a1.f1	488	6.6e-46	203 673	sp P21976 NUPM_NEUCR NADH-UBIQUINONE OXIDOREDUCTASE 20.8 KD SUBUNIT >gi 168781(M55323) complex I 22-kDa polypep
j4e05a1.f1	489	1.3e-45	212 652	sp P24918 NUAM_NEUCR NADH-UBIQUINONE OXIDOREDUCTASE 78 KD SUBUNIT PRECURSOR(COMPLEX I-78KD) (CI-78KD) >pir s17=NADH dehydrogenase
o9g03a1.f1	483	2.5e-45	86 568	sp Q08822 ETFD_YEAST PROBABLE ELECTRON TRANSFER FLAVOPROTEIN-UBIQUINONE OXIDOREDUCTASE PRECURSOR (ETF-QO) (ETF-U
m7e03a1.r1	360	4.6e-32	14 466	gnl PID e349376 (Z99260) ubiquinone reductase [Schizosaccharomyces pombe]
15b07a1.f1	354	1.1e-31	362 598	sp P22142 NUCM_NEUCR NADH-UBIQUINONE OXIDOREDUCTASE 49 KD SUBUNIT PRECURSOR(COMPLEX I-49KD) (CI-49KD) >pir s13
Contig506_c9f11a1.f1	343	1.6e-30	93 503	sp P25710 NUJM_N NADH-UBIQUINONE OXIDOREDUCTASE 21.3 KD SUBUNIT>pir s14277 NADH dehydrogenase (ubiquinone) (EC 1.6.5.
j4e05a1.r1	341	2.3e-29	9 341	pir s59926 NADH dehydrogenase (ubiquinone) (EC 1.6.5.3) 78K chain precursor -Neurospora crassa >gi 55
z7e09a1.r1	294	2.5e-25	69 404	sp P24919 NUFM_NEUCR NADH-UBIQUINONE OXIDOREDUCTASE 29.9 KD SUBUNIT PRECURSOR(COMPLEX I-29.9KD) (CI-29.9KD) >pi
Contig1077_o6a09a1.r1	264	3.8e-22	117 347	pir s47150 NADH dehydrogenase (ubiquinone) (EC 1.6.5.3) 14K chain -Neurospora crassa >gi 499315 (Z18945) NADH:ub
Contig317_g5h08a1.f1	264	1.4e-21	203 607	sp P53318 COQ6_Y UBIQUINONE BIOSYNTHESIS MONOOXYGENASE COQ6 >pir s64587hypothetical protein YGR255c - yeast (Saccharom
w8g05a1.r1	239	1.5e-19	219 419	sp P24919 NUFM_NEUCR NADH-UBIQUINONE OXIDOREDUCTASE 29.9 KD SUBUNIT PRECURSOR(COMPLEX I-29.9KD) (CI-29.9KD) >pi
Contig1249_m6a09a1.r1	238	1.6e-18	48 278	gnl PID e349376 (Z99260) ubiquinone reductase [Schizosaccharomyces pombe]
g5h08a1.r1	200	7.4e-13	30 518	sp P53318 COQ6_YEAST UBIQUINONE BIOSYNTHESIS MONOOXYGENASE COQ6 >pir s64587hypothetical protein YGR255c - yeast
Contig813_w8f02a1.f1	153	2.3e-10	145 378	sp Q02369 NI2M_B NADH-UBIQUINONE OXIDOREDUCTASE B22 SUBUNIT (COMPLEX I-B22) (CI-B22) >pir s28256 NADH dehydrogenase (u

b. Complex II-Succinate-ubiquinone
<succinate dehydrogenase>

c. Complex III-Ubiquinone to cytochrome C
<cytochrome b>

Contig715_p0e03a1.r1	755	3.1e-74	10 489	sp P00161 CYB_EM CYTOCHROME B >pir CBASN ubiquinol--cytochrome-c reductase(EC 1.10.2.2) cytochrome b - Emericella nid
p0e03a1.f1	727	3e-71	24 464	sp P00161 CYB_EMENI CYTOCHROME B >pir CBASN ubiquinol--cytochrome-c reductase(EC 1.10.2.2) cytochrome b - Eme
h0a08a1.f1	244	4.9e-20	434 649	sp P40312 CYB5_YEAST CYTOCHROME B5 >pir s63052 cytochrome b5 - yeast(Saccharomyces cerevisiae) >gnl PID e22183
j4c03a1.f1	234	5.7e-19	283 531	sp P38626 NC5R_YEAST PUTATIVE NADH-CYTOCHROME B5 REDUCTASE (P35)

Contig1096_m5d06a1.f1 186 6.9e-14 171 428 >pir||S49935cytochrome-b5 reductase (EC 1.6.2.
 Contig20_m2d10a1.f1 183 1.4e-13 177 407 prf||751845A cytochrome b2 1-103 [Saccharomycetales]
 x3a05a1.f1 163 4.6e-11 186 314 prf||751845A cytochrome b2 1-103 [Saccharomycetales]
 g9e09a1.f1 175 7.6e-11 316 606 sp|P36060|MCR1_YEAST NADH-CYTOCHROME B5 REDUCTASE PRECURSOR (P34 /
 P32)>pir||S37800 cytochrome-b5 reductase (EC
 z8c03a1.r1 156 9.4e-08 190 477 sp|P36060|MCR1_YEAST NADH-CYTOCHROME B5 REDUCTASE PRECURSOR (P34 /
 P32)>pir||S37800 cytochrome-b5 reductase (EC
 Contig254_h1a12a1.f1 107 1.7e-05 1 87 sp|P00175|CYB2_YEAST CYTOCHROME B2 PRECURSOR (L-LACTATE
 <cytochrome c> DEHYDROGENASE(CYTOCHROME)) (L-LACTATE FERRICYTOCHROME C
 Contig531_o4f04a1.r1 602 4.5e-58 71 409 gi|2062405 (U79011) cytochrome b5 [Borago officinalis]
 Contig723_u4g07a1.r1 451 5.4e-42 13 444 sp|P38091|CYC_EM CYTOCHROME C >gi|1899007 (M83141) cytochrome c
 {Emericellanidulans}
 Contig477_d3b01a1.f1 317 8.9e-28 397 711 sp|P32891|DLD1_Y D-LACTATE DEHYDROGENASE (CYTOCHROME) PRECURSOR
 (D-LACTATEFERRICYTOCHROME C OXIDOREDUCTASE) (D-LCR) >p
 Contig732_x7b08a1.f1 288 9.9e-25 222 419 sp|P04037|COX4_Y CYTOCHROME C OXIDASE POLYPEPTIDE IV PRECURSOR
 >pir||OLBY4cytochrome-c oxidase (EC 1.9.3.1) chain IV p
 Contig175_m6h01a1.f1 213 8.9e-17 259 588 sp|Q01519|COXG_Y CYTOCHROME C OXIDASE POLYPEPTIDE VIB (AED)
 >pir||S31256cytochrome-c oxidase (EC 1.9.3.1) chain VIB -
 Contig640_c0e03a1.f1 212 1.2e-16 224 541 sp|P32799|COXE_Y CYTOCHROME C OXIDASE POLYPEPTIDE VIA
 PRECURSOR>pir||A48520 cytochrome-c oxidase (EC 1.9.3.1) chain VI
 Contig963_k5a06a1.f1 176 7.5e-13 377 556 pdb|2CYP| Cytochrome c Peroxidase (E.C.1.11.1.5)
 (Ferrocytochrome c (Colon)H2O2 Reductase)
 o5b10a1.r1 175 1.1e-12 159 308 gnl|PID|e1256442 (AL021767) possible ubiquinol-cytochrome c
 reductasecomponent [Schizosaccharomyces pombe]
 <cytochrome oxidase> sp|Q12287|COXS_YEAST CYTOCHROME C OXIDASE ASSEMBLY PROTEIN COX17
 <CBP4 PROTEIN-cytoC, ubiquinol assembly> >pir||S62056COX17 protein - yeast (Saccharomyc
 Contig1400_w9h07a1.f1 121 5.2e-06 248 562 sp|P37267|CBP4_Y CBP4 PROTEIN PRECURSOR >pir||S64488 regulatory
 proteinCBP4 precursor - yeast (Saccharomyces cerevisia)

d.Other electron transport pathways

<NADH OXIDASE>

*Contig1365_o9c07a1.f1 182 2.4e-12 208 660 sp|P32382|NADO_T NADH OXIDASE >pir||S25102 NADH oxidase
 -Thermoanaerobacter brockii >gi|48123 (X67220) NADH oxidase[Th

<NADPH DEHYDROGENASE>

x1h09a1.r1 385 5.5e-35 136 729 gi|2232254 (AF005237) old-yellow-enzyme homolog [Catharanthus
 roseus]

e.Electron carriers

<flavoprotein>

Contig1240_m2d10a1.r1 751 8.3e-74 28 891 gnl|PID|e349663 (Z99292) flavoprotein [Schizosaccharomyces pombe]

Contig64_m0e09a1.r1 402 8.4e-37 1 606 sp|Q10499|YDGE_S PUTATIVE FLAVOPROTEIN C26F1.14C
 Contig50_h8g02a1.r1 325 2.7e-28 104 508 gnl|PID|e349663 (Z99292) flavoprotein [Schizosaccharomyces pombe]
 Contig1250_g7a10a1.r1 172 1.9e-12 64 252 sp|P53575|ETFB_B ELECTRON TRANSFER FLAVOPROTEIN BETA-SUBUNIT
 (BETA-ETF) (ELECTRON TRANSFER FLAVOPROTEIN SMALL SUBUNIT)

<FLAVOHEMOPROTEIN>

i8f11a1.r1 455 2e-42 85 567 sp|P39662|HMPA_ALCEU FLAVOHEMOPROTEIN (HAEMOGLOBIN-LIKE
 PROTEIN) (FLAVOHEMOGLOBIN) >pir||A53396 flavohemoprotein
 w9e03a1.r1 206 1.8e-15 46 327 sp|P24232|HMPA_ECOLI FLAVOHEMOPROTEIN (HAEMOGLOBIN-LIKE
 PROTEIN) (FLAVOHEMOGLOBIN) (DIHYDROPTERIDINE REDUCTASE (

<QUINONE>

w8a03a1.r1 361 2e-32 4 531 sp|P38230|QOR_YEAST PROBABLE QUINONE OXIDOREDUCTASE (NADPH:QUINONE
 REDUCTASE)>pir||S45904 quinone oxidoreducta

f. Component enzymes and molecules

<FLAVIN OXIDOREDUCTASE>

Contig1495_d3e02a1.f1 835 1.1e-82 53 598 sp|Q00415|HPPD_C 4-HYDROXYPHENYLPYRUVATE DIOXYGENASE (4HPPD) (HPD)
 (T-CELLREACTIVE PROTEIN) >gi|601846 (L38493) T-cell
 Contig1360_c5g11a1.r1 622 4.3e-60 58 912 sp|Q00415|HPPD_C 4-HYDROXYPHENYLPYRUVATE DIOXYGENASE (4HPPD) (HPD)
 (T-CELLREACTIVE PROTEIN) >gi|601846 (L38493) T-cell
 Contig11_y8g03a1.r1 263 4.8e-22 176 490 sp|P54550|YQJM_B PROBABLE NADH-DEPENDENT FLAVIN OXIDOREDUCTASE
 YQJM>gnl|PID|d1013299 (D84432) YqjM [Bacillus subtilis]
 Contig191_m8f03a1.r1 139 3.1e-08 312 476 sp|P54550|YQJM_B PROBABLE NADH-DEPENDENT FLAVIN OXIDOREDUCTASE
 YQJM>gnl|PID|d1013299 (D84432) YqjM [Bacillus subtilis]

<RESPIRATORY complex assembly>

i2e08a1.r1 783 4e-77 10 606 sp|P40341|RCA1_YEAST MITOCHONDRIAL RESPIRATORY CHAIN COMPLEXES
 ASSEMBLYPROTEIN RCA1 (TAT-BINDING HOMOLOG 12) >p

<MITOCHONDRIAL FAD CARRIER PROTEIN>

s9f04a1.f1 247 2.4e-20 208 567 gnl|PID|d1022297 (AB004539) MITOCHONDRIAL FAD CARRIER PROTEINFAD
 CARRIER PROTEIN FLX1[Schizosaccharomyces pombe] >gnl|PID|e125

g. ATP synthase

<ATP SYNTHASE>

y8h10a1.r1 867 4.1e-86 16 540 sp|P23704|ATPB_NEUCR ATP SYNTHASE BETA CHAIN, MITOCHONDRIAL
 PRECURSOR>pir||JC1112 H+-transporting ATP synthase
 k5d06a1.r1 743 6.2e-73 7 504 sp|P22550|VATB_CANTR VACUOLAR ATP SYNTHASE SUBUNIT B (V-ATPASE 57 KD
 SUBUNIT)>pir||S13080 H+-transporting ATPas
 Contig1172_h1f03a1.r1 719 2.4e-70 13 570 sp|P24487|ATPA_S ATP SYNTHASE ALPHA CHAIN, MITOCHONDRIAL
 PRECURSOR>pir||A39036 H+-transporting ATP synthase (EC 3.6.1.
 Contig256_h0h10a1.f1 501 3.3e-62 249 701 sp|P37211|ATPA_N ATP SYNTHASE ALPHA CHAIN, MITOCHONDRIAL
 PRECURSOR>pir||JC1111 H+-transporting ATP synthase (EC 3.6.1.
 Contig763_s8f09a1.f1 613 3.9e-59 321 725 sp|P23704|ATPB_N ATP SYNTHASE BETA CHAIN, MITOCHONDRIAL
 PRECURSOR>pir||JC1112 H+-transporting ATP synthase (EC 3.6.1.3
 Contig1377_e9e12a1.r1 564 5e-54 55 726 sp|O13349|ATPF_K ATP SYNTHASE SUBUNIT 4, MITOCHONDRIAL

j0b09a1.r1	543	1.1e-51	12 425	PRECURSOR>gi 2425071 (AF019222) F1Fo-ATP synthase subunit 4 (K1 sp P11592 VATA_NEUCR VACUOLAR ATP SYNTHASE CATALYTIC SUBUNIT A (V-ATPASE 67 KDSUBUNIT) >pir PXNCV7 H+-transpor
Contig1510_h1f11a1.f1	505	1e-47	88 516	sp P16000 ATPL_E ATP SYNTHASE PROTEIN 9, MITOCHONDRIAL PRECURSOR(LIPID-BINDING PROTEIN) >gi 168019 (M30144) mitochondr
Contig1289_i8d01a1.f1	490	3.4e-46	9 515	sp O13350 ATP7_K ATP SYNTHASE D CHAIN, MITOCHONDRIAL >gi 2425073(AF019223) F1Fo-ATP synthase subunit 7 [Kluyveromyces
w4g11a1.r1	475	1.5e-44	117 488	sp P56525 ATPD_NEUCR ATP SYNTHASE DELTA CHAIN, MITOCHONDRIAL PRECURSOR
g8b10a1.r1	455	2.2e-42	107 514	sp Q01278 VATE_NEUCR VACUOLAR ATP SYNTHASE SUBUNIT E (V-ATPASE E SUBUNIT)(V-ATPASE 26 KD SUBUNIT) >gi 600167 (U
j0b09a1.f1	410	1.9e-37	138 461	sp P11592 VATA_NEUCR VACUOLAR ATP SYNTHASE CATALYTIC SUBUNIT A (V-ATPASE 67 KDSUBUNIT) >pir PXNCV7 H+-transpor
o5f02a1.r1	383	9.2e-35	176 517	sp P39111 VATF_YEAST VACUOLAR ATP SYNTHASE 14 KD SUBUNIT (V-ATPASE F SUBUNIT)>pir A55118 H+-transporting ATPas
Contig647_b0h11a1.f1	296	1.5e-25	166 498	sp P38077 ATPG_Y ATP SYNTHASE GAMMA CHAIN, MITOCHONDRIAL PRECURSOR>pir S55891 H+-transporting ATP synthase (EC 3.6.1.
b0h11a1.r1	268	1.3e-22	13 384	sp P49377 ATPG_KLULA ATP SYNTHASE GAMMA CHAIN, MITOCHONDRIAL PRECURSOR>pir S56153 H+-transporting ATP synthase
Contig433_n8c05a1.f1	211	8.7e-16	178 600	sp P41807 VM13_Y VACUOLAR ATP SYNTHASE 54 KD SUBUNIT (V-ATPASE 54 KDSUBUNIT) >pir A47429 H+-transporting ATPase (EC 3
m8f04a1.r1	135	1.2e-07	17 145	sp P09457 ATPO_YEAST ATP SYNTHASE OLIGOMYCIN SENSITIVITY CONFERRAL PROTEINPRECURSOR, MITOCHONDRIAL (OSCP) (ATP
Contig1686_a5b12a1.f1	113	7.3e-06	142 291	pir S53404 ATP synthase regulatory factor homolog YLR327c - yeast(Saccharomyces cerevisiae) >gi 662125 (U20618)
<PLASMA MEMBRANE ATPASE (PROTON PUMP)>				
Contig1349_c5b10a1.f1	1142	2.9e-115	27 737	gi 3366659 (AF036763) P-ATPase [Emericella nidulans]
m7e01a1.r1	755	3.6e-74	30 590	gi 3366659 (AF036763) P-ATPase [Emericella nidulans]=(PROTON PUMP)
Contig1421_c8b08a1.f1	542	1.2e-51	312 815	gi 2197050 (AF001033) putative 20kDa subunit of the V-ATPase [Neurospora crassa]
q0g10a1.f1	546	1.7e-51	4 504	gi 1237128 (U36396) vacuolar ATPase 98 kDa subunit [Neurospora crassa]-H+-transporting
n3f03a1.r1	459	6.4e-42	30 401	sp P49380 PMA1_KLULA PLASMA MEMBRANE ATPASE (PROTON PUMP) >gi 598435 (L37875)proton-ATPase [Kluyveromyces lacti
Contig757_v1e09a1.r1	448	1.3e-41	7 480	sp P31413 VATL_N VACUOLAR ATP SYNTHASE 16 KD PROTEOLIPID SUBUNIT>pir S43893 H+-transporting ATPase (EC 3.6.1.35) lipi
n3f03a1.f1	438	1.4e-39	204 773	sp Q07421 PMA1_AJECA PLASMA MEMBRANE ATPASE (PROTON PUMP) >gi 409249 (L07305)ATPase [Ajellomyces capsulatus] >p
Contig962_j7d08a1.f1	392	1.4e-34	24 470	gi 3366659 (AF036763) P-ATPase [Emericella nidulans]
d2c08a1.f1	324	1.7e-28	352 759	gi 1208770 (U48365) V-type ATPase 16 kDa proteolipid subunit [Pleurochrysis carterae] >gi 2149129 (U81
q0g10a1.r1	329	5.7e-28	10 495	gi 1237128 (U36396) vacuolar ATPase 98 kDa subunit [Neurospora crassa]

h. Alternative respiratory path

<ALTERNATIVE OXIDASE>

Contig340_g4b11a1.f1 778 1.4e-76 1 735 gnl|PID|d1032995 (AB016540) alternative oxidase [Aspergillus niger]

15. Reducing carriers (8)

<a. glutaredoxin>

Contig1357_alb04c9.r1 276 2e-23 130 426 gi|3249567 (AF047694) glutaredoxin [Vernicia fordii]

*Contig1556_c9f10a1.r1 239 1.6e-19 137 448 gi|3335374 (AC003028) glutaredoxin-like protein [Arabidopsis thaliana]

<b. glutathione>

<gamma-glutamyl transpeptidase-synthesis and deg of glutathione>

r4h11a1.r1 471 4.3e-44 46 822 sp|P19440|GGT1_HUMAN GAMMA-GLUTAMYLTRANSPEPTIDASE 1
PRECURSOR(GAMMA-GLUTAMYLTRANSFERASE 1) >pir||EKHUEXgamma-gl

<glutathione S-transferase-reduces peroxides, reducing agent>

Contig1732_c8d03a1.f1 133 5.7e-08 256 654 gi|2583081 (AF026977) microsomal glutathione S-transferase 3
[Homo sapiens]

*Contig1246_v3f09a1.r1 136 1.6e-06 208 426 sp|P04907|GTH3_M GLUTATHIONE S-TRANSFERASE III (GST-III) (CLASS
PHI)>pir||XUZM32 glutathione transferase (EC 2.5.1.18)

<GLUTATHIONE PEROXIDASE>

Contig1403_c7g02a1.f1 518 4.7e-49 231 719 sp|P38143|GSHI_Y GLUTATHIONE PEROXIDASE HOMOLOG YBR244W
>pir||S46121probable glutathione peroxidase (EC 1.11.1.9) - ye

<c. thioredoxin>

Contig1775_d1a07a1.r1 560 1.6e-53 269 595 sp|P29429|THIO_E THIOREDOXIN >pir||S27053 thioredoxin -
Emericellanidulans >bbs|120057 thioredoxin [Aspergillus nidula

j9b04a1.r1 515 8.8e-49 50 484 sp|P18408|MT16_YEAST PHOSPHOADENOSINE PHOSPHOSULFATE REDUCTASE
(PAPSREDUCTASE, THIOREDOXIN DEPENDENT) (PADOPS R

j9b04a1.f1 255 3.5e-21 350 583 sp|P18408|MT16_YEAST PHOSPHOADENOSINE PHOSPHOSULFATE REDUCTASE
(PAPSREDUCTASE, THIOREDOXIN DEPENDENT) (PADOPS R

II. Cell Growth, Cell Division

A. Cell walls (12)

<septin>

Contig1351_c5d08a1.r1 1174 1.4e-118 226 1284 sp|P48009|SPN4_S SEPTIN HOMOLOG SPN4 >gi|987283 (U29890) septin
homolog[Schizosaccharomyces pombe] >gnl|PID|e1168676 (

Contig447_d5d02a1.r1 1173 1.8e-118 15 773 gi|1791305 (U83489) septin B [Emericella nidulans]

d5d02a1.f1 352 1.7e-31 247 480 gi|1791305 (U83489) septin B [Emericella nidulans]

<RODLET PROTEIN-spore-wall fungal hydrophobin>

Contig1836_alb02f2.f1 545 5.8e-52 90 560 sp|P28346|RODL_E RODLET PROTEIN PRECURSOR >pir||A40323 Rodletless

Contig1791_c6e09a1.f1	355	8.5e-32	975 1325	protein- Emericella nidulans >gi 168086 (M61113) rod sp P28346 RODL_E RODLET PROTEIN PRECURSOR >pir A40323 Rodletless
Contig971_k0g04a1.r1	165	1.2e-11	6 101	protein- Emericella nidulans >gi 168086 (M61113) rod sp P28346 RODL_E RODLET PROTEIN PRECURSOR >pir A40323 Rodletless
Contig1330_g6b08a1.f1	162	2.5e-11	213 308	protein- Emericella nidulans >gi 168086 (M61113) rod sp P28346 RODL_E RODLET PROTEIN PRECURSOR >pir A40323 Rodletless
<SPORE-WALL FUNGAL HYDROPHOBIN-not rodlet>				
Contig1806_b0d04a1.r1	381	1.3e-34	75 479	sp P52750 DEWA_E SPORE-WALL FUNGAL HYDROPHOBIN DEWA PRECURSOR >pir S67924spore-wall fungal hydrophobin DewA - Emeric
Contig1711_b0c12a1.f1	381	1.4e-34	175 579	sp P52750 DEWA_E SPORE-WALL FUNGAL HYDROPHOBIN DEWA PRECURSOR >pir S67924spore-wall fungal hydrophobin DewA - Emeric
Contig1863_ala04f2.f1	381	1.4e-34	186 590	sp P52750 DEWA_E SPORE-WALL FUNGAL HYDROPHOBIN DEWA PRECURSOR >pir S67924spore-wall fungal hydrophobin DewA - Emeric
<INTEGRIN ALPHA CHAIN-LIKE PROTEIN-cell adhesion>				
g4g04a1.r1	257	8.6e-20	96 560	sp P53705 INT1_CANAL INTEGRIN ALPHA CHAIN-LIKE PROTEIN (ALPHA-INT1)>gi 1144531 (U35070) integrin-like protein a
g7a09a1.r1	159	9.9e-08	235 447	sp P53705 INT1_CANAL INTEGRIN ALPHA CHAIN-LIKE PROTEIN (ALPHA-INT1)>gi 1144531 (U35070) integrin-like protein a=C. albicans
B. Biomembranes (see also D,3.c biomembrane precursors),Cytoskeleton, organelle biogenesis (40)				
<peroxisom>				
Contig1486_r2g02a1.f1	315	1.4e-27	130 588	gnl PID e1169881 (AJ002536) PMP20=PEROXISOMAL MEMBRANE PROTEIN [Schizosaccharomyces pombe]
w7h12a1.r1	184	7.8e-25	357 521	sp P78723 PEXE_PICAN PEROXISOMAL MEMBRANE PROTEIN PER10 (PEROXIN-14)>gi 1777749 (U46195) Per10p [Pichia angusta
Contig1043_d2h01a1.r1	251	9.8e-21	248 541	gnl PID e1132728 (Z99167) putative peroxisomal organisation and biogenesisprotein [Schizosaccharomyces pombe]
Contig866_y6h08a1.f1	176	4.4e-12	3 203	sp P21245 P47A_C PEROXISOMAL MEMBRANE PROTEIN PMP47A >pir A23667 47Kperoxisomal membrane protein - yeast (Candida boi
Contig1058_o0h05a1.f1	142	3.2e-09	292 435	gi 2769700 (U58050) peroxisomal-like protein [Aspergillus fumigatus]
Contig194_i3c05a1.r1	157	3.3e-09	37 564	sp Q12462 PEXB_Y PEROXISOMAL MEMBRANE PROTEIN PMP27 (PEROXIN-11)>pir A56509 peroxisomal membrane protein PMP27 - yeas
Contig1023_d1a12a1.f1	137	4.7e-06	12 218	gi 2992543 (AF021797) peroxisomal receptor for PTS2-containing proteins Pex7p[Pichia pastoris]
<KINESIN>				
x1d12a1.r1	562	1.3e-52	19 348	sp P17120 BIMC_EMENI KINESIN-LIKE PROTEIN BIMC >pir A34795 kinesin-relatedprotein bimc - Emericella nidulans >
Contig755_q0b12a1.f1	309	6.9e-26	353 538	sp P28739 KLPA_E KINESIN-LIKE PROTEIN KLPA >pir A44337 kinesin-relatedprotein KLPA - Emericella nidulans >gi 2704 (X6
<tubulin>				
r3f05a1.f1	484	1.9e-45	3 290	sp P18695 TBG_EMENI TUBULIN GAMMA CHAIN >pir S03916 tubulin gamma chain

c6h05a1.r1	280	1.5e-23	23 184	-Emericella nidulans >gi 2363 (X15479 sp P24633 TBA1_EMENI TUBULIN ALPHA-1 CHAIN >pir S13336 tubulin alpha-1 chain- Emericella nidulans
m5g06a1.r1	196	5e-12	128 319	sp P18695 TBG_EMENI TUBULIN GAMMA CHAIN >pir S03916 tubulin gamma chain -Emericella nidulans >gi 2363 (X15479
<ankyrin>				
c8f04a1.f1	274	3.5e-23	151 711	gi 2447128 (U42580) contains 10 ankyrin-like repeats; similar to humanankyrin, corresponds to Swiss-P
c3c08a1.r1	261	1.2e-20	18 617	gi 1841966 (U65916) ankyrin [Rattus norvegicus]
<VACUOLAR ASSEMBLY PROTEIN VPS39>				
m6b12a1.r1	146	1.8e-06	260 637	sp Q07468 VP39_YEAST VACUOLAR ASSEMBLY PROTEIN VPS39 (VACUOLAR MORPHOGENESIS PROTEIN VAM6) >pir S67613 probable
<cytoskeleton assembly control protein>				
Contig1107_c6c12a1.f1	259	1.8e-28	216 563	pir S63211 cytoskeleton assembly control protein SLA2 - yeast (Saccharomyces cerevisiae) >gnl PID e239710 (Z71519=TRANSMEMBRANE PROTEIN MOP2
1.actin-see also mitosis				
<actin>				
m8d09a1.r1	916	3.1e-91	29 664	sp P78712 ARP3_NEUCR ACTIN-LIKE PROTEIN 3 >gi 1718497 (U79737) actin-related protein 3 [Neurospora crassa]
z5e10a1.f1	438	7.1e-40	117 659	sp P53946 ARP5_YEAST ACTIN-LIKE PROTEIN ARP5 >pir S58718 probable nuclear protein YNL059c - yeast (Saccharomyces
Contig1684_alc05f2.f1	386	4.6e-35	567 788	sp P20359 ACTG_E ACTIN, GAMMA >pir JT0385 actin gamma - Emericella nidulans >gi 168005 (M22869) gamma-actin [Emericella
Contig42_z5a09a1.f1	121	0.00015	214 366	sp P53459 ACT6_D ACTIN 6 >gi 1098579 (U27837) actin [Diphyllobothrium dendriticum]
<PROFILIN-assembly of actin monomers>				
*Contig1634_c8a05a1.f1	243	5.5e-28	1287 1559	pdb 1HLU A Chain A, Structure Of Bovine Beta-Actin-Profilin Complex With Actin Bound Atp Phosphates Solvent Acces
Contig1788_g5b09a1.f1	283	3.6e-24	181 546	sp P39825 PROF_S PROFILIN >pir A53952 profilin - fission yeast (Schizosaccharomyces pombe)
<ARP2/3 COMPLEX-actin polymerization>				
Contig1692_c4c11a1.f1	813	2.6e-80	174 1052	sp O14241 AR34_S PROBABLE ARP2/3 COMPLEX 34 KD SUBUNIT (P34-ARC) >gnl PID e339970 (Z98981) probable Arp2-3 complex subu
m6f08a1.r1	617	1.5e-59	60 563	sp O15509 AR20_HUMAN ARP2/3 COMPLEX 20 KD SUBUNIT (P20-ARC) >gi 2282040(AF006087) p20-Arc [Homo sapiens] >gi 24
Contig495_olb04a1.f1	499	5e-47	25 582	sp P78774 AR41_S PROBABLE ARP2/3 COMPLEX 41 KD SUBUNIT (P41-ARC)
c0h11a1.r1	464	2.3e-43	98 484	sp P32381 ARP2_YEAST ACTIN-LIKE PROTEIN ARP2 >pir S20225 actin-like protein ACT2 - yeast (Saccharomyces cerevisiae)
Contig726_w6b05a1.f1	181	9.5e-24	116 361	sp O15145 AR21_H ARP2/3 COMPLEX 21 KD SUBUNIT (P21-ARC) >gi 2282038(AF006086) p21-Arc [Homo sapiens]
<fimbrin-actin bundling>				

m6ella1.r1	507	6.8e-48	202 840	sp P32599 FIMB_YEAST FIMBRIN (ABP67) >pir S29320 fimbrin - yeast(Saccharomyces cerevisiae) >gi 4420 (X63867) f
Contig229_h8h08a1.f1	278	1e-22	378 614	sp P32599 FIMB_Y FIMBRIN (ABP67) >pir S29320 fimbrin - yeast(Saccharomyces cerevisiae) >gi 4420 (X63867) fimbrin [Sac
<COFILIN-actin binding>				
Contig998_r7c08a1.f1	248	1.8e-20	358 798	sp Q03048 COFI_Y COFILIN >pir A44397 cofilin - yeast (Saccharomyces cerevisiae) >pdb 1CFY A Saccharomyces cerevisiae >
<ACTIN-BINDING PROTEIN>				
Contig248_h1d05a1.f1	270	8.9e-23	179 625	sp P25229 CAPA_X ACTIN-BINDING PROTEIN CHAIN A (ABP-A) >pir S07105actin-binding protein chain a, nuclear - African cl
2. choline				
<choline dehydrogenase>				
f2b11a1.r1	269	5.8e-22	80 571	gi 1657509 (U73857) choline dehydrogenase [Escherichia coli]
<chitin synthase>				
Contig542_c7b10a1.f1	780	1.7e-89	242 691	pir JC2315 chitin synthase (EC 2.4.1.16) chsB - Emericella nidulans>gnl PID d1005340 (D21269) chitin synthase [E
f5c06a1.f1	246	5.5e-19	268 528	pir JC2408 chitin synthase (EC 2.4.1.16), class I - Emericella nidulans>prf 2102237A chitin synthase
3. other				
<phosphatidylethanolamine methyltransferase>				
Contig1042_o4a10a1.f1	184	6.4e-11	296 721	gi 2209107 (AF004113) phosphatidylethanolamine methyltransferase [Schizosaccharomyces pombe]
<ACETYLGLUCOSAMINE-6-PHOSPHATE DEACETYLASE>				
Contig1645_g2a12a1.f1	351	2.3e-31	249 902	sp P34480 NAGA_C PUTATIVE N-ACETYLGLUCOSAMINE-6-PHOSPHATE DEACETYLASE (GLCNAC 6-P DEACETYLASE) >pir S31124 hypothetica
Contig1139_g3h11a1.r1	348	4.8e-31	442 1053	sp P34480 NAGA_C PUTATIVE N-ACETYLGLUCOSAMINE-6-PHOSPHATE DEACETYLASE (GLCNAC 6-P DEACETYLASE) >pir S31124 hypothetica
<DIACETYLMURAMIDASE>				
Contig1142_g2c04a1.f1	614	3.2e-59	83 679	sp P00721 LYCH_C N,O-DIACETYLMURAMIDASE (LYSOZYME CH) >pir MUKAD lysozyme (EC 3.2.1.17) - fungus (Chalara sp.)
Contig520_o4b02a1.r1	380	4.1e-52	90 407	sp P00721 LYCH_C N,O-DIACETYLMURAMIDASE (LYSOZYME CH) >pir MUKAD lysozyme (EC 3.2.1.17) - fungus (Chalara sp.)
<Glycosyltransferase-glycopeptidolipid biosyn>				
n8d01a1.r1	377	4.2e-34	48 488	gi 871531 (X87947) glycosyltransferase [Saccharomyces cerevisiae]
n8d01a1.f1	249	6.6e-20	165 512	gnl PID d1022233 (AB004534) Glycosyltransferase [Schizosaccharomyces pombe]
C. cell cycle control (17)				
<cell division protein>				
c6f09a1.r1	423	3.4e-58	17 454	gnl PID d1022237 (AB004534) cdc2 kinase homologue [Schizosaccharomyces

					pombe]	
Contig930_c5g03a1.f1	242	8.4e-20	656 1009	sp P51953 CDK7_C	CELL DIVISION PROTEIN KINASE 7 (40 KD PROTEIN KINASE)(P40 MO15) (CDC2/CDK2,4-ACTIVATING KINASE) >gnl	
<CELL DIVISION CONTROL PROTEIN>						
Contig583_c4g06a1.r1	815	1.6e-80	276 1301	sp P53699 CC4_CA	CELL DIVISION CONTROL PROTEIN 4 >gnl PID e234056 (X96763)CDC4 gene product [Candida albicans]	
p0g11a1.r1	709	2.6e-69	7 465	sp P25694 CC48_YEAST	CELL DIVISION CONTROL PROTEIN 48 >pir s67669 celldivision control protein CDC48 - yeast (
o8a05a1.r1	646	1.3e-62	40 606	sp P25694 CC48_YEAST	CELL DIVISION CONTROL PROTEIN 48 >pir s67669 celldivision control protein CDC48 - yeast (
j7e08a1.r1	426	6e-38	40 543	sp P43069 CC25_CANAL	CELL DIVISION CONTROL PROTEIN 25	
Contig1269_u4c02a1.r1	247	6.7e-20	142 507	gnl PID e334053	(Z98533) hypothetical cell division control protein[Schizosaccharomyces pombe]	
<cell cycle protein>						
m6h05a1.r1	480	5.1e-45	345 695	gnl PID d1033372	(AB000281) krev-1 [Neurospora crassa]	
c4f02a1.r1	449	9.3e-42	43 735	sp P50582 HSK1_SCHPO	CELL CYCLE PROTEIN KINASE HSK1 >pir s56143 proteinkinase hsk1 (EC 2.7.1.-) - fission yeas	
d4e12a1.f1	346	7.7e-30	47 517	gnl PID e1291642	(AL023288) AAA family ATPase protein [Schizosaccharomycespombe]	
Contig593_c4d04a1.f1	292	1.3e-24	166 534	sp P50582 HSK1_S	CELL CYCLE PROTEIN KINASE HSK1 >pir s56143 proteinkinase hsk1 (EC 2.7.1.-) - fission yeast (Schizosa	
*Contig997_j7h03a1.f1	126	9.5e-07	316 555	gi 2697005	(U59435) cell cycle protein p38-2G4 homolog [Homo sapiens]	
<SCH9 protein-cell progress through G1>						
Contig554_c6c06a1.f1	212	1.2e-34	295 564	gi 4426	(X12560) SCH9 protein (AA 1-824) [Saccharomyces cerevisiae]	
<G1/S-SPECIFIC CYCLIN-essential for movement from g1-S>						
r5d06a1.r1	152	2.9e-08	423 734	sp P24867 CG16_YEAST	G1/S-SPECIFIC CYCLIN PCL1 (CYCLIN HCS26) >pir A40027cyclin G1 homolog HCS26 - yeast (Sacc	
<cullin-neg regulator of cell cycle>						
f0f02a1.r1	433	2.3e-39	11 592	sp Q13616 CUL1_HUMAN	CULLIN HOMOLOG 1 G1 to G0 phase(CUL-1) >gi 1381142 (U58087) Hs-CUL-1[Homo sapiens]	
<SENESCENCE>						
*Contig1193_x3c04a1.f1	286	1.9e-24	414 950	sp Q64374 SM30_M	SENESCENCE MARKER PROTEIN-30 (SMP-30) (REGUCALCIN) (RC)>gi 1144000 (U28937) senescence marker protein	
<apoptosis>						
Contig890_v7d01a1.f1	200	2.1e-15	382 606	sp P46966 DAD1_H	DEFENDER AGAINST CELL DEATH 1 (DAD-1) >pir A54437apoptotic cell death regulator DAD1 ~ human >gnl PI	

E. Mitosis/cytokinesis (15)

1. MITOSIS <MITOSIS>

c4f05a1.r1 981 4.1e-98 19 567 sp|P30303|MPIP_EMENI M-PHASE INDUCER PHOSPHATASE
>pir||S24395protein-tyrosine-phosphatase (EC 3.1.3.48) nimt -
<DNA DAMAGE CHECKPOINT PROTEIN-allows entry into Mitosis>
*Contig1730_c1a04a1.f1 659 5.6e-85 115 612 sp|P42656|RA24_S DNA DAMAGE CHECKPOINT PROTEIN RAD24
>gnl|PID|e1251047(AL021817) dna damage checkpoint protein [schizo

<centromere>
Contig1160_w5c12a1.f1 938 1.4e-93 333 980 sp|O43100|CBF5_E CENTROMERE/MICROTUBULE BINDING PROTEIN
CBF5(CENTROMERE-BINDING FACTOR 5) (NUCLEOLAR PROTEIN CBF5) >gi
<CHROMOSOME SEGREGATION PROTEIN-with microtubule, migration of chromosomes>
Contig627_c1b12a1.r1 672 1.7e-64 132 704 sp|P38989|SMC2_Y CHROMOSOME SEGREGATION PROTEIN SMC2 (DA-BOX PROTEIN
SMC2)>pir||A56157 chromosome segregation protein
o6g05a1.r1 299 1.2e-24 12 536 sp|P33307|CSE1_YEAST CHROMOSOME SEGREGATION PROTEIN CSE1
>pir||A48083chromosome segregation protein CSE1 - yeas
o6g05a1.f1 197 1e-13 68 679 sp|P33307|CSE1_YEAST CHROMOSOME SEGREGATION PROTEIN CSE1
>pir||A48083chromosome segregation protein CSE1 - yeas
<dynamin-molecular motor, associated with microtubule, endocytosis>
Contig862_r7d07a1.f1 920 1.1e-91 13 708 sp|P21576|VPS1_Y VACUOLAR SORTING PROTEIN 1 >pir||S25820
dynamin-relatedprotein VPS1 - yeast (Saccharomyces cerevisiae
g4c03a1.r1 401 7.2e-36 92 412 sp|P54861|DNM1_YEAST DYNAMIN-RELATED PROTEIN DNMI >pir||S64742
dynamin-relatedprotein DNMI - yeast (Saccharomyc
Contig339_g4c03a1.f1 309 6.7e-26 172 498 sp|Q09748|YB68_S DYNAMIN-LIKE PROTEIN C12C2.08 >gi|984696
(Z54140)dynamin-related protein [Schizosaccharomyces pombe]

<dynein-molecular motor>
Contig171_m6g04a1.f1 123 7.8e-07 310 405 gnl|PID|e326960 (Z97340) strong similarity to dynein light chain
[Arabidopsisthaliana]

<nuclear positioning>
e7d01a1.r1 927 1.9e-92 24 620 gnl|PID|e1286087 (AJ003163) apsB [Emericella nidulans]
<DMR-N9 PROTEIN-regulation of mitosis>
k5h10a1.r1 294 1.2e-24 9 497 sp|Q09019|DMR9_HUMAN DMR-N9 PROTEIN (PROTEIN 59) >pir||A49364 59
protein,brain - human (fragment) >gi|306712 (L
<CALTRACTIN-ASSOCIATED WITH THE POLES OF THE MITOTIC SPINDLES>
s8b04a1.f1 172 1.9e-12 163 582 sp|P53441|CATR_NAEGR CALTRACTIN (CENTRIN) >gi|972963 (U21725)
centrin[Naegleria gruberi]

2.cytokinesis
<cytokinesis>
k9g02a1.f1 264 4e-22 362 619 gnl|PID|e347848 (AJ001587) sid3 [Schizosaccharomyces pombe]-septum
deposition
<f-actin-contractile ring during cytokinesis>
Contig989_j9f08a1.f1 152 5.7e-09 145 420 sp|P20111|AACS_C ALPHA-ACTININ, SKELETAL MUSCLE ISOFORM (F-ACTIN
CROSSLINKING PROTEIN) >pir||S02032 alpha-actinin 2, s
<TROPOMYOSIN-component of contractile ring>
o4g05a1.r1 241 8.9e-20 128 508 sp|Q02088|TPM_SCHPO TROPOMYOSIN >pir||S27127 tropomyosin -

yeast(*Schizosaccharomyces pombe*) >gi|173517 (L04126)

F. Meiosis (2)

<Rad9-required for meiotic chromosome condensation and synapsis>

w7a08a1.f1 172 9.6e-09 9 455 gi|1353390 (U34998) Rad9 [*Coprinus cinereus*]

<condensin-chromosome condensation protein>

j0a03a1.r1 154 2.5e-09 124 330 gi|3298547 (AC004681) putative condensin protein [*Arabidopsis thaliana*]

III. DNA synthesis

A. DNA replication (8)

<DNA POLYMERASE>

Contig467_o0b09a1.f1 539 2.6e-50 3 410 sp|P46588|DPOD_C DNA POLYMERASE DELTA LARGE CHAIN (DNA POLYMERASE III)>gi|951001 (X88804) DNA-directed DNA polymerase

<replication factor>

Contig793_v3e05a1.f1 836 9.6e-83 128 868 sp|P35250|AC14_H ACTIVATOR 1 40 KD SUBUNIT (REPLICATION FACTOR C 40 KDSUBUNIT) (A1 40 KD SUBUNIT) (RF-C 40 KD SUBUNIT)=human

Contig443_d5e10a1.f1 694 1.1e-67 194 1006 sp|Q92372|RFA1_S REPLICATION FACTOR-A PROTEIN 1 (SINGLE-STRANDED DNA-BINDING PROTEIN P68 SUBUNIT) >gi|1502413 (U59385)s

<Single-stranded DNA-binding protein>

<DnaJ protein>

Contig1596_i2e07a1.r1 485 6.2e-67 808 1233 sp|P25491|MAS5_Y MITOCHONDRIAL PROTEIN IMPORT PROTEIN MAS5 (PROTEIN YDJ1)>pir||S26703 dnaJ protein homolog YDJ1 - yeast

m5e08a1.r1 322 2.8e-28 145 807 sp|P25491|MAS5_YEAST MITOCHONDRIAL PROTEIN IMPORT PROTEIN MAS5 (PROTEIN YDJ1)>pir||S26703 dnaJ protein homolog

Contig1123_x8c07a1.f1 292 4.2e-25 260 745 sp|Q09912|PSI_SC PSI PROTEIN >pir||S55900 DNAJ-like protein homolog -fission yeast (*Schizosaccharomyces pombe*) >gi|953

n8g02a1.r1 138 2.1e-08 206 394 gi|1127833 (U40992) heat shock protein hsp40 homolog [*Homo sapiens*]>gi|1518918 (U41290) DNAJ homolog

h4a08a1.f1 128 0.00016 268 483 gi|1707079 (U80451) contains strong similarity to a DNAJ-like domain(PS:PS00636) [*Caenorhabditis eleg*]

B. DNA modification and repair (7)

<DNA LYASE>

x1g07a1.r1 362 1.5e-32 55 588 sp|P22936|APN1_YEAST DNA-(APURINIC OR APYRIMIDINIC SITE) LYASE (APENDONUCLEASE) (APURINIC-APYRIMIDINIC ENDONUCL

<endonuclease IV>

<DNA REPAIR PROTEIN>

w6c06a1.r1 737 2.9e-72 8 544 sp|P41410|RA54_SCHPO DNA REPAIR PROTEIN RHP54 >gnl|PID|e325327 (Z97208) Rad54p[*Schizosaccharomyces pombe*]

d4c06a1.r1 359 1e-30 102 539 sp|Q03468|ERC6_HUMAN EXCISION REPAIR PROTEIN ERCC-6 (COCKAYNE SYNDROME PROTEINCSB) >pir||A44224 DNA repair heli

w8d07a1.r1	234	1.4e-17	22 480	sp P53692 RA18_SCHPO DNA REPAIR PROTEIN RAD18 >gi 1150622 (X80929) rad18 geneproduct [Schizosaccharomyces pombe
Contig654_b0g01a1.f1	229	5.8e-17	242 469	sp P12753 RA50_Y DNA REPAIR PROTEIN RAD50 (153 KD PROTEIN) >pir [BWBYDLRAD50 protein - yeast (Saccharomyces cerevisiae
*Contig756_vlg09a1.r1	185	1.3e-10	22 342	gnl PID e349697 (Z99292) dna repair protein [Schizosaccharomyces pombe]
j7f09a1.r1	151	1.9e-06	89 460	sp P12753 RA50_YEAST DNA REPAIR PROTEIN RAD50 (153 KD PROTEIN) >pir [BWBYDLRAD50 protein - yeast (Saccharomyces

C. DNA packaging (7)

1. Histone

<Histones, class H1 (or I, or f1)>

Contig1551_g2h03a1.f1	180	2.4e-13	86 298	sp P53551 H1_YEA HISTONE H1 >pir [S69056 histone H1 - yeast (Saccharomycescerevisiae) >gi 1244786 (U43703) Lpi17p [Sac
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<Histones, class H2a (or IIb1, or f2a2)>

Contig1795_k8h08a1.f1	500	3.7e-47	216 611	sp P08844 H2A_EM HISTONE H2A >pir [A27332 histone H2A - Emericella nidulans>gi 168053 (M18258) histone H2A [Emericella
*Contig1045_n8g06a1.r1	175	8.6e-10	169 708	gi 3395780 (AF058445) histone macroH2A1.1 [Gallus gallus]

<Histones, class H2b (or IIb2, or f2b)>

Contig1765_g4e06a1.f1	443	3.7e-41	121 540	sp P23754 H2B_EM HISTONE H2B >pir [S11937 histone H2B - Emericella nidulans>gi 296335 (X55547) H2B [Emericella nidulan
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<Histones, class H3 (or III, or f3)>

Contig1767_dlh05a1.f1	669	4.3e-65	96 503	sp P23753 H3_EME HISTONE H3 >pir [S11938 histone H3 - Emericella nidulans>gi 296337 (X55548) H3 [Emericella nidulans]
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<Histones, class H4 (or IV, or f2a1)>

Contig1427_i8e09a1.r1	409	1.6e-37	141 386	sp P23750 H41_EM HISTONE H4.1 >pir [S11939 histone H4.1 - Emericellanidulans >gi 296341 (X55549) H4.1 [Emericella nidu
Contig1739_j7e02a1.f1	406	3e-37	237 482	sp P23751 H42_EM HISTONE H4.2 >pir [S11940 histone H4.2 -

2. DNA-binding (3)

<DNA-binding protein amdA>

m3b09a1.r1	374	9.6e-33	53 592	pir [S61908 DNA-binding protein amdA - Emericella nidulans >gi 454103 (L28810)regulatory protein [Emer
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<NONHISTONE>

Contig1353_g2f05a1.f1	280	7e-24	56 316	sp P11633 NHPB_Y NONHISTONE CHROMOSOMAL PROTEIN 6B >pir [B35072 nonhistonechromosomal protein NHP6B - yeast (Saccharom
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<SINGLE-STRAND TELOMERIC DNA-BINDING PROTEIN>

Contig332_g4e10a1.f1 166 1.8e-09 510 776 sp|P25555|GBP2_Y SINGLE-STRAND TELOMERIC DNA-BINDING PROTEIN
GBP2(G-STRAND BINDING PROTEIN 2) (RAP1 LOCALIZATION FACTO

IV. Gene Expression

A. Transcription

1. RNA Polymerase (9)

<RNA POLYMERASE I, rRNA>

c7b07a1.f1 352 3.8e-30 389 703 sp|P22138|RPA2_YEAST DNA-DIRECTED RNA POLYMERASE I 135 KD POLYPEPTIDE
(A135)(RNA POLYMERASE I SUBUNIT 2) >pir||
Contig80_13b11a1.r1 397 1e-34 24 551 sp|P15398|RPA1_S DNA-DIRECTED RNA POLYMERASE I 190 KD
POLYPEPTIDE>pir||JS0080 DNA-directed RNA polymerase (EC 2.7.7.6)
13b11a1.f1 225 2.1e-16 248 481 sp|P15398|RPA1_SCHPO DNA-DIRECTED RNA POLYMERASE I 190 KD
POLYPEPTIDE>pir||JS0080 DNA-directed RNA polymerase (
Contig479_d2h10a1.f1 228 2.6e-18 136 486 sp|P32529|RPA9_Y DNA-DIRECTED RNA POLYMERASE I 13.7 KD POLYPEPTIDE
(A12.2)>pir||A48107 DNA-directed RNA polymerase (EC

<RNA POLYMERASE II, mRNA>

a0b11a1.f1 476 2.2e-43 129 485 sp|P08518|RPB2_YEAST DNA-DIRECTED RNA POLYMERASE II 140 KD POLYPEPTIDE
(B150)(RNA POLYMERASE II SUBUNIT 2) >pir

<RNA POLYMERASE III, tRNA>

Contig535_c8b02a1.f1 342 2.2e-30 269 805 sp|P32910|RPC6_Y DNA-DIRECTED RNA POLYMERASE III 36 KD POLYPEPTIDE
(C34)>pir||A45107 DNA-directed RNA polymerase (EC 2

<RNA Polymerase>

Contig1398_j9h11a1.f1 220 1.8e-17 117 548 gi|3372230 (AF017074) RNA polymerase I, II and III 16.5 kDa
subunit[Arabidopsis thaliana]

2. Regulation (48)

<transcription factor>

xlg11a1.r1 1084 4.9e-109 9 635 gi|3411264 (AF080600) homeodomain DNA-binding transcription
factor [Emericellanidulans]
s3f07a1.r1 912 8.4e-91 28 681 sp|P52958|CT1A_FUSSO CUTINASE TRANSCRIPTION FACTOR 1 ALPHA
>gi|1262912(U51671) cutinase transcription factor 1
x3c12a1.r1 372 1.4e-33 68 415 gnl|PID|e1311364 (Z99168) putative heat shock transcription
factor[Schizosaccharomyces pombe]
*Contig380_f5h12a1.r1 307 4.7e-26 63 506 gi|1100209 (L49345) transcription factor ZFM1 [Homo sapiens]
*Contig286_g8f03a1.f1 310 6.8e-26 237 857 gnl|PID|e280495 (Z46606) helicase-like transcription factor [Homo
sapiens]
g6f09a1.r1 289 4.5e-24 60 296 sp|P52890|ATF1_SCHPO TRANSCRIPTION FACTOR ATF1 (TRANSCRIPTION FACTOR
MTS1)(PROTEIN SSS1) >pir||S66147 transcrip
*Contig1222_g6h05a1.r1 219 5.9e-14 55 537 sp|P48361|AS10_Y ASK10 PROTEIN >pir||S64402 probable transcription

Contig927_c4c12a1.f1	182	1.8e-13	311 541	factorASK10 - yeast (<i>Saccharomyces cerevisiae</i>) >gnl sp P40096 NCBI_Y CLASS 2 TRANSCRIPTION REPRESSOR >pir S50662
Contig1252_g4c01a1.r1	150	1e-06	11 292	hypotheticalprotein YER159c - yeast (<i>Saccharomyces cerev</i> sp P28349 NIT4_N NITROGEN ASSIMILATION TRANSCRIPTION FACTOR
*Contig132_m5h10a1.r1	139	2.4e-05	190 669	NIT-4>pir A41696 regulatory protein nit-4 - <i>Neurospora cr</i> pir S43749 transcription factor nft1 - fission yeast
o9c12a1.r1	123	4.6e-05	376 561	(<i>Schizosaccharomycespombe</i>) pir S61704 probable transcription factor YPL230w - yeast
n8h07a1.r1	132	0.0002	24 500	(<i>Saccharomycescerevisiae</i>) >gi 1181258 (X9456 sp P28349 NIT4_NEUCR NITROGEN ASSIMILATION TRANSCRIPTION FACTOR
Contig1461_o6c05a1.f1	130	0.00021	548 1069	NIT-4>pir A41696 regulatory protein nit-4 - Ne sp P28348 NIRA_E NITROGEN ASSIMILATION TRANSCRIPTION FACTOR
Contig546_c6g06a1.f1	126	0.00024	571 720	NIRA>pir A41697 nitrate assimilation regulatory protein n sp P56095 AP1_KL AP-1-LIKE TRANSCRIPTION FACTOR >gi 2245654
<SUPRESSOR OF STEM-LOOP PROTEIN-transcription initiation, pol binding>				
i0d11a1.r1	230	3.6e-18	261 473	(AF006499)transcription factor K1YAP1 (<i>Kluyveromyces lacti</i> gnl PID d1032706 (AB016221) SSL1 [<i>Schizosaccharomyces pombe</i>]=SUPPRESSOR OF STEM-LOOP PROTEIN, translation initiation and UV resistance
<TRANSCRIPTION INITIATION FACTOR TFIID>				
Contig1018_j7a02a1.f1	632	3.8e-61	522 899	sp Q12731 TF2D_E TRANSCRIPTION INITIATION FACTOR TFIID (TATA-BOX FACTOR)(TATA SEQUENCE-BINDING PROTEIN) (TBP) >gi 8878
x1d01a1.r1	520	8.2e-49	7 636	sp P38129 T2D4_YEAST TRANSCRIPTION INITIATION FACTOR TFIID 90 KD SUBUNIT(TAFII-90) >pir S34023 TATA box-bindin
Contig1605_e9a11a1.r1	490	4.3e-46	441 863	sp Q12731 TF2D_E TRANSCRIPTION INITIATION FACTOR TFIID (TATA-BOX FACTOR)(TATA SEQUENCE-BINDING PROTEIN) (TBP) >gi 8878
Contig1682_g9c11a1.f1	409	1.7e-37	512 751	sp Q12731 TF2D_E TRANSCRIPTION INITIATION FACTOR TFIID (TATA-BOX FACTOR)(TATA SEQUENCE-BINDING PROTEIN) (TBP) >gi 8878
f0d08a1.r1	259	9.7e-33	96 539	sp P53040 T2D5_YEAST TRANSCRIPTION INITIATION FACTOR TFIID 60 KD SUBUNIT(TAFII-60) >pir S64120 TATA box-bindin
Contig1037_c1e03a1.f1	217	2.4e-24	498 725	sp P38129 T2D4_Y TRANSCRIPTION INITIATION FACTOR TFIID 90 KD SUBUNIT(TAFII-90) >pir S34023 TATA box-binding protein-a
i8b08a1.r1	144	1.7e-06	185 484	sp P49846 T2D4_DROME TRANSCRIPTION INITIATION FACTOR TFIID 85 KD SUBUNIT (P85)(TAFII-80) >pir A54593 TFIID sub
<TRANSCRIPTION INITIATION PROTEIN SPT6>				
n8a10a1.r1	352	5.4e-30	81 650	sp P23615 SPT6_YEAST TRANSCRIPTION INITIATION PROTEIN SPT6 >pir A36468 SPT6protein - yeast (<i>Saccharomyces cere</i>
<HAP3 protein-transcription factor>				
Contig1371_e9d05a1.r1	459	5.5e-60	18 296	pir JC6080 HAP3 protein - <i>Emericella nidulans</i> >gi 1017716 (U35341) Hapc[<i>Emericella nidulans</i>]
<RNA helicase>				
g6h04a1.r1	821	3.6e-81	55 825	gi 2408027 (Z99162) atp-dependent rna helicase [<i>Schizosaccharomyces pombe</i>]
c7d09a1.r1	640	5.6e-62	24 716	sp Q09747 YB66_SCHPO PUTATIVE ATP-DEPENDENT RNA HELICASE C12C2.06

w5b11a1.r1	542	4.7e-51	14 412	>gi 984214(Z54140) probable ATP-dependent RNA sp Q09719 YA47_SCHPO PUTATIVE ATP-DEPENDENT RNA HELICASE-translation initiation C31A2.07C>pir S59645 hypothetical protein SPAC31A2.07
i8b11a1.r1	518	4.7e-49	12 392	sp P24782 DBP2_SCHPO P68-LIKE PROTEIN >pir S14048 RNA helicase dbp2 - fissionyeast (Schizosaccharomyces pombe)
j9a01a1.r1	527	1e-48	51 485	gi 172764 (M83755) STH1 protein, [Saccharomyces cerevisiae],snf2 RNA helicase homolog?
Contig1348_e7a04a1.r1	489	5.5e-46	159 845	sp P20447 DBP3_Y PROBABLE ATP-DEPENDENT RNA HELICASE DBP3 (HELICASE CA3)>pir S30805 probable RNA helicase CA3 - yeast
n2a07a1.r1	473	9.3e-44	213 668	sp Q09903 YAJ3_SCHPO PUTATIVE ATP-DEPENDENT RNA HELICASE c30d11.03>pir S62561 hypothetical protein SPAC30D11.0
g6h04a1.f1	420	1.1e-38	193 531	sp Q07478 HE47_YEAST PROBABLE ATP-DEPENDENT RNA HELICASE P47 HOMOLOG>pir S67620 hypothetical protein YDL084w -
Contig1557_g3e04a1.f1	275	3.7e-35	405 716	sp P20447 DBP3_Y PROBABLE ATP-DEPENDENT RNA HELICASE DBP3 (HELICASE CA3)>pir S30805 probable RNA helicase CA3 - yeast
Contig1337_w4b05a1.f1	181	2.5e-10	577 825	gnl PID e248472 (X98130) RNA helicase [Arabidopsis thaliana] >gnl PID e245943(X97970) RNA helicase [Arabidopsis thali
l5f03a1.f1	162	2.7e-09	389 601	sp Q09719 YA47_SCHPO PUTATIVE ATP-DEPENDENT RNA HELICASE c31A2.07C>pir S59645 hypothetical protein SPAC31A2.07
<HELICASE MOT1-essential>				
g3h10a1.r1	709	6.7e-68	15 749	sp P32333 MOT1_YEAST PROBABLE HELICASE MOT1 >pir S22775 MOT1 protein - yeast(Saccharomyces cerevisiae) >gi 171
y8h08a1.f1	207	2.8e-13	214 546	sp P32333 MOT1_YEAST PROBABLE HELICASE MOT1 >pir S22775 MOT1 protein - yeast(Saccharomyces cerevisiae) >gi 171
<regulatory protein creA-carbon catabolite repression>				
Contig1541_g5c01a1.f1	825	1.4e-81	10 588	sp Q01981 CREA_E DNA-BINDING PROTEIN CREA >gi 168035 (L03563) CREA[Emericella nidulans]
<HAC1-unfolded protein response pathway, transcrip activation>				
Contig1782_c7a10a1.f1	130	1.2e-05	435 527	pir S56223 HAC1 protein - yeast (Saccharomyces cerevisiae)
<TRANSCRIPTIONAL REPRESSOR>				
k5g05a1.f1	451	5.9e-42	10 384	sp P78706 RCO1_NEUCR TRANSCRIPTIONAL REPRESSOR RCO-1 >gi 1698504 (U57061)rco-1 gene product [Neurospora crassa]
Contig1520_g6a01a1.f1	258	9.8e-21	646 930	sp O14063 IMA1_S IMPORTIN ALPHA SUBUNIT (KARYOPHERIN ALPHA SUBUNIT)(SERINE-RICH RNA POLYMERASE I SUPPRESSOR PROTEIN) >
Contig673_a0c03a1.r1	959	8.5e-96	131 877	sp O14063 IMA1_S IMPORTIN ALPHA SUBUNIT (KARYOPHERIN ALPHA SUBUNIT)(SERINE-RICH RNA POLYMERASE I SUPPRESSOR PROTEIN) >
<QUINATE REPRESSOR>-repressor protein in the quinic acid utilization pathway, cluster>				
d4a10a1.f1	410	1.5e-36	104 658	pir JH0262 Qutr protein QUINATE REPRESSOR - Emericella nidulans >gi 168084 (M77664) repressorprotein [Emericella nidula
<PacC-factor regulates acid and base expressed genes>				
Contig573_c5d05a1.f1	138	1.3e-07	196 276	pir S54308 DNA binding protein - Emericella nidulans >gi 695416

(Z47081) DNAbinding protein [Emericella nidulans=pacC=transcription factor
mediating pH regulation

<RNA-BINDING POST-TRANSCRIPTIONAL REGULATOR>

Contig82_l3b05a1.r1 451 5.6e-42 136 654 sp|O13759|CSX1_S RNA-BINDING POST-TRANSCRIPTIONAL REGULATOR
CSX1>gnl|PID|e1198263 (Z99292) rna binding post-transcript

<HapE-transcription regulation, penicillin and acetamidase biosynthesis>

Contig1480_h4c03a1.f1 636 3e-111 149 556 gi|2098795 (U96847) HapE [Emericella nidulans]

-Asexual development-Central regulatory pathway

<regulatory protein brlA-transcription factor>

o4h01a1.r1 913 6.2e-91 11 511 sp|P10069|BRLA_EMENI REGULATORY PROTEIN BRLA (BRISTLE A PROTEIN)
>pir||A28913regulatory protein brlA - Emericel

c5h06a1.r1 897 3.2e-89 131 778 sp|P10069|BRLA_EMENI REGULATORY PROTEIN BRLA (BRISTLE A PROTEIN)
>pir||A28913regulatory protein brlA - Emericel

<REGULATORY PROTEIN WETA>

<STUA transcription factor>

<CELL PATTERN FORMATION-ASSOCIATED PROTEIN: SPATIAL LOCALIZATION OF ABAA AND BRLA>

Contig1369_d5d07a1.f1 543 1e-51 542 874 sp|P36011|STUA_E CELL PATTERN FORMATION-ASSOCIATED PROTEIN
>pir||A44068cell pattern formation-associated protein - Eme

p0c08a1.r1 133 3.4e-07 412 486 sp|P36011|STUA_EMENI CELL PATTERN FORMATION-ASSOCIATED PROTEIN
>pir||A44068cell pattern formation-associated pr-E. nidulans

3. Processing (19)

a. SPLICEOSOME

<SPLICEOSOME ASSOCIATED PROTEIN>

10f01a1.f1 224 6.2e-18 5 202 gnl|PID|e340019 (Z98979) putative spliceosome associated
protein[Schizosaccharomyces pombe]

<splicing factor>

Contig231_h8h02a1.r1 592 6.2e-57 50 832 gi|2911284 (U97681) putative splicing factor [Schizosaccharomyces
pombe]

Contig1332_a1d02c9.r1 243 5.4e-19 375 797 gnl|PID|e325342 (Z97209) splicing factor [Schizosaccharomyces pombe]

n3e12a1.r1 226 7.3e-18 396 683 gi|2749972 (AF012278) putative pre-mRNA splicing factor
[Schizosaccharomycespombe]

<small nuclear ribonucleoprotein>

Contig71_m0c10a1.r1 344 1.3e-30 19 441 gnl|PID|e339912 (Z98974) putative small nuclear
ribonucleoprotein[Schizosaccharomyces pombe]

Contig1261_s9f03a1.f1 310 5.2e-27 365 607 gnl|PID|e349593 (Z99259) small nuclear ribonucleoprotein
[Schizosaccharomycespombe]

Contig328_m7c12a1.r1 264 3.9e-22 166 396 gnl|PID|e1292641 (AL023534) small nuclear ribonucleoprotein
F[Schizosaccharomyces pombe]

Contig907_r5d10a1.f1 234 5.6e-19 252 455 gnl|PID|e1294550 (AL023706) small nuclear
ribonucleoprotein[Schizosaccharomyces pombe]

<U4/U6 SNRNA-ASSOCIATED SPLICING FACTOR PRP24>

m6f12a1.r1 259 3.7e-21 65 637 sp|P49960|PR24_YEAST U4/U6 SNRNA-ASSOCIATED SPLICING FACTOR PRP24 (U4/U6SNRPPROTEIN) >pir||s54480 U6 snRNP prot

<SPLICING FACTOR U2AF 65 KD SUBUNIT>

y6c08a1.r1 316 1.7e-27 9 530 sp|P26369|U2AF_MOUSE SPLICING FACTOR U2AF 65 KD SUBUNIT (U2 AUXILIARY FACTOR65 KD SUBUNIT) (U2 SNRNP AUXILIARY

y6c08a1.f1 259 4.9e-21 180 467 sp|P26369|U2AF_MOUSE SPLICING FACTOR U2AF 65 KD SUBUNIT (U2 AUXILIARY FACTOR65

<MITOCHONDRIAL RNA SPLICING PROTEIN>

i7h06a1.r1 224 2.8e-17 18 314 sp|Q01926|MRS2_YEAST MITOCHONDRIAL RNA SPLICING PROTEIN MRS2 PRECURSOR>pir||s62064 MRS2 protein - yeast (S. cerevisiae)

<tRNA splicing endonuclease>

n5h01a1.r1 241 8.7e-29 7 351 gnl|PID|e1298615 (AL023859) putative tRNA splicing endonuclease gamma subunit[Schizosaccharomyces pombe]

<HETEROGENEOUS NUCLEAR RIBONUCLEOPROTEIN K>

Contig1199_e9b12a1.f1 143 2.1e-08 266 424 sp|Q07244|ROK_HU HETEROGENEOUS NUCLEAR RIBONUCLEOPROTEIN K (HNRNP K)(DC-STRETCH BINDING PROTEIN) (CSEBP) (TRANSFORMATIO

<hnRNP-E1 protein>

Contig116_k0g05a1.r1 268 1.5e-22 92 559 pir||s42472 hnRNP-E1 protein - human >pir||s65678 PCBP-1 protein - human>gi|460771 (X78137) hnRNP-E1 [Homo sapien

<NAM8 PROTEIN-MITOCHONDRIAL SPLICING>

Contig1512_i2h05a1.f1 254 2.6e-18 1210 1413 gi|4026 (X64763) NAM8 gene product [Saccharomyces cerevisiae] >prf||1814447BNAM8 gene [Saccharomyces cerevisi

b.polyA addition

<POLYA>

l3h08a1.r1 260 3.4e-20 29 607 sp|Q10569|CPSA_BOVIN CLEAVAGE AND POLYADENYLATION SPECIFICITY FACTOR, 160 KDSUBUNIT (CPSF 160 KD SUBUNIT) >pir|

n1e12a1.r1 208 2.2e-15 194 430 gi|632500 (U17394) polyadenylation factor 64 kDa subunit [Xenopus laevis]

l3h08a1.f1 165 4.3e-10 168 431 sp|Q10570|CPSA_HUMAN CLEAVAGE AND POLYADENYLATION SPECIFICITY FACTOR, 160 KDSUBUNIT (CPSF 160 KD SUBUNIT) >gi|1

c.5' capping

<MRNA CAPPING ENZYME>

c8e06a1.r1 377 3.8e-34 59 604 sp|P40997|MCE1_SCHPO MRNA CAPPING ENZYME (MRNA GUANYLYLTRANSFERASE) (GTP--RNAGUANYLYLTRANSFERASE) >gi|562123 (U

d.other

<nucleolin-rRNA processing>

Contig1368_e9a08a1.f1 357 5.3e-32 341 781 bbs|110564 NSR1=nucleolin homolog [Saccharomyces cerevisiae, Peptide, 249 aa]

<3'-TERMINAL PHOSPHATE CYCLASE-RNA processing?>

h1d06a1.r1	264	3.9e-22	61 480	sp Q08096 RTC1_YEAST	RNA 3'-TERMINAL PHOSPHATE CYCLASE (RNA-3'-PHOSPHATECYCLASE) (RNA CYCLASE) >pir s66692 hyp
<FIBRILLARIN>					
Contig350_g3f08a1.f1	380	1.6e-34	184 483	sp P15646 FBRL_Y	FIBRILLARIN (NUCLEOLAR PROTEIN 1) >pir s25421 nucleolarprotein NOP1 - yeast (Saccharomyces cerevisia)
<MINOR CAPSID PROTEIN C>					
j4f08a1.r1	755	3.5e-74	27 548	sp P03711 VCAC_LAMB	MINOR CAPSID PROTEIN C (GPC) (CONTAINS: CAPSID ASSEMBLYPROTEIN NU3) >pir VHBPCL minor cap
<QUEUINE TRNA-RIBOSYLTRANSFERASE-trRNA, guanine modification>					
Contig336_g4d01a1.r1	473	2.8e-44	10 609	sp P15178 SYD_RA	ASPARTYL-TRNA SYNTHETASE (ASPARTATE--TRNA LIGASE) (ASPRS)>pir SYRTDT aspartate--trna ligase (EC 6.1.
Contig63_m0f06a1.r1	418	1.9e-38	210 737	gnl PID e1250585	(AL021813) phenylalanyl-trna synthetase alpha chain[Schizosaccharomyces pombe]
Contig181_m7a04a1.f1	357	3.5e-31	73 387	sp P46655 SYEC_Y	GLUTAMYL-TRNA SYNTHETASE, CYTOPLASMIC (GLUTAMATE--TRNALIGASE) (GLURS) (P85) >pir s53934 probable glu
Contig534_c8b04a1.f1	344	1.3e-30	342 776	sp P07284 SYSC_Y	SERYL-TRNA SYNTHETASE, CYTOPLASMIC (SERINE--TRNA LIGASE)(SERRS) >pir YSBYC serine--trna ligase (EC 6
Contig839_v3f08a1.f1	337	1.7e-29	152 601	gnl PID e1263903	(AL022103) histidyl-trna synthetase [Schizosaccharomycespombe]
Contig572_o5f10a1.r1	317	1.4e-26	214 726	sp P40825 SYAC_Y	ALANYL-TRNA SYNTHETASE, CYTOPLASMIC (ALANINE--TRNALIGASE) (ALARS) >pir s62065 alanine--trna ligase (
r8a04a1.f1	160	1.8e-10	9 170	sp Q23623 TGT_CAEL	PUTATIVE QUEUINE TRNA-RIBOSYLTRANSFERASE (TRNA-GUANINETRANSGLYCOSYLASE) (GUANINE INSERTION
4. tRNA synthetase (12)					
g8c04a1.r1	735	4.9e-72	25 558	sp P40825 SYAC_YEAST	ALANYL-TRNA SYNTHETASE, CYTOPLASMIC (ALANINE--TRNALIGASE) (ALARS) >pir s62065 alanine--tr
Contig1136_u4f08a1.f1	675	4.7e-65	102 1055	sp O13651 SYIC_S	ISOLEUCYL-TRNA SYNTHETASE, CYTOPLASMIC (ISOLEUCINE--TRNALIGASE) (ILERS) >gnl PID d1022285 (AB004538)
y8e05a1.r1	648	7.4e-63	12 545	gnl PID e1240164	(Y12589) phenylalanyl-trna synthetase [Candida albicans]
v3f08a1.r1	526	7e-50	42 545	gnl PID e1263903	(AL022103) histidyl-trna synthetase [Schizosaccharomycespombe]
c8b04a1.r1	511	2.5e-48	76 651	sp O14018 SYSC_SCHPO	SERYL-TRNA SYNTHETASE, CYTOPLASMIC (SERINE--TRNA LIGASE)(SERRS) >gnl PID e351309 (Z97210)
mlh01a1.r1	496	9.7e-47	106 651	sp P38088 SYG_YEAST	GLYCYL-TRNA SYNTHETASE (GLYCINE--TRNA LIGASE) (GLYRS)>pir s48285 probable glycine--trna 1
n0h04a1.f1	449	9.6e-42	75 437	sp P04802 SYDC_YEAST	ASPARTYL-TRNA SYNTHETASE, CYTOPLASMIC (ASPARTATE--TRNALIGASE) (ASPRS) >pir SYBYDC asparta
r1a02a1.r1	402	1.5e-36	207 641	sp Q05506 SYRC_YEAST	PROBABLE ARGINYL-TRNA SYNTHETASE, CYTOPLASMIC(ARGININE--TRNA LIGASE) (ARGRS) >pir s70106
i0b08a1.r1	398	2.5e-36	12 443	sp P04803 SYWM_YEAST	TRYPTOPHANYL-TRNA SYNTHETASE, MITOCHONDRIAL(TRYPTOPHAN--TRNA LIGASE) (TRPRS) >pir YWBYM t

rla02a1.f1	244	4.4e-19	151 366	sp Q05506 SYRC_YEAST PROBABLE ARGINYL-TRNA SYNTHETASE, CYTOPLASMIC(ARGININE--TRNA LIGASE) (ARGRS) >pir s70106
m7a04a1.r1	223	1e-16	85 624	gnl PID e339154 (Z98849) putative glutamyl-trna synthetas [Schizosaccharomycespombe]
Contig1126_u4c01a1.r1	201	3.5e-15	169 531	sp P43835 SYW_HA TRYPTOPHANYL-TRNA SYNTHETASE (TRYPTOPHAN--TRNA LIGASE)(TRPRS) >pir c64083 tryptophan--trna ligase (E

5. RNA Degradation (3)

<ribonuclease>

n0b06a1.r1	401	1.1e-36	35 433	sp P24657 RNTR_TRIVI RIBONUCLEASE TRV >pir JX0197 ribonuclease T2 (EC3.1.27.1) - fungus (Trichoderma viride) >
o4d08a1.r1	314	2.9e-26	14 463	gnl PID e349364 (Z99259) ribonuclease II RNB family protein[Schizosaccharomyces pombe]

<NONSENSE-MEDIATED MRNA DECAY PROTEIN-DECAY OF MRNAS CONTAINING PREMATURE STOP CODONS>

w4a10a1.r1	471	4.6e-44	61 528	pir s48909 NMD3 protein - yeast (Saccharomyces cerevisiae) >gi 458900(U00027) Nmd3p: Putative Upf1p =NONSENSE-MEDIATED MRNA DECAY PROTEIN 3
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B. Protein Biosynthesis

1. initiation (16)

<EUKARYOTIC TRANSLATION INITIATION>

Contig1022_n1e01a1.r1	851	2.3e-84	45 1043	sp Q10425 IF3X_S PROBABLE EUKARYOTIC TRANSLATION INITIATION FACTOR 3 BETASUBUNIT (EIF-3 BETA) >gnl PID e1168609 (Z7069
Contig1049_m3c05a1.r1	522	1.7e-49	154 573	sp P56286 IF2A_S EUKARYOTIC TRANSLATION INITIATION FACTOR 2 ALPHA SUBUNIT(EIF-2-ALPHA) >gnl PID e1216795 (AL021046) tr
Contig1590_a1e03f2.f1	500	3.7e-47	104 499	sp P47813 IF1A_H EUKARYOTIC TRANSLATION INITIATION FACTOR 1A (EIF-1A)(EIF-4C) >gi 306725 (L18960) protein synthesis fa
u4a05a1.f1	410	1.3e-37	282 593	sp P40217 IF34_YEAST EUKARYOTIC TRANSLATION INITIATION FACTOR 3 DELTA SUBUNIT(EIF-3 DELTA) (EIF3 P39) (TRANSLAT
Contig1493_d4b03a1.f1	323	1.7e-27	487 846	sp Q10425 IF3X_S PROBABLE EUKARYOTIC TRANSLATION INITIATION FACTOR 3 BETASUBUNIT (EIF-3 BETA) >gnl PID e1168609 (Z7069
z6f08a1.r1	240	1.3e-19	207 521	sp P78954 IF4E_SCHPO EUKARYOTIC TRANSLATION INITIATION FACTOR 4E (EIF-4E)(EIF4E) (MRNA CAP-BINDING PROTEIN) (EI

<INITIATION FACTOR>

Contig299_g7c05a1.r1	654	1.7e-63	4 495	sp P47943 IF4A_S EUKARYOTIC INITIATION FACTOR 4A (EIF-4A) >gnl PID e114182(X80796) translation initiation factor eIF-4
Contig1750_h4c11a1.f1	594	3.9e-57	266 742	sp P23301 IF52_Y INITIATION FACTOR 5A-2 (EIF-5A) (EIF-4D) (HYPUSINECONTAINING PROTEIN HP2) >pir FIBYA1 translation in
r3d06a1.r1	530	2.5e-50	29 853	gnl PID e1291633 (AL023287) hypothetical translation initiation factor[Schizosaccharomyces pombe]
Contig1153_h4c11a1.r1	367	4.6e-33	320 550	sp P23301 IF52_Y INITIATION FACTOR 5A-2 (EIF-5A) (EIF-4D) (HYPUSINECONTAINING PROTEIN HP2) >pir FIBYA1 translation in
j5c01a1.f1	313	2.5e-27	56 508	sp P32502 E2BB_YEAST TRANSLATION INITIATION FACTOR EIF-2B DELTA

d5c04a1.f1	247	2.4e-20	254 565	SUBUNIT(EIF-2B GDP-GTP EXCHANGE FACTOR) (GUANIN sp P32774 TOA2_YEAST TRANSCRIPTION INITIATION FACTOR IIA SMALL CHAIN (TFIIA13.5 KD SUBUNIT) >pir A41810 transc
p0g09a1.r1	200	7.4e-14	72 470	sp Q10475 YDF3_SCHPO PROBABLE EUKARYOTIC INITIATION FACTOR C17C9.03>gnl PID e241757 (Z73099) probable initiatio
j9a04a1.r1	161	2.3e-10	16 339	sp P56288 E2BG_SCHPO PROBABLE TRANSLATION INITIATION FACTOR EIF-2B GAMMASUBUNIT (EIF-2B GDP-GTP EXCHANGE FACTOR
*Contig614_c2e07a1.f1	120	1.5e-05	396 590	gnl PID e1291633 (AL023287) hypothetical translation initiation factor[Schizosaccharomyces pombe]
l3f02a1.r1	128	7.1e-05	172 420	sp P32502 E2BB_YEAST TRANSLATION INITIATION FACTOR EIF-2B DELTA SUBUNIT(EIF-2B GDP-GTP EXCHANGE FACTOR) (GUANIN

2. elongation (16)

<ELONGATION FACTOR, eucaryotic and archaeal>

Contig1720_c8h08a1.f1	1458	1.8e-211	94 1032	sp Q01765 EF1A_P ELONGATION FACTOR 1-ALPHA (EF-1-ALPHA) >gnl PID e229326(X96614) EF1-alpha translation elongation fact
Contig1745_g7f04a1.r1	825	1.2e-81	146 790	sp P53978 EF3B_Y ELONGATION FACTOR 3B (EF-3B) >pir S62926 probablemembrane protein YNL014w - yeast (Saccharomyces cer
o9f08a1.r1	695	3.9e-67	26 580	gnl PID e212187 (X92971) translocation elongation factor [Saccharomycescerevisiae]
Contig938_e7d04a1.r1	678	1.9e-65	112 1035	sp P53978 EF3B_Y ELONGATION FACTOR 3B (EF-3B) >pir S62926 probablemembrane protein YNL014w - yeast (Saccharomyces cer
o9f08a1.f1	524	7.6e-49	78 557	gnl PID e1294522 (AL023704) translocation elongation factor[Schizosaccharomyces pombe]
z5g01a1.f1	504	6.3e-47	289 708	sp O14460 EF2_SCHPO ELONGATION FACTOR 2 (EF-2) >gnl PID d1024469 (D83975)elongation factor 2 [Schizosaccharomy
Contig1388_c3h01a1.f1	483	2.1e-45	167 817	gnl PID d1032532 (AB016046) elongation factor 1 beta [Schizosaccharomycespombe]
Contig847_w9b11a1.r1	434	5e-39	8 565	pir S25363 translation elongation factor eEF-3 - yeast (Candida albicans)>gi 2521 (Z12822) translation elongatio
Contig1190_r7h02a1.r1	418	1.8e-38	269 997	sp P49373 TFS2_S TRANSCRIPTION ELONGATION FACTOR S-II (TFIIS) >pir S63845transcription elongation factor TFIIS - fiss
o6a10a1.r1	345	8.6e-30	8 262	sp P25039 EFG1_YEAST ELONGATION FACTOR G, MITOCHONDRIAL 1 PRECURSOR (MEF-G-1)>pir S61642 translation elongation
w5g09a1.f1	325	1.1e-27	327 602	gi 2832315 (AF045014) translation release factor eRF3 [Podospora anserina]
*Contig1412_fld12a1.f1	251	1.9e-19	436 636	gnl PID e351430 (Z99568) elongation factor [Schizosaccharomyces pombe]
Contig4_v7g03a1.r1	184	1.2e-12	239 679	sp P49373 TFS2_S TRANSCRIPTION ELONGATION FACTOR S-II (TFIIS) >pir S63845transcription elongation factor TFIIS - fiss
j7e11a1.r1	169	6e-09	157 408	sp P25997 EF3_CANAL ELONGATION FACTOR 3 (EF-3) >gi 2498 (Z11484) elongationfactor 3 [Candida albicans]
<elongation factor 2> n3d10a1.r1	591	4.4e-56	21 686	gnl PID d1005229 (D21163) similar to human elongation factor 2 mRNA

(HSEF2).[Homo sapiens]

<TRANSLATION FACTOR>

g5f06a1.f1 232 9.9e-19 343 567 sp|P79060|SUI1_SCHPO PROTEIN TRANSLATION FACTOR SUI1

3. termination (1)

<PEPTIDE CHAIN RELEASE FACTOR>

Contig810_r8e11a1.r1 1398 2.6e-142 27 965 gi|2996008 (AF053983) translation release factor subunit 1 [Podosporaanserina]

4. Ribosomal proteins (88)

<a.40S ribosomal protein> (35)

Contig1727_c8e12a1.f1 806 1.3e-79 53 568 sp|P52810|RS9_PO 40S RIBOSOMAL PROTEIN S9 (S7) >gnl|PID|e242707 (X96613)cytoplasmic ribosomal protein S7 [Podospora an

Contig1574_c9a10a1.f1 788 1e-77 349 1023 sp|P05752|RS6_SC 40S RIBOSOMAL PROTEIN S6 >pir||R3ZP6E ribosomal proteins6.e, cytosolic - fission yeast (Schizosacchar

Contig1529_alc06c9.r1 787 1.3e-77 68 694 sp|P26783|RS5_YE 40S RIBOSOMAL PROTEIN S5 (RP14) (YS8) >pir||S55720ribosomal protein S5.e - yeast (Saccharomyces cerev

Contig59_mlg12a1.r1 770 8.5e-76 29 634 sp|P40910|RS3A_C 40S RIBOSOMAL PROTEIN RP10 >pir||S49366 ribosomal proteins0.e.B, cytosolic - yeast (Candida albicans)

Contig1219_dlc01a1.f1 742 7.5e-73 125 721 sp|P05754|RS8_YE 40S RIBOSOMAL PROTEIN S8 (S14) (YS9) (RP19) >pir||S45591ribosomal protein S8.e, cytosolic - yeast (Sa

Contig1770_dh11a1.r1 717 3.4e-70 368 787 sp|P27073|RS19_E 40S RIBOSOMAL PROTEIN S19 (S16) >pir||JQ1349 ribosomalprotein S19.e, cytosolic - Emericella nidulans

Contig1573_dlc08a1.f1 656 9.9e-64 283 762 sp|P26781|RS41_Y 40S RIBOSOMAL PROTEIN RP41 (YS12) (S18A / S18B)>pir||S41784 ribosomal protein S11.e, cytosolic - yeas

Contig1297_g7h08a1.f1 653 2.1e-63 299 757 sp|P34737|RS15_P 40S RIBOSOMAL PROTEIN S15 (S12) >pir||A53793 ribosomalprotein S12, cytosolic - Podospora anserina >gi

Contig485_dih08a1.r1 627 1.1e-60 223 840 sp|P25443|RS4_YE 40S RIBOSOMAL PROTEIN S4 (OMNIPOTENT SUPPRESSOR PROTEINSUP44) (RP12) (S2E) >pir||R3BYS2 ribosomal prot

Contig1106_alc02c9.r1 610 7.9e-59 315 737 sp|P34737|RS15_P 40S RIBOSOMAL PROTEIN S15 (S12) >pir||A53793 ribosomalprotein S12, cytosolic - Podospora anserina >gi

Contig1417_r2f10a1.f1 600 7.4e-58 161 571 sp|P27770|RS17_N 40S RIBOSOMAL PROTEIN S17 (CRP3) >pir||S34441 ribosomalprotein L17.e - Neurospora crassa >gi|168796 (

Contig1491_d5g06a1.f1 596 2.3e-57 58 486 sp|P40213|R61A_Y 40S RIBOSOMAL PROTEIN RS16 HOMOLOG (RP61R HOMOLOG)>pir||S67619 ribosomal protein S16.e.B - yeast (Sac

Contig1399_d2h09a1.f1 579 1.4e-55 212 580 gi|3114615 (AF052483) 40S ribosomal protein S12 [Erysiphe graminis f. sp.hordei]

Contig1639_hle05a1.r1 547 3.7e-52 228 626 sp|P35271|RS18_Y 40S RIBOSOMAL PROTEIN S18E >pir||S50886 ribosomal proteins18.e, cytosolic - yeast (Saccharomyces cere

Contig1524_u4e07a1.r1 508 4.8e-48 53 466 sp|P19115|RS14_N 40S RIBOSOMAL PROTEIN S14 (CRP2) >gi|2995 (X53734)ribosomal protein crp-2 [Neurospora crassa]

*Contig1089_f0f06a1.r1 448 1.1e-41 3 458 sp|Q10101|RS7_SC PROBABLE 40S RIBOSOMAL PROTEIN S7

Contig1779_c6f04a1.f1 395 4.8e-36 94 399 sp|P26782|RS24_Y 40S RIBOSOMAL PROTEIN S24E (RP50) >pir||S48410

Contig1593_g6c01a1.f1	392	9.6e-36	47 331	sp Q08745 YO93_Y	ribosomalprotein S24.e - yeast (<i>Saccharomyces cerevisi</i>
Contig993_m8f08a1.r1	384	6.7e-35	106 351	gnl PID e1313483	PUTATIVE 40S RIBOSOMAL PROTEIN IN SNF2-CPA1 INTERGENICREGION >pir s67197 ribosomal protein s10.e.A - (AL031154) 40s ribosomal protein s27
Contig1570_c9b09a1.f1	371	1.5e-33	134 457	sp P23403 RS20_X	type[Schizosaccharomyces pombe] 40S RIBOSOMAL PROTEIN S20 (S22) >pir A37974
Contig311_n0e01a1.f1	365	7.2e-33	164 436	sp P05753 RS4E_Y	ribosomalprotein S20 - African clawed frog >gi 214758 (M 40S RIBOSOMAL PROTEIN S4 (S7) (YS6) (RP5)
Contig1571_d5d04a1.f1	238	2e-19	54 233	sp Q12087 RS30_Y	>pir s20054ribosomal protein s4.e, cytosolic - yeast (Sacc 40S RIBOSOMAL PROTEIN S30 >pir s67074 ribosomal
Contig980_n0f03a1.f1	226	3.9e-18	252 413	sp Q10421 RS28_S	proteinsS30.e, cytosolic - yeast (<i>Saccharomyces cerev</i> PROBABLE 40S RIBOSOMAL PROTEIN S28
Contig873_r4c07a1.f1	210	1.8e-16	192 368	(S33)>gnl PID e1168607	(Z70691) ribosomal protein S28(Schizosaccha sp P53733 YN8L_Y
Contig1691_c3g10a1.f1	200	6.6e-15	76 291	>pir s63368probable ribosomal protein S19, mitochondrial - ye sp P79009 RS25_S	PUTATIVE 40S RIBOSOMAL PROTEIN YNR037C 40S RIBOSOMAL PROTEIN S25 (S31)
y6c06a1.r1	747	2.2e-73	88 615	>gnl PID d1019840(AB000398)	ribosomal protein S31 [Schizosaccharomyce
w8d11a1.r1	621	5.6e-60	61 492	sp Q01291 RSP4_NEUCR	40S RIBOSOMAL PROTEIN SA HOMOLOG (RIBOSOME-ASSOCIATEDPROTEIN 1) >gi 1039443 (U36470) putat
ald06f2.f1	501	2.8e-47	163 549	gnl PID e1293406	(AL023594) 40s ribosomal protein s4 type [Schizosaccharomycespombe]
w4e01a1.r1	433	4.6e-40	58 528	sp P04648 RS22_YEAST	40S RIBOSOMAL PROTEIN S22 (YS24) (YP58) >pir R4BY24ribosomal protein s15a.e.c10 - yeast (sp P32899 YHU8_YEAST
Contig182_i7h11a1.f1	245	3.8e-20	141 494	>pir s33911hypothetical protein YHR148w - yeast (S gnl PID e1287762	PUTATIVE 40S RIBOSOMAL PROTEIN YHR148W (AL022598) 40s mitochondrial ribosomal
y6c06a1.f1	218	2.8e-17	159 512	protein[Schizosaccharomyces pombe] sp Q01291 RSP4_NEUCR	40S RIBOSOMAL PROTEIN SA HOMOLOG (RIBOSOME-ASSOCIATEDPROTEIN 1) >gi 1039443 (U36470) putat
o6g03a1.f1	191	1.4e-13	186 386	sp P27929 NAM9_YEAST	NAM9 PROTEIN PRECURSOR >pir s55146 ribosomal proteinsS4.e precursor - yeast (<i>Saccharomyces</i>
o5e07a1.r1	186	2.9e-13	69 290	sp P32902 RT04_YEAST	MITOCHONDRIAL 40S RIBOSOMAL PROTEIN MRP4 >pir A42115ribosomal protein S2, mitochondrial -
w4e03a1.f1	184	2e-11	164 352	sp Q10234 RT05_SCHPO	PROBABLE MITOCHONDRIAL 40S RIBOSOMAL PROTEIN S5>gnl PID e223738 (Z69727) probable ribosoma
i3f02a1.r1	126	8.6e-07	7 93	sp P02382 RMS5_EMENI	MITOCHONDRIAL RIBOSOMAL PROTEIN S5 >pir QXASRI 218 rRNAintron protein - <i>Emericella nidulans</i>
<b.60S ribosomal protein> (50)					
Contig1635_a1e02f2.f1	959	8e-96	235 843	sp O13418 RL15_A	60S RIBOSOMAL PROTEIN L15 >gnl PID e1181730 (Y15321)putative ribosomal protein L15 [<i>Aspergillus niger</i>
Contig1080_i7d05a1.r1	800	5e-79	45 860	sp P35679 RL2_SC	60S RIBOSOMAL PROTEIN L2 >gi 312177 (X73146) ribosomalprotein L2 [<i>Schizosaccharomyces pombe</i>]

Contig1656_c1c10a1.f1	790	5.9e-78	62 712	gnl PID e330379 (Z97992) 60s ribosomal protein L10 [Schizosaccharomyces pombe]
Contig1757_d5g11a1.f1	747	2.5e-73	111 710	sp P26784 R13A_Y 60S RIBOSOMAL PROTEIN L13A (RP22) >pir S48401 ribosomalprotein L16.e.A, cytosolic - yeast (Saccharom
Contig1652_o6h02a1.f1	722	9.4e-71	44 793	gnl PID e1285382 (AL022304) 60s ribosomal protein [Schizosaccharomyces pombe]
Contig1436_r6h09a1.r1	628	8.9e-61	249 767	sp P47913 RL1X_Y 60S RIBOSOMAL PROTEIN L18A >pir S59848 ribosomal proteinL18a.e.c15 - yeast (Saccharomyces cerevisiae
Contig1192_g4c02a1.f1	625	2.2e-60	179 868	sp P31334 RM09_Y MITOCHONDRIAL 60S RIBOSOMAL PROTEIN L9 PRECURSOR (YML9)>pir R5BYL3 ribosomal protein L3 precursor, m
Contig1536_c8f11a1.f1	619	8.6e-60	503 994	sp P35979 RL12_M 60S RIBOSOMAL PROTEIN L12 >pir JN0778 ribosomal proteinL12 - mouse >gi 398048 (L04280) ribosomal pro
Contig1395_alb03c9.r1	598	1.3e-57	202 699	sp P26321 RL1_YE 60S RIBOSOMAL PROTEIN L1 (L5) (YL3) (RIBOSOMAL 5 SRNA-BINDING PROTEIN) >gi 173232 (M65056) 5S ribosom
Contig1091_b0f01a1.f1	561	1.2e-53	204 608	sp P04451 RL1A_Y 60S RIBOSOMAL PROTEIN L17 >pir R5BY17 ribosomal proteinL23.e, cytosolic - yeast (Saccharomyces cerev
Contig955_b0a07a1.r1	406	6.9e-52	328 645	sp O13418 RL15_A 60S RIBOSOMAL PROTEIN L15 >gnl PID e1181730 (Y15321)putative ribosomal protein L15 [Aspergillus niger
Contig1603_c4f01a1.f1	533	1.1e-50	31 504	sp Q02753 R21A_Y 60S RIBOSOMAL PROTEIN L21E A >pir S28921 ribosomalprotein L21.e.A, cytosolic - yeast (Saccharomyces
Contig1759_j7c10a1.f1	524	9.3e-50	48 365	sp P52809 RL44_P 60S RIBOSOMAL PROTEIN L44 (L41) >gnl PID d1011717(D67040) ribosomal protein L41 [Candida utilis]
Contig1608_c8d07a1.f1	513	1.3e-48	362 925	sp P05735 RL19_Y 60S RIBOSOMAL PROTEIN L19 (L23) (YL14) (RP33) (RP15L)>pir S44597 ribosomal protein L19.e, cytosolic
Contig1344_c1c05a1.f1	508	4.8e-48	198 644	sp P17076 RL4A_Y 60S RIBOSOMAL PROTEIN L7A-2 (L4-2) (YL5) (RP6)>pir R5BY7A ribosomal protein L7a.e.A - yeast (Sacchar
Contig575_c5b05a1.r1	441	5.8e-41	62 430	gnl PID e349694 (Z99296) 60s ribosomal protein L32 [Schizosaccharomyces pombe]
Contig816_y6a08a1.f1	422	5.6e-39	90 422	sp P40525 YIF2_Y PROBABLE 60S RIBOSOMAL PROTEIN YIL052C >pir S48427ribosomal protein L34.e.B, cytosolic - yeast (Sacc
Contig1499_n8g05a1.r1	420	1.1e-38	103 411	sp P41056 R372_Y 60S RIBOSOMAL PROTEIN L37B (YL37) (RP47) >pir S44069ribosomal protein L35a.e.c15 - yeast (Saccharomy
Contig1449_e4g11a1.r1	387	3e-35	63 395	sp P04649 RL34_Y 60S RIBOSOMAL PROTEIN L34 (YL28) >pir R5BY1E ribosomalprotein L31.e.A, cytosolic - yeast (Saccharomy
Contig1705_h1g02a1.f1	387	3.4e-35	129 473	gi 292435 (L07287) ribosomal protein L26 [Homo sapiens]
Contig1803_c8a04a1.f1	387	3.4e-35	71 376	sp P52808 RL30_S 60S RIBOSOMAL PROTEIN L30 (L32) >gi 1621046 (U52080)ribosomal protein Rpl32p [Schizosaccharomyces pom
Contig1696_l3e05a1.f1	377	4e-34	50 301	gi 2665824 (AF035770) ribosomal protein L37 [Schistosoma mansoni]
Contig682_y8h12a1.f1	376	4.4e-34	80 352	sp P49631 R37A_Y PROBABLE 60S RIBOSOMAL PROTEIN L37A >pir S54068ribosomal protein L37a.e - yeast (Saccharomyces cerev
Contig1513_d4c07a1.f1	373	9.8e-34	540 923	sp Q12690 R13D_Y PROBABLE 60S RIBOSOMAL PROTEIN L13E B >pir S67618ribosomal protein L13.e.A, cytosolic - yeast (Sacch
Contig1125_r5f10a1.f1	368	3.2e-33	334 609	sp P38665 RL24_K 60S RIBOSOMAL PROTEIN L24 (L30) >gi 173317

Contig1019_c1h09a1.f1	353	1.2e-31	150 410	(L05777)ribosomal protein L30 [Kluyveromyces lactis] sp P06380 RL16_Y 60S RIBOSOMAL PROTEIN L16 (YL16) (39A) (RP39)>pir S59767 ribosomal protein L11.e.A, cytosolic - yeas
Contig38_z3e04a1.r1	350	2.6e-31	180 512	sp Q02753 R21A_Y 60S RIBOSOMAL PROTEIN L21E A >pir S28921 ribosomalprotein L21.e.A, cytosolic - yeast (Saccharomyces
Contig1168_u4f09a1.r1	343	1.5e-30	157 477	sp Q02753 R21A_Y 60S RIBOSOMAL PROTEIN L21E A >pir S28921 ribosomalprotein L21.e.A, cytosolic - yeast (Saccharomyces
Contig1799_d3h04a1.f1	295	1.9e-25	62 451	sp O14069 YEA4_s PROBABLE 60S RIBOSOMAL PROTEIN C2E11.04 >gnl PID e339274(Z98850) ribosomal protein [Schizosaccharomyc
Contig267_h0a09a1.f1	269	1.1e-22	90 470	sp P35996 RM38_Y MITOCHONDRIAL 60S RIBOSOMAL PROTEIN L38 (YML38)>pir S38000 ribosomal protein L14, mitochondrial - ye
Contig653_b0g02a1.f1	266	2.2e-22	261 470	sp P08978 RL2A_N 60S RIBOSOMAL PROTEIN L27A (L29) (CRP1) >pir R6NC7Aribosomal protein L27a.e - Neurospora crassa >gi
Contig754_t2e02a1.f1	248	1.6e-20	122 412	sp P05745 RL39_Y 60S RIBOSOMAL PROTEIN YL39 >pir S50922 ribosomal proteinL36.e.A, cytosolic - yeast (Saccharomyces ce
Contig645_c0a03a1.r1	234	5.1e-19	287 478	sp P24049 RL17_R 60S RIBOSOMAL PROTEIN L17 (L23) (AMINO ACIDSTARVATION-INDUCED PROTEIN) (ASI) >pir R5RT17 ribosomal p
Contig1622_u4f10a1.r1	193	1.2e-14	72 398	gi 3098460 (AF040713) 60S ribosomal protein P2 [Cryptochiton stelleri]
Contig1441_e9c05a1.f1	169	4.2e-12	8 181	sp P05740 RL7A_Y 60S RIBOSOMAL PROTEIN YL17-A >pir S38012 ribosomalprotein L17.e.A, cytosolic - yeast (Saccharomyces
Contig1284_j4h04a1.r1	142	6.3e-09	344 493	sp P36525 RM24_Y MITOCHONDRIAL 60S RIBOSOMAL PROTEIN L24 PRECURSOR (YML24)>pir S50921 ribosomal protein YmL24, mitoch
Contig1552_c5c05a1.f1	140	2.6e-08	33 188	sp P05739 R16B_Y 60S RIBOSOMAL PROTEIN YL16B >pir S55970 ribosomalprotein L6.e.B, cytosolic - yeast (Saccharomyces ce
Contig551_c6d06a1.r1	153	4.5e-08	361 573	sp O14337 RM07_s PROBABLE MITOCHONDRIAL 60S RIBOSOMAL PROTEIN L7 PRECURSOR>gnl PID e325415 (Z97211) probable mitochond
*Contig1149_w8a04a1.r1	120	0.00016	324 488	sp P36517 RM04_Y MITOCHONDRIAL 60S RIBOSOMAL PROTEIN L4 PRECURSOR (YML4)>pir S59407 ribosomal protein YmL4 precursor,
o4g08a1.r1	443	3.8e-41	135 500	sp P51997 RL2B_PUCGR 60S RIBOSOMAL PROTEIN L23A (L25) >gi 1707876 (U44800)ribosomal protein L23a [Puccinia gram
v1a02a1.r1	433	4.7e-40	84 524	sp P53875 YNS5_YEAST PUTATIVE 60S MITOCHONDRIAL RIBOSOMAL PROTEIN YNL185C>pir S63140 probable ribosomal protei
x1d02a1.r1	364	8.3e-33	6 296	sp P27659 RL3_MOUSE 60S RIBOSOMAL PROTEIN L3 (J1 PROTEIN) >gi 52741 (Y00225)J1 protein [Mus musculus] >prf 16
c8b05a1.r1	351	2.2e-31	104 454	gnl PID e1285350 (AL022299)60S ribosomal protein [Schizosaccharomyces pombe]>gnl PID e1291883 (AJ001133) ribos
e4b10a1.r1	315	1.4e-27	30 335	sp Q09668 RL22_SCHPO 60S RIBOSOMAL PROTEIN L22 >gnl PID e334258 (Z98595) 60sribosomal protein l22 [Schizosaccha
Contig1679_d5d09a1.f1	174	1.2e-12	375 632	sp P50344 RLA1_C 60S ACIDIC RIBOSOMAL PROTEIN P1 (ALLERGEN CLA H 12) (CLAH XII) >gi 143425 (X85180) ribosomal protein
z3g02a1.f1	151	3.6e-10	201 437	sp P22354 RM20_YEAST MITOCHONDRIAL 60S RIBOSOMAL PROTEIN L20 PRECURSOR (YML20)>pir S38163 ribosomal protein Ym

z5g03a1.f1	148	6.8e-10	102 290	sp P41805 RL10_YEAST 60S RIBOSOMAL PROTEIN L10 (L9) (UBIQUINOL-CYTOCHROME CREDUCTASE COMPLEX SUBUNIT VI REQUIRI
c9e07a1.f1	162	3e-09	108 395	sp P05317 RLA0_YEAST 60S ACIDIC RIBOSOMAL PROTEIN P0 (L10E) >gi 4371 (X06959)ribosomal protein A0 (AA 1-312) [S
Contig1435_j4b03a1.f1	145	1.7e-09	310 423	sp P73300 RL36_s 50S RIBOSOMAL PROTEIN L36 >gnl PID d1018061 (D90905) 50Sribosomal protein L36 [Synechocystis sp.]
n8f06a1.f1	142	3.1e-09	355 447	sp P05747 RL43_YEAST 60S RIBOSOMAL PROTEIN YL43 >pir S71066 ribosomal proteinL29.e, cytosolic - yeast (Sacchar
<ribosomal protein> c9e07a1.r1	459	8.4e-43	79 588	pir R5BY0E acidic ribosomal protein P0.e, cytosolic - yeast (Saccharomycescerevisiae) >gi 171806 (M37
o6f09a1.f1	446	1.5e-41	112 489	gi 3003044 (AF054907) putative 5S rRNA binding ribosomal protein [Neurospora crassa]
Contig1007_f0f06a1.f1	330	3.8e-29	257 523	gi 2737908 (U73847) ribosomal protein [Neurospora crassa]

5. Post-translational modifications (14)

a.methylation

<SERINE HYDROXYMETHYLTRANSFERASE>

g4f08a1.r1	900	8.2e-99	36 692	sp O13426 GLYC_CANAL SERINE HYDROXYMETHYLTRANSFERASE, CYTOSOLIC (SERINEMETHYLASE) (GLYCINE HYDROXYMETHYLTRANSFE
Contig1363_fle12a1.f1	663	3.1e-94	561 1067	sp O13426 GLYC_C SERINE HYDROXYMETHYLTRANSFERASE, CYTOSOLIC (SERINEMETHYLASE) (GLYCINE HYDROXYMETHYLTRANSFERASE) (SHMT
g4f08a1.f1	144	1.7e-06	389 592	sp P34898 GLYC_NEUCR SERINE HYDROXYMETHYLTRANSFERASE, CYTOSOLIC (SERINEMETHYLASE) (GLYCINE HYDROXYMETHYLTRANSFE=N. crassa

b.glycosylation

<glycosylation>

c7g06a1.f1	492	2.6e-46	195 668	sp P32621 GDA1_YEAST GUANOSINE-DIPHOSPHATASE (GDPASE) >pir A40732guanosine-diphosphatase (EC 3.6.1.42) - yeast
<GPI-ANCHOR TRANSMIDASE> Contig26_dlf08a1.r1	216	1.7e-16	166 321	sp P49018 GPI8_Y GPI-ANCHOR TRANSMIDASE >pir S59796 probable membraneprotein YDR331w - yeast (Saccharomyces cerevisia
<MNN9 PROTEIN> j4f07a1.f1	464	2.4e-43	182 595	sp P53697 MNN9_CANAL MNN9 PROTEIN >gi 1488302 (U63642) Mnn9p [Candidaalbicans]
<UDP-GLUCOSE:GLYCOPROTEIN GLUCOSYLTRANSFERASE> w5a11a1.r1	353	3.6e-30	192 467	gi 860712 (U28735) coded for by C. elegans cDNA cm06e4; coded for by C.elegans cDNA CEESP39F; coded

c.myristoylization

<PEPTIDE N-MYRISTOYLTRANSFERASE>

n0g11a1.r1 495 1.2e-46 8 451 sp|P34763|NMT_AJECA GLYCYLPEPTIDE N-TETRADECANOYLTRANSFERASE
(PEPTIDEN-MYRISTOYLTRANSFERASE) (MYRISTOYL-COA:PR Ajellomyces capsulatus)

d. other

<protein disulfide-isomerase>

Contig1743_e4a07a1.r1 1365 8.1e-139 260 1276 sp|Q12730|PDI_AS PROTEIN DISULFIDE ISOMERASE PRECURSOR (PDI)
>pir||S57942protein disulfide-isomerase (EC 5.3.4.1) - As
Contig89_m5b09a1.r1 320 9.9e-28 172 453 sp|Q00248|PDI_AS PROTEIN DISULFIDE ISOMERASE PRECURSOR
(PDI)>gnl|PID|d1013598 (D85900) protein disulfide isomerase [As

<cyclophilin>

Contig1618_g6g11a1.f1 636 1.1e-61 90 563 sp|P18253|CYPH_S PEPTIDYL-PROLYL CIS-TRANS ISOMERASE (PPIASE)
(ROTAMASE)(CYCLOPHILIN) (CYCLOSPORIN A-BINDING PROTEIN)
Contig1641_e9e07a1.f1 416 2.9e-38 355 744 gi|3288923 (AF071225) cyclophilin B [Rattus norvegicus]
k0f12a1.f1 379 2.6e-34 177 488 sp|P38911|FKB3_YEAST FK506-BINDING NUCLEAR PROTEIN (PEPTIDYL-PROLYL
CIS-TRANSISOMERASE) (PPIASE) (PROLINE ROTAMase)(cyclophilin)
Contig111_k5c02a1.f1 251 8.3e-21 325 990 pir||S62327 cyclophilin-like protein wis2 - fission yeast
(Schizosaccharomycespombe) >gnl|PID|e205292 (X91981) wi

6. Folding and Targeting (68)

a. folding

<CALNEXIN HOMOLOG-folding of glycoproteins>

o4g04a1.r1 510 3e-48 18 515 sp|Q39817|CALX_SOYBN CALNEXIN HOMOLOG PRECURSOR >gi|669003 (U20502)
calnexin[Glycine max]
Contig835_r2e08a1.r1 430 9.5e-40 213 734 sp|P36581|CALX_S CALNEXIN HOMOLOG PRECURSOR >pir||A56106 calnexin
homologcnx1 - fission yeast (Schizosaccharomyces pom

<PEPTIDYL-PROLYL CIS-TRANS ISOMERASE (catalyzes folding)>

<FK506-BINDING PROTEIN-protein folding inhibitor>

Contig1483_c4f02a1.f1 394 6.3e-36 172 492 sp|P48375|FKB2_D 12 KD FK506-BINDING PROTEIN (FKBP)
(PEPTIDYL-PROLYLCIS-TRANS ISOMERASE) (PPIASE) (MACROLIDE BINDING P

b. chaperones

<chaperone>

Contig547_c6f12a1.r1 922 6.8e-92 7 741 gnl|PID|e290095 (Y08867) putative ER chaperone [Aspergillus
awamorii]>gnl|PID|e290123 (Y08868) putative ER chaperone
o0h11a1.r1 693 1.3e-67 10 441 gnl|PID|e290095 (Y08867) putative ER chaperone [Aspergillus
awamorii]>gnl|PID|e290123 (Y08868) putative ER
Contig548_c6f12a1.f1 408 6e-48 148 417 gnl|PID|e290095 (Y08867) putative ER chaperone [Aspergillus
awamorii]>gnl|PID|e290123 (Y08868) putative ER chaperone
Contig991_c4a06a1.f1 370 2.1e-33 103 321 gi|2367623 (AF016187) chaperone/heat shock protein [Emmericella
nidulans]
Contig312_g6c05a1.f1 336 8.9e-30 204 404 gi|2367623 (AF016187) chaperone/heat shock protein [Emmericella
nidulans]

<prefoldin-chaperone which delivers unfolded proteins to another chaperonin>

Contig522_c8d04a1.f1 219 2.1e-17 156 467 gnl|PID|e1297429 (Y17393) prefoldin subunit 2 [Mus musculus]

<heat shock protein Hsp88>

Contig603_c3h04a1.r1 895 5.2e-89 255 1115 gi|3242972 (AF069523) heat shock protein Hsp88 [Neurospora crassa]

Contig540_c7e04a1.f1 142 4.8e-08 408 524 gi|3242972 (AF069523) heat shock protein Hsp88 [Neurospora crassa]

<heat-shock protein30>

Contig1840_c1c01a1.f1 933 4.7e-93 84 626 sp|P40920|HS30_E 30 KD HEAT SHOCK PROTEIN >pir||S50131 heat-shock protein30 - Emericella nidulans >gnl|PID|d1007414 (D

Contig1864_a0e10a1.f1 933 4.8e-93 323 865 sp|P40920|HS30_E 30 KD HEAT SHOCK PROTEIN >pir||S50131 heat-shock protein30 - Emericella nidulans >gnl|PID|d1007414 (D

Contig1800_c6d03a1.f1 855 8.9e-85 451 990 sp|P40920|HS30_E 30 KD HEAT SHOCK PROTEIN >pir||S50131 heat-shock protein30 - Emericella nidulans >gnl|PID|d1007414 (D

Contig1820_a5c12a1.f1 832 2.4e-82 101 640 sp|P40920|HS30_E 30 KD HEAT SHOCK PROTEIN >pir||S50131 heat-shock protein30 - Emericella nidulans >gnl|PID|d1007414 (D

Contig1866_c1c02a1.f1 816 1.2e-80 342 881 sp|P40920|HS30_E 30 KD HEAT SHOCK PROTEIN >pir||S50131 heat-shock protein30 - Emericella nidulans >gnl|PID|d1007414 (D

Contig1772_a0a02a1.r1 798 9.6e-79 88 615 sp|P40920|HS30_E 30 KD HEAT SHOCK PROTEIN >pir||S50131 heat-shock protein30 - Emericella nidulans >gnl|PID|d1007414 (D

Contig1612_g7f07a1.r1 733 7.5e-72 95 583 sp|P40920|HS30_E 30 KD HEAT SHOCK PROTEIN >pir||S50131 heat-shock protein30 - Emericella nidulans >gnl|PID|d1007414 (D

Contig1812_g4b07a1.f1 612 4.8e-59 249 800 sp|P40920|HS30_E 30 KD HEAT SHOCK PROTEIN >pir||S50131 heat-shock protein30 - Emericella nidulans >gnl|PID|d1007414 (D

Contig1862_c1b11a1.f1 342 2.1e-30 261 758 sp|P19752|HS30_N 30 KD HEAT SHOCK PROTEIN >pir||A38360 heat shock protein30 - Neurospora crassa >gi|168820 (M55672) he

s8b01a1.f1 143 1.1e-22 119 280 sp|P40920|HS30_EMENI 30 KD HEAT SHOCK PROTEIN >pir||S50131 heat-shock protein30 - Emericella nidulans >gnl|PID|

Contig1576_f5d04a1.f1 189 3.4e-14 311 520 sp|P40920|HS30_E 30 KD HEAT SHOCK PROTEIN >pir||S50131 heat-shock protein30 - Emericella nidulans >gnl|PID|d1007414 (D

Contig917_c3g11a1.f1 181 2.4e-13 312 521 sp|P40920|HS30_E 30 KD HEAT SHOCK PROTEIN >pir||S50131 heat-shock protein30 - Emericella nidulans >gnl|PID|d1007414 (D

<heat shock protein 70>

Contig1610_a1d03f2.f1 1682 1.9e-172 19 1257 gi|1498496 (U64207) heat shock protein 70 [Penicillium citrinum]

Contig1144_x7a06a1.f1 882 1.2e-87 176 712 gnl|PID|e267541 (X98931) heat shock protein 70 [Emericella nidulans]

d2d10a1.f1 634 7.6e-83 155 703 gnl|PID|e267541 (X98931) heat shock protein 70 [Emericella nidulans]

r5g03a1.r1 667 6.4e-65 84 671 sp|P22774|HS7M_SCHPO MITOCHONDRIAL HEAT SHOCK 70 KD PROTEIN

PRECURSOR>pir||S18670 heat shock protein SSP1 precu

*Contig1366_c3d01a1.r1 356 3.9e-31 229 513 sp|P22774|HS7M_S MITOCHONDRIAL HEAT SHOCK 70 KD PROTEIN

PRECURSOR>pir||S18670 heat shock protein SSP1 precursor - fiss

Contig1567_c3d01a1.f1 127 9.5e-05 449 664 sp|P22774|HS7M_S MITOCHONDRIAL HEAT SHOCK 70 KD PROTEIN

PRECURSOR>pir||S18670 heat shock protein SSP1 precursor - fiss

<HEAT SHOCK PROTEIN HSP1>

Contig113_k5b07a1.f1	275	1.3e-49	205 465	sp P40292 HS82_A	HEAT SHOCK PROTEIN HSP1 (65 KD IGE-BINDING PROTEIN)>gi 1930153 (U92465) heat shock protein [Aspergill
k5b07a1.r1	371	1.6e-33	1 306	sp P40292 HS82_ASPFU	HEAT SHOCK PROTEIN HSP1 (65 KD IGE-BINDING PROTEIN)>gi 1930153 (U92465) heat shock protein
Contig677_a0d04a1.f1	299	6.4e-26	22 195	sp P40292 HS82_A	HEAT SHOCK PROTEIN HSP1 (65 KD IGE-BINDING PROTEIN)>gi 1930153 (U92465) heat shock protein [Aspergill
<HEAT SHOCK PROTEIN 104>					
Contig1831_c3b01a1.r1	1354	1.2e-137	3 1691	sp P31539 H104_Y	HEAT SHOCK PROTEIN 104 >pir S61476 heat shock protein104 - yeast (Saccharomyces cerevisiae) >gi 5578
*Contig981_e9e06a1.r1	386	5.2e-34	240 668	sp P31539 H104_Y	HEAT SHOCK PROTEIN 104 >pir S61476 heat shock protein104 - yeast (Saccharomyces cerevisiae) >gi 5578
Contig1489_d4a01a1.r1	231	3.6e-16	74 631	sp P31539 H104_Y	HEAT SHOCK PROTEIN 104 >pir S61476 heat shock protein104 - yeast (Saccharomyces cerevisiae) >gi 5578
<T-COMPLEX PROTEIN-chaperone of actin, tubulin>					
m8h03a1.r1	750	1.2e-73	63 647	sp P39078 TCPD_YEAST	T-COMPLEX PROTEIN 1, DELTA SUBUNIT (TCP-1-DELTA)(CCT-DELTA) >pir S67690 chaperonin ANC2,
c9e10a1.r1	622	4.5e-60	24 482	sp P87153 TCPH_SCHPO	PROBABLE T-COMPLEX PROTEIN 1, ETA SUBUNIT (TCP-1-ETA)(CCT-ETA) >gnl PID e315886 (Z95397) C
k5e01a1.r1	584	4.8e-56	3 521	sp P12612 TCPA_YEAST	T-COMPLEX PROTEIN 1, ALPHA SUBUNIT (TCP-1-ALPHA)(CCT-ALPHA) >pir A39793 TCP1 protein - ye
Contig83_l5b12a1.f1	470	5.5e-44	204 719	sp P87153 TCPH_S	PROBABLE T-COMPLEX PROTEIN 1, ETA SUBUNIT (TCP-1-ETA)(CCT-ETA) >gnl PID e315886 (Z95397) Cct7p [Schiz
k5e01a1.f1	230	6.1e-32	395 607	sp P28769 TCPA_ARATH	T-COMPLEX PROTEIN 1, ALPHA SUBUNIT (TCP-1-ALPHA)(CCT-ALPHA) >pir JN0448 t-complex polypep, chaperonin for acitn, tubulin
x9d05a1.r1	348	1.3e-30	52 348	sp P87153 TCPH_SCHPO	PROBABLE T-COMPLEX PROTEIN 1, ETA SUBUNIT (TCP-1-ETA)(CCT-ETA) >gnl PID e315886 (Z95397) C
Contig697_p0a05a1.f1	132	4.2e-07	150 236	gnl PID e1314064	(AL031174) t-complex protein 1 gamma subunit homolog[Schizosaccharomyces pombe]
<complex I intermediate associated protein-chaperone in assembly of NADH: Ubiquinone Oxidoreductase>					
w4h08a1.r1	162	1.4e-23	88 354	gnl PID e1198586	(AJ001726) complex I intermediate associated protein CIA35[Neurospora crassa]
w4h08a1.f1	217	3.5e-17	167 430	gnl PID e1198586	(AJ001726) complex I intermediate associated protein CIA35[Neurospora crassa]
<DNAJ-LIKE PROTEIN>					
Contig844_x8c07a1.r1	130	2.4e-07	7 141	sp P50027 DNJH_S	DNAJ-LIKE PROTEIN SLR0093 >gnl PID d1011217 (D64004) DnaJ[Synechocystis sp.]
c.protein sorting					
<protein sorting>					
d3c09a1.r1	344	1e-28	94 873	sp Q07878 VP13_YEAST	VACUOLAR PROTEIN SORTING-ASSOCIATED PROTEIN VPS13>pir S64791 probable membrane protein YL
<CARBOXYPEPTIDASE Y-sorting of vacuolar protein>					

Contig1585_c3d11a1.r1 1431 8.1e-146 9 1142 sp|P30574|CBPY_C CARBOXYPEPTIDASE Y PRECURSOR (CARBOXYPEPTIDASE YSCY)
<MVP1 PROTEIN-vacuolar protein sorting>
Contig275_g9f06a1.r1 393 7.9e-36 29 700 sp|P40959|MVP1_Y MVP1 PROTEIN >pir||S53033 MVP1 protein -
yeast(Saccharomyces cerevisiae) >gi|728651 (Z48613) unknown[
Contig1203_g9f06a1.f1 225 3.3e-17 241 633 sp|P40959|MVP1_Y MVP1 PROTEIN >pir||S53033 MVP1 protein -
yeast(Saccharomyces cerevisiae) >gi|728651 (Z48613) unknown[
<vacuolar protein sorting homolog h-vps45>
f5f09a1.r1 463 3.2e-43 31 699 gi|1477466 (U35246) vacuolar protein sorting homolog h-vps45
[Homo sapiens]
<clathrin>
g3d10a1.r1 650 1e-61 28 768 sp|P22137|CLH_YEAST CLATHRIN HEAVY CHAIN >pir||A36349 clathrin heavy chain
1 -yeast (Saccharomyces cerevisiae)
Contig1038_o5c12a1.r1 219 2.1e-17 478 774 gnl|PID|d1026032 (AB011822) clathrin light chain [Schizosaccharomyces
pombe]
Contig504_c9g08a1.f1 210 1.1e-15 140 277 sp|Q00776|AP54_Y CLATHRIN COAT ASSEMBLY PROTEIN AP54 (CLATHRIN
COATASSOCIATED PROTEIN AP54) (GOLGI ADAPTOR AP-1 54 KD
Contig826_w4e09a1.f1 147 3.9e-08 134 343 sp|Q10161|CLH_SC PROBABLE CLATHRIN HEAVY CHAIN >gnl|PID|e220677
(Z69240)clathrin heavy chain [Schizosaccharomyces pomb
<RIBOSYLATION FACTOR>
Contig1661_k5d07a1.r1 904 5.7e-90 101 643 sp|P34727|ARF_AJ ADP-RIBOSYLATION FACTOR >pir||D49993
ADP-ribosylationfactor - Ajellomyces capsulata >gi|407693 (L2511
<SIGNAL RECOGNITION PARTICLE>
Contig1155_x7d07a1.f1 538 3.2e-51 428 778 sp|Q00179|SR54_A SIGNAL RECOGNITION PARTICLE 54 RD PROTEIN
HOMOLOG>pir||JC4572 signal recognition particle 54K protein
r1f06a1.f1 130 1.1e-07 228 596 sp|P36057|SRPB_YEAST PUTATIVE SIGNAL RECOGNITION PARTICLE RECEPTOR
BETASUBUNIT (SR-BETA) >pir||S37984 probable
<MITOCHONDRIAL IMPORT RECEPTOR>
Contig294_n0a07a1.f1 114 2.9e-06 501 659 gnl|PID|d1022291 (AB004538) MITOCHONDRIAL IMPORT RECEPTOR SUBUNIT
TOM40[Schizosaccharomyces pombe]
<synaptobrevin-protein trafficking>
Contig1563_e4a05a1.f1 375 6.5e-34 434 709 gi|2769755 (AF010288) synaptobrevin [Aspergillus parasiticus]
*Contig965_m3h03a1.r1 195 2.5e-14 186 653 sp|Q16943|VP33_A VESICLE-ASSOCIATED MEMBRANE PROTEIN/SYNAPTOBREVIN
BINDINGPROTEIN (VAP-33) >pir||A57245 VAMP-binding p
Contig830_z4b01a1.f1 156 1e-10 356 550 sp|P47192|SYBR_A SYNAPTOBREVIN-RELATED PROTEIN >gi|600710
(M90418)formerly called HAT24; synaptobrevin-related protein
<VESICLE TRANSPORT V-SNARE PROTEIN>
*Contig987_c3f10a1.r1 319 5.6e-28 262 891 sp|P78768|VTI1_S VESICLE TRANSPORT V-SNARE PROTEIN VTI1
HOMOLOG>gnl|PID|d1014475 (D89116) similar to Saccharomyces cer
<COATOMER ALPHA SUBUNIT-trafficcing to golgi, nonclathrin vesicles>
Contig577_c5b03a1.f1 1366 6.5e-139 3 815 gi|3170523 (AF053883) coatomer alpha subunit [Emericella
nidulans)
<COATOMER BETA SUBUNIT-trafficcing to golgi, nonclathrin vesicles>
r2a12a1.f1 566 2e-53 278 805 sp|P23514|COPB_RAT COATOMER BETA SUBUNIT (BETA-COAT PROTEIN)

gld10a1.r1 398 2.9e-35 29 679 (BETA-COP)>pir||s13520 beta-COP protein - rat >g
sp|P41810|COPB_YEAST COATOMER BETA SUBUNIT (BETA-COAT PROTEIN)
i7g05a1.r1 302 3.9e-26 1 468 (BETA-COP)>pir||s54534 coatomer complex beta cha
gi|2809537 (AF043120) beta prime coatomer protein [Mus musculus]
<COATOMER ZETA SUBUNIT-traffic to golgi, nonclathrin vesicles>
k9f10a1.f1 137 4.1e-08 293 535 sp|P53600|COPZ_YEAST COATOMER ZETA SUBUNIT (ZETA-COAT PROTEIN)
(ZETA-COP)>pir||s52521 hypothetical protein YPL0
<COATOMER GAMMA SUBUNIT>
Contig178_i8d09a1.r1 171 5.2e-11 9 371 sp|P87140|COPG_S PROBABLE COATOMER GAMMA SUBUNIT (GAMMA-COAT
PROTEIN) (GAMMA-COP) >gnl|PID|e316116 (Z95396) unknown [Sc
<NPL6 PROTEIN-nuclear protein localization>
Contig174_i8g02a1.r1 166 2.3e-09 282 518 sp|P32832|NPL6_Y NPL6 PROTEIN >pir||s30792 NPL6 protein -
yeast(Saccharomyces cerevisiae) >gi|172050 (M98434) nuclear

7.Turnover-protein degradation-including vacuolar (91)

<protein-L-isoaspartate O-methyltransferase-esterification for degradation>

Contig1569_g3g03a1.f1 490 4.4e-46 761 1474 sp|Q27869|PIMT_D PROTEIN-L-ISOASPARTATE(D-ASPARTATE)
O-METHYLTRANSFERASE (PROTEIN-BETA-ASPARTATE METHYLTRANSFERASE) (PI

<PROTEASE REGULATORY SUBUNIT>

Contig1452_m2d09a1.f1 1039 2.8e-104 42 767 sp|P33299|PRS7_Y 26S PROTEASE REGULATORY SUBUNIT 7 HOMOLOG (CIM5
PROTEIN) (TAT-BINDING HOMOLOG 3) >pir||s34354 tat-bind
Contig636_c0h10a1.f1 960 6.9e-96 108 845 sp|P41836|PRS8_S 26S PROTEASE REGULATORY SUBUNIT 8 HOMOLOG (LET1
PROTEIN)>pir||s45176 transcription factor SUG1 homolo
Contig656_o8g12a1.r1 851 2.4e-84 7 822 sp|P33297|PRSA_Y 26S PROTEASE REGULATORY SUBUNIT S6A (TAT-BINDING
PROTEINHOMOLOG 1) (TBP-1) >pir||s46605 YTA1 protein
Contig1319_g6f01a1.r1 810 5.2e-80 102 653 sp|P78578|PRS6_A 26S PROTEASE REGULATORY SUBUNIT 6B HOMOLOG
>gi|1777414(U15601) 26S proteasome subunit [Aspergillus ni
o8g12a1.f1 588 1.7e-56 129 614 sp|P33297|PRSA_YEAST 26S PROTEASE REGULATORY SUBUNIT S6A (TAT-BINDING
PROTEINHOMOLOG 1) (TBP-1) >pir||s46605 YT
Contig634_c0h10a1.r1 581 9.6e-56 126 821 sp|P47210|PRS8_H 26S PROTEASE REGULATORY SUBUNIT 8 (PROTEASOME
SUBUNITP45) (THYROID HORMONE RECEPTOR INTERACTING PROTE
Contig717_u4g08a1.r1 338 5.6e-30 151 477 sp|P33299|PRS7_Y 26S PROTEASE REGULATORY SUBUNIT 7 HOMOLOG (CIM5
PROTEIN) (TAT-BINDING HOMOLOG 3) >pir||s34354 tat-bind

<proteasome>

Contig1183_g6f01a1.f1 930 1.1e-92 203 826 sp|P78578|PRS6_A 26S PROTEASE REGULATORY SUBUNIT 6B HOMOLOG
>gi|1777414(U15601) 26S proteasome subunit [Aspergillus ni
Contig808_z4f05a1.f1 780 7.6e-77 106 735 sp|Q09682|PRC9_S PUTATIVE PROTEASOME COMPONENT C9/Y13 (MACROPAIN
SUBUNIT) (MULTICATALYTIC ENDOPEPTIDASE COMPLEX SUBUNIT
Contig1547_m8e08a1.f1 696 6.2e-68 139 867 sp|P25786|PRC2_H PROTEASOME COMPONENT C2 (MACROPAIN SUBUNIT
C2) (PROTEASOME NU CHAIN) (MULTICATALYTIC ENDOPEPTIDASE COM
Contig990_m8e02a1.f1 691 2.1e-67 27 683 gnl|PID|e1263959 (AL022117) putative proteasome subunit
[Schizosaccharomycespombe]
Contig1453_u4b10a1.r1 681 2.5e-66 85 786 sp|P23724|PRC5_Y POTENTIAL PROTEASOME COMPONENT C5

Contig721_t2e09a1.f1	560	7.4e-53	126 677	(MULTICATALYTIC ENDOPEPTIDASE COMPLEX SUBUNIT C5) >pir 842436 multi gnl PID d1011888 (D78151) human 26S proteasome subunit p97 [Homo sapiens]
Contig1033_d1f02a1.f1	544	8.2e-52	106 639	gi 476044 (X78991) proteasomal subunit Pre3 [Saccharomyces cerevisiae]>gi 854572 (X87611) proteasome component
n0f06a1.r1	438	1.3e-40	119 457	sp P21242 PRC8_YEAST PROTEASOME COMPONENT C1 (MACROPAIN SUBUNIT C1)(PROTEINASE YSCE SUBUNIT 1) (MULTICATALYTIC
l5h10a1.f1	325	1.3e-28	90 518	gnl PID d1020529 (AB003102) 26S proteasome subunit p44.5 [Homo sapiens]
z2a09a1.f1	280	1.8e-23	154 504	gnl PID d1020530 (AB003103) 26S proteasome subunit p55 [Homo sapiens]
k5a04a1.r1	262	1.2e-21	156 614	gi 3450889 (AF083890) 20S proteasome subunit 9 [Arabidopsis thaliana]
n0f06a1.f1	216	3.7e-17	205 429	sp P21242 PRC8_YEAST PROTEASOME COMPONENT C1 (MACROPAIN SUBUNIT C1)(PROTEINASE YSCE SUBUNIT 1) (MULTICATALYTIC
<CLPB PROTEIN>				
Contig1439_n3d06a1.f1	281	8.9e-23	261 623	sp P53532 CLPB_C CLPB PROTEIN >gi 1163118 (U43536) heat-inducible expression; two ATP-binding domains; ClpB homolog, si
<ubiquitin>				
Contig1865_alg03f2.f1	1457	1.4e-148	142 1023	pir D29456 ubiquitin precursor UBI4 - yeast (Saccharomyces cerevisiae)>gi 4734 (X05731) ubiquitin (AA 1-381) [Sa
Contig1048_d3c02a1.r1	1091	9.2e-110	104 1201	sp P22515 UBA1_Y UBIQUITIN-ACTIVATING ENZYME E1 1
Contig1479_j7d10a1.f1	1084	2.6e-107	271 1098	>pir S38048ubiquitin--protein ligase (EC 6.3.2.19) - yeast (Sacchar
Contig1475_a0f02a1.r1	958	1e-95	198 782	gnl PID e351301 (Z99531) ubiquitin system protein [Schizosaccharomyces pombe]
o8f12a1.r1	896	4.3e-89	3 554	pir D29456 ubiquitin precursor UBI4 - yeast (Saccharomyces cerevisiae)>gi 4734 (X05731) ubiquitin (AA 1-381) [Sa
Contig1650_d3c02a1.f1	856	7e-85	130 1023	gi 2262193 (U62795) ubiquitin ligase Pub1 [Schizosaccharomyces pombe]>gi 2408007 (Z99161) ubiquitin ligase
m8d11a1.r1	788	1.1e-77	42 827	gi 4715 (X55386) ubiquitin-activating enzyme [Saccharomyces cerevisiae]
Contig1093_k0e12a1.f1	469	1.5e-74	293 577	sp Q09879 UBPB_SCHPO PUTATIVE UBIQUITIN CARBOXYL-TERMINAL HYDROLASE C8A4.01C(UBIQUITIN THIOLESTERASE) (UBIQUITI
Contig1509_n8a02a1.r1	622	3.9e-60	22 390	gnl PID e1251311 (AJ223328) polyubiquitin [Nicotiana tabacum]>gnl PID e1311722 (AJ007936) polyubiquitin [Gibberella pu
Contig621_o8a06a1.r1	609	9.5e-59	181 561	pir A29456 ubiquitin / ribosomal protein CEP52 - yeast (Saccharomyces cerevisiae) >gi 4728 (X05728) ubiquitin [Sa
Contig271_g9h01a1.f1	521	2.2e-49	348 803	gi 166336 (M88684) polyubiquitin [Aglaothamnion neglectum]
Contig53_m3f08a1.r1	437	1.8e-40	24 356	>prf 1908440Apoly-ubiquitin [Aglaothamnion neglectu
Contig376_g1d09a1.r1	436	2.7e-40	29 403	gi 3309661 (AF075599) ubiquitin conjugating enzyme 12 [Homo sapiens]
Contig24_a0f02a1.f1	388	2.6e-35	242 484	gi 1143188 (U32627) ubiquitin precursor [Candida albicans]
				gi 2408071 (Z99166) ubiquitin fusion degradation protein [Schizosaccharomyces pombe]
				gi 571519 (U16852) polyubiquitin [Gracilaria verrucosa]

c4a04a1.f1	355	8.6e-32	139 558	>prf 2109223Apoly-ubiquitin [Gracilaria verrucosa] sp P14682 UBC3_YEAST UBIQUITIN-CONJUGATING ENZYME E2-34 KD (UBIQUITIN-PROTEINLIGASE) (UBIQUITIN CARRIER PROTEIN)
Contig389_f2b09a1.r1	349	3.5e-30	323 754	sp P39940 RSP5_Y UBIQUITIN--PROTEIN LIGASE RSP5 >pir S43217 hypotheticalprotein YER125w - yeast (Saccharomyces cerevi
g3h09a1.r1	330	3.8e-29	80 505	sp Q92353 UBPC_SCHPO PUTATIVE UBIQUITIN CARBOXYL-TERMINAL HYDROLASE C6G9.08(UBIQUITIN THIOLESTERASE) (UBIQUITIN
Contig1105_c6c07a1.f1	324	1.7e-28	3 299	gi 3265058 (AF060232) monoubiquitin/carboxy extension protein fusion[Botryotinia fuckeliana]
Contig98_k9g12a1.f1	320	4.4e-28	335 562	gnl PID e354806 (AJ003818) ubiquitin-like protein [Schizosaccharomyces pombe]
r6c08a1.r1	283	9.4e-23	143 541	sp P39538 UBPC_YEAST UBIQUITIN CARBOXYL-TERMINAL HYDROLASE 12 (UBIQUITINTHIOLESTERASE 12) (UBIQUITIN-SPECIFIC P
Contig1277_j4a05a1.f1	257	2.1e-21	173 424	sp Q12306 SMT3_Y UBIQUITIN-LIKE PROTEIN SMT3 >pir S63999 SMT3 protein -yeast (Saccharomyces cerevisiae) >gi 881372 (U
Contig1352_g2f11a1.f1	262	7.8e-21	452 706	gi 2262193 (U62795) ubiquitin ligase Pub1 [Schizosaccharomyces pombe]>gi 2408007 (Z99161) ubiquitin ligase [Schi
d3d05a1.f1	247	3.7e-19	93 359	gi 1843535 (U82122) E6-AP ubiquitin-protein ligase [Mus musculus]
Contig1818_d2g09a1.f1	226	3.8e-18	231 377	pir S62680 ubiquitin-extension protein - Emericella nidulans
z7e03a1.f1	226	3.8e-18	185 604	sp P15374 UBL3_HUMAN UBIQUITIN CARBOXYL-TERMINAL HYDROLASE ISOZYME L3 (UCH-L3)(UBIQUITIN THIOLESTERASE L3) >pir
v3g05a1.f1	229	8.2e-18	212 556	sp Q09738 UBPA_SCHPO PUTATIVE UBIQUITIN CARBOXYL-TERMINAL HYDROLASE C13A11.04C(UBIQUITIN THIOLESTERASE) (UBIQUI
e9g11a1.f1	230	2.1e-17	135 515	gnl PID e1251102 (AL021838) ubiquitin carboxyl-terminal hydrolase[Schizosaccharomyces pombe]
Contig436_e0c04a1.f1	220	4e-17	210 500	sp P48510 DSK2_Y UBIQUITIN-LIKE PROTEIN DSK2 >gi 786151 (L40587)ubiquitin-like protein [Saccharomyces cerevisiae]
o0f09a1.r1	213	8.8e-15	31 429	gnl PID e351301 (Z99531) ubiquitin system protein [Schizosaccharomyces pombe]
e0e09a1.r1	182	1.2e-12	318 608	sp Q92353 UBPC_SCHPO PUTATIVE UBIQUITIN CARBOXYL-TERMINAL HYDROLASE C6G9.08(UBIQUITIN THIOLESTERASE) (UBIQUITIN
Contig527_c8b10a1.f1	174	1.8e-09	330 785	gnl PID e1298610 (AL023859) putative ubiquitin protein ligase[Schizosaccharomyces pombe]
*Contig1346_alb01c9.r1	142	7e-09	189 515	gi 2668744 (AF034946) ubiquitin conjugating enzyme [Zea mays]
Contig18_q0b07a1.f1	114	2.9e-06	427 510	pir S43786 ubiquitin--protein ligase (EC 6.3.2.19) - Arabidopsis thaliana
<UBIQUITIN-CONJUGATING ENZYME>				
Contig1649_m8h12a1.f1	632	3.8e-61	394 942	sp P16577 UBC4_W UBIQUITIN-CONJUGATING ENZYME E2-23 KD (UBIQUITIN-PROTEINLIGASE) (UBIQUITIN CARRIER PROTEIN) >pir A34
Contig939_c7g10a1.f1	318	1.5e-55	450 635	gi 3323498 (AF030296) ubiquitin conjugating enzyme UBC1 {glomerella cingulata}
d3d05a1.r1	293	4.3e-24	129 602	gi 1872514 (U84404) E6-associated protein E6-AP/ubiquitin-protein ligase [Homo sapiens] >gi 2361031 (A

Contig1112_c6d08a1.f1	206	5.5e-16	501 677	sp O00102 UBC7_S	UBIQUITIN-CONJUGATING ENZYME E2-18 KD (UBIQUITIN-PROTEINLIGASE) (UBIQUITIN CARRIER PROTEIN) >gnl PID
*Contig797_y6b01a1.f1	195	8e-15	95 217	sp P52493 UBC2_N	UBIQUITIN-CONJUGATING ENZYME E2-17 KD (UBIQUITIN-PROTEINLIGASE 2) (UBIQUITIN CARRIER PROTEIN) >gnl PI
<pepsinogen>					
Contig200_i2g09a1.r1	918	1.9e-91	72 653	gi 530795	(U03278) pepsinogen [Aspergillus niger]
m3c08a1.r1	288	1.1e-24	277 567	gi 530795	(U03278) pepsinogen [Aspergillus niger]
e4h01a1.f1	247	4.7e-20	16 159	gi 530795	(U03278) pepsinogen [Aspergillus niger]
<aspartic protease>					
m5f01a1.f1	181	4.6e-11	205 642	sp P32329 YAP3_YEAST	ASPARTIC PROTEINASE 3 PRECURSOR (YAPSIN 1) >pir S64957aspergillopepsin I (EC 3.4.23.18) Y
Contig117_m5f01a1.r1	119	0.0001	299 547	sp Q12303 YL12_Y	PUTATIVE ASPARTYL PROTEASE YLR121C PRECURSOR >pir S64958probable membrane protein YLR121c - yeast (S
<proline peptidase>					
<aminopeptidase>					
*Contig1816_alh01c9.r1	991	3.7e-99	668 1759	sp P38174 AMP2_Y	METHIONINE AMINOPEPTIDASE 2 (METAP 2) (PEPTIDASE M 2)>gi 1045302 (U17437) methionine aminopeptidase 2
Contig1347_g3b06a1.f1	915	4.1e-91	172 1263	sp P37302 APE3_Y	AMINOPEPTIDASE Y PRECURSOR >pir A54134 aminopeptidase Y(EC 3.4.11.-) - yeast (Saccharomyces cerevisi
Contig1504_o3d01a1.f1	624	2.6e-60	237 1124	gi 3366	(Y07522) aminopeptidase yscI (AA 1-514) [Saccharomyces cerevisiae]
Contig946_r7a12a1.r1	586	5.9e-56	30 1268	gi 3368	(X63998) aminopeptidase yscII [Saccharomyces cerevisiae]
p0f10a1.f1	428	1.6e-39	65 490	gnl PID e339951	(Z98980) aminopeptidase i precursor [Schizosaccharomycespombe]
Contig501_c9h01a1.f1	399	1.9e-36	4 504	sp P38174 AMP2_Y	METHIONINE AMINOPEPTIDASE 2 (METAP 2) (PEPTIDASE M 2)>gi 1045302 (U17437) methionine aminopeptidase 2
c9a01a1.r1	319	6.2e-27	58 531	sp P18962 DAP2_YEAST	DIPEPTIDYL AMINOPEPTIDASE B (DPAP B) (YSCV) >pir S46780hypothetical protein YHR028c - yea
Contig210_m7d03a1.f1	286	1.9e-24	170 418	pir S45411	methionyl aminopeptidase (EC 3.4.11.18) MAP2 - yeast(Saccharomyces cerevisiae) >gi 496684 (X79489) D-
w6h10a1.f1	294	3.3e-24	9 470	sp P18962 DAP2_YEAST	DIPEPTIDYL AMINOPEPTIDASE B (DPAP B) (YSCV) >pir S46780hypothetical protein YHR028c - yea
j7a03a1.r1	226	1.2e-15	118 528	gi 2773225	(AF039716) Similar to aminopeptidase; coded for by c. elegans cDNAYk91g4.3; coded for by C
j7a03a1.f1	148	1.8e-07	98 358	gnl PID e1169567	(X95762) Aminopeptidase P-like [Homo sapiens]
<iminopeptidase>					
g7b07a1.r1	378	3.3e-34	70 597	sp P46547 PIP_AERSO	PROLINE IMINOPEPTIDASE (PROLYL AMINOPEPTIDASE)>pir JC4184 prolyl aminopeptidase (EC 3.4.1
g7b07a1.f1	255	8.2e-21	163 552	sp P46547 PIP_AERSO	PROLINE IMINOPEPTIDASE (PROLYL AMINOPEPTIDASE)>pir JC4184 prolyl aminopeptidase (EC 3.4.1
<serine protease>					
Contig322_g5d04a1.r1	742	8.4e-73	89 760	gnl PID e319086	(Y13338) intracellular vacuolar serine proteinase

n0c11a1.r1	602	5e-58	4 462	[Aspergillusfumigatus] gnl PID e319086 (Y13338) intracellular vacuolar serine proteinase [Aspergillusfumigatus]
<ASPARTATE PROTEASE> o8c12a1.r1	391	1.2e-35	104 550	gi 1469396 (U43775) secreted aspartic proteinase precursor [Glomerellacingulata]
<metallopeptidase> Contig1854_a5c06a1.f1	625	5.2e-78	271 957	sp P46073 ME24_A 24 KD METALLOPROTEINASE PRECURSOR (DEUTEROLYSIN)>pir JC4378 metalloproteinase (EC 3.4.--.) 23K - Asp
Contig1687_j5b07a1.f1	608	1.4e-58	257 1042	gnl PID e1293248 (AL023590) putative metallopeptidase [Schizosaccharomycespombe]
Contig1855_a1e05c9.r1	524	9.9e-50	99 659	pir JC4379 metalloproteinase (EC 3.4.--.) 23K - Aspergillus fumigatus>gi 780794 (U24146) MEP20 [Aspergillus fumi
Contig235_h4h04a1.f1	462	3.8e-43	277 615	sp P46073 ME24_A 24 KD METALLOPROTEINASE PRECURSOR (DEUTEROLYSIN)>pir JC4378 metalloproteinase (EC 3.4.--.) 23K - Asp
<ca dependent protease> l3d08a1.r1	132	1.4e-05	235 378	prf 1613155A Ca dependent Cys protease p94 [Rattus norvegicus]
<alkaline protease> Contig1828_a5e07a1.f1	1909	1.7e-196	205 1413	gi 470731 (L31778) alkaline protease [Aspergillus nidulans]
<ACID PROTEASE A> x1b07a1.r1	347	5.4e-31	7 444	sp P24665 PRTA_ASPNG ASPERGILLOPEPSIN II PRECURSOR (ACID PROTEASE A) (PROCTASEA) >pir A41025 aspergillopepsin
<CAAX PRENYL PROTEASE-cleavage of alpha factor for activatio> Contig421_e9b02a1.r1	487	8.5e-46	8 544	sp P47154 ST24_Y CAAX PRENYL PROTEASE 1 (PRENYL PROTEIN-SPECIFICENDOPROTEASE 1) (PPSEP 1) (A-FACTOR CONVERTING ENZYME)
n3b02a1.f1	167	5.3e-09	221 427	sp P47154 ST24_YEAST CAAX PRENYL PROTEASE 1 (PRENYL PROTEIN-SPECIFICENDOPROTEASE 1) (PPSEP 1) (A-FACTOR CONVERT
<insulinase-peptidase M16 family> j0g08a1.r1	374	1.1e-32	73 501	sp O14077 YEAC_SCHPO PUTATIVE ZINC-PROTEASE C2E11.12C >gnl PID e339165(Z98850) hypothetical protease=insulinase family
<Lon serine protease> y7c06a1.f1	140	1e-05	373 555	sp P36775 LONM_YEAST MITOCHONDRIAL ATP-DEPENDENT PROTEASE PRECURSOR>pir S43938 proteinase PIM1 (EC 3.4.21.-)=LON gene serine protease

V. Processes

A. Cell rescue, defense, osmotic adaptation, starvation response, development (asexual, sexual)
includes antibiotics, toxins.
See also B.cell signalling, signal transduction and C. transmembrane transport

1. development (8)

a. conidiation-asexual

<velvet A>

Contig1621_u4c01a1.f1 970 6.1e-97 256 1089 gi|3329358 (U95045) velvet A [Emericella nidulans]
 <CONIDIATION-SPECIFIC PROTEIN 6>
 r3c09a1.r1 164 1.5e-11 88 324 sp|P34762|CON6_NEUCR CONIDIATION-SPECIFIC PROTEIN 6 >gi|415714
 (L26036)conidiation protein [Neurospora crassa]

b. pigment production
 <GREEN PIGMENT SYNTHASE>
 g9a11a1.r1 668 1.9e-63 11 478 sp|Q03149|WA_EMENI CONIDIAL GREEN PIGMENT SYNTHASE >pir||s28353
 probablepolyketide synthase - Emericella nidu

<PORPHYRIN>
 <porphyrinogen oxidase>
 Contig458_d4d03a1.r1 522 1.8e-49 116 874 sp|P35055|HEM6_S COPROPORPHYRINOGEN III OXIDASE
 PRECURSOR(COPROPORPHYRINOGENASE) (COPROGEN OXIDASE) >pir||s39905coprop
 Contig857_r3e10a1.f1 314 1.8e-27 110 484 gi|171316 (J03873) coproporphyrinogen oxidase (EC 1.3.3.3)
 [Saccharomycescerevisiae]

<polyketide synthase>
 Contig1214_g9a11a1.f1 664 9.1e-109 483 950 gi|3136092 (AF025541) polyketide synthase [Aspergillus fumigatus]
 Contig295_g7e09a1.f1 279 5.5e-22 1 528 pir||s60224 melanin biosynthetic polyketide synthase PKS1 -
 Colletotrichumlagenarium >gnl|PID|d1019697 (D83643) p

<LACCASE>
 k5d05a1.f1 255 5.2e-20 191 502 sp|P17489|LAC1_EMENI LACCASE PRECURSOR (BENZENEDIOL:OXYGEN
 OXIDOREDUCTASE)(URISHIOL OXIDASE) (LACCASE I) >gi|24

2.defense (41)

a.Secondary metabolites
 -Penicullin production
 <penicill>
 Contig416_n5f11a1.r1 1022 1.8e-102 87 1016 sp|P21133|AAAA_E ACYL-COENZYME
 A:6-AMINOPENICILLANIC-ACID-ACYLTRANSFERASEPRECURSOR (ISOPENICILLIN N
 ACYLTRANSFERASE) >
 r3c06a1.f1 405 4.5e-37 283 513 sp|P21133|AAAA_EMENI ACYL-COENZYME
 A:6-AMINOPENICILLANIC-ACID-ACYLTRANSFERASEPRECURSOR (ISOPENICILLIN N ACYLTRA

-sterigmatocystin biosynthesis
 <sterigmatocystin>
 Contig1677_g2d05a1.f1 1543 1.1e-157 72 983 sp|Q00707|STCL_E PROBABLE STERIGMATOCYSTIN BIOSYNTHESIS P450
 MONOOXYGENASESTCL >gi|1235628 (U34740) putative p450 mono
 Contig1811_alg02c9.r1 1444 3.2e-147 5 1033 gi|1293655 (U51327) versicolorin B synthase [Aspergillus
 parasiticus]>gi|1293657 (U51328) versicolorin B synthas
 Contig1291_c8f01a1.f1 1328 7.6e-134 260 1003 sp|Q12397|STCA_E PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS
 POLYKETIDESYNTHASE (PKS) >gi|972728 (L39121) polyketide syntha
 Contig529_o4e08a1.r1 1310 5.5e-133 49 849 sp|Q00727|STCV_E PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS DEHYDROGENASE
 STCV>gi|1235634 (U34740) putative dehydrogenase
 Contig1627_alc06f2.f1 1231 1.2e-124 61 852 sp|Q00791|STCU_E VERSICOLORIN REDUCTASE >gi|1235633 (U34740)

Contig	Start	End	Score	Gene	Description
Contig1600_m8f12a1.f1	1125	2.3e-113	533 1156	sp Q00730 STCW_E	putativeketoreductase [Emericella nidulans] PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS MONOOXYGENASE
Contig1527_r3b09a1.r1	1107	1.8e-111	268 963	STCW>gi 1235635 (U34740)	putative FAD-containing
Contig1079_alf01c9.r1	845	4.4e-110	66 584	sp Q00674 STCE_E	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS KETOREDUCTASE
c8f01a1.r1	1098	3.4e-109	11 658	STCE>gi 1235622 (U34740)	putative ketoreductase
Contig1442_e9c04a1.r1	1066	3.9e-107	23 847	sp Q12609 STCF_E	PROBABLE STERIGMATOCYSTIN BIOSYNTHESIS P450
n2e10a1.r1	1059	7.8e-106	28 669	MONOOXYGENASESTCF >gi 1235624 (U34740)	putative p450 mono
Contig1158_g5a07a1.r1	1033	1.1e-103	119 715	sp Q12397 STCA_EMENI	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS
Contig736_x7g01a1.f1	993	2.2e-99	45 782	POLYKETIDESYNTASE (PKS) >gi 972728 (L39121)	polyke
Contig21_r4c09a1.r1	973	2.8e-97	85 792	gi 1293655 (U51327)	versicolorin B synthase [Aspergillus parasiticus]>gi 1293657 (U51328)
g9b02a1.f1	939	2.2e-92	120 713	sp Q00681 STCJ_EMENI	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS FATTY
Contig1009_c9f01a1.f1	744	1.8e-85	150 752	ACIDSYNTASE ALPHA SUBUNIT >gi 1235626 (U3474	
Contig1406_d2f11a1.r1	496	2.6e-84	5 283	sp Q00730 STCW_E	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS MONOOXYGENASE
g9h10a1.f1	844	3.9e-82	2 541	STCW>gi 1235635 (U34740)	putative FAD-containing
Contig452_d4h03a1.r1	818	7.3e-81	106 639	sp Q00714 STCS_E	PROBABLE STERIGMATOCYSTIN BIOSYNTHESIS P450
z4c07a1.r1	800	6.1e-79	29 514	MONOOXYGENASESTCS >gi 1235631 (U34740)	putative p450 mono
g9b02a1.r1	799	2.1e-77	13 468	sp Q00674 STCE_E	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS KETOREDUCTASE
c0d01a1.r1	788	4.1e-76	12 476	STCE>gi 1235622 (U34740)	putative ketoreductase
u4e02a1.r1	547	7.4e-71	47 340	sp Q00706 STCK_EMENI	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS FATTY
Contig1592_e4e01a1.f1	701	1.9e-68	217 600	ACIDSYNTASE BETA SUBUNIT >gi 1235627 (U34740	
z7a04a1.r1	630	6.4e-61	19 453	sp Q12609 STCF_E	PROBABLE STERIGMATOCYSTIN BIOSYNTHESIS P450
Contig657_b0f03a1.f1	610	7.9e-59	133 513	MONOOXYGENASESTCF >gi 1235624 (U34740)	putative p450 mono
				sp Q12608 STCB_E	PROBABLE STERIGMATOCYSTIN BIOSYNTHESIS P450
				MONOOXYGENASESTCB >gi 1235620 (U34740)	putative p450 mono
				sp Q12397 STCA_EMENI	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS
				POLYKETIDESYNTASE (PKS) >gi 972728 (L39121)	polyke
				sp Q00675 STCI_E	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS
				LIPASE/ESTERASESTCI >gi 1235625 (U34740)	putative lipase/ester
				sp Q00713 STCQ_EMENI	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS PROTEIN
				STCQ>gi 1235630 (U34740)	similar to A. para
				sp Q00706 STCK_EMENI	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS FATTY
				ACIDSYNTASE BETA SUBUNIT >gi 1235627 (U34740	
				sp Q12397 STCA_EMENI	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS
				POLYKETIDESYNTASE (PKS) >gi 972728 (L39121)	polyke
				sp Q00707 STCL_EMENI	PROBABLE STERIGMATOCYSTIN BIOSYNTHESIS P450
				MONOOXYGENASESTCL >gi 1235628 (U34740)	putativ
				sp Q00675 STCI_E	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS
				LIPASE/ESTERASESTCI >gi 1235625 (U34740)	putative lipase/ester
				sp Q00710 STCO_EMENI	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS PROTEIN
				STCO>gi 1235629 (U34740)	similar to A. para
				sp Q00727 STCV_E	PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS DEHYDROGENASE
				STCV>gi 1235634 (U34740)	putative dehydrogenase

x7g01a1.r1	562	1e-53	45 572	sp Q00714 STCS_EMENI PROBABLE STERIGMATOCYSTIN BIOSYNTHESIS P450 MONOOXYGENASESTCS >gi 1235631 (U34740) putativ
Contig961_m3e08a1.r1	532	1.5e-50	62 595	sp Q00727 STCV_E PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS DEHYDROGENASE STCV>gi 1235634 (U34740) putative dehydrogenase
Contig1619_c5e01a1.f1	529	3.2e-50	547 906	sp Q00713 STCQ_E PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS PROTEIN STCQ>gi 1235630 (U34740) similar to A. parasiticus put
h4g09a1.f1	397	3e-36	206 463	sp Q00717 STCT_EMENI PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS PROTEIN STCT>gi 1235632 (U34740) putative translati
c0f02a1.f1	391	1.2e-35	171 704	gi 1200177 (U24698) norsolorinic acid reductase [Aspergillus parasiticus]
Contig779_z5d08a1.r1	302	3.5e-26	3 188	sp Q00674 STCE_E PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS KETOREDUCTASE STCE>gi 1235622 (U34740) putative ketoreductase
h8e06a1.f1	226	4.3e-18	169 456	sp Q00668 STCC_EMENI PUTATIVE STERIGMATOCYSTIN BIOSYNTHESIS PEROXIDASE STCC>gi 1235621 (U34740) putative peroxi
d5d05a1.r1	174	4.1e-10	46 468	sp Q12608 STCB_EMENI PROBABLE STERIGMATOCYSTIN BIOSYNTHESIS P450 MONOOXYGENASESTCB >gi 1235620 (U34740) putativ

b.other

<clavulanate-inhibits beta-lactamases>

<VEGETABLE INCOMPATIBILITY PROTEIN-vegetative incompatibility,defense>

Contig820_z4c11a1.f1	134	8.6e-07	6 251	sp Q00808 HET1_P VEGETATIBLE INCOMPATIBILITY PROTEIN HET-E-1 >gi 607003(L28125) beta transducin-like protein [Podospor
x5f10a1.r1	146	6.5e-06	81 326	sp Q00808 HET1_PODAN VEGETATIBLE INCOMPATIBILITY PROTEIN HET-E-1 >gi 607003(L28125) beta transducin-like protei

<pisatin demethylase-inactivates plant compound pisatin>

x8d06a1.f1	263	2.3e-21	168 575	pir S45583 pisatin demethylase (a cytochrome p450)- fungus (Nectria haematococca) >gi 487426(L20976)-defense,pea phytoalexin detoxifying
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r8g06a1.f1	169	3.5e-11	15 458	sp P38364 PIDE_NECHA PISATIN DEMETHYLASE >pir S34286 pisatin demethylase -fungus (Nectria haematococca) >gi 31
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<D-AMINO ACID OXIDASE-oxidation of cephalosporin C>

cld08a1.f1	151	1.4e-07	297 470	sp Q99042 OXDA_TRIVR D-AMINO ACID OXIDASE (DAMOX) (DAO) (DAAO)>gnl PID e187982 (Z50019) D-amino acid oxidase [T
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3.detoxification (9)

<oxygen resistance>

Contig103_k5g12a1.f1	372	1.3e-33	164 499	gi 2979688 (AF035619) singlet oxygen resistance protein [Cercosporanicotianae]
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<SHO1 OSMOSENSOR>

Contig843_z3e05a1.f1	133	1.5e-07	377 475	sp P40073 SS81_Y SSU81 PROTEIN (SHO1 OSMOSENSOR) >pir S50621 SSU81protein - yeast (Saccharomyces cerevisiae) >gi 6033
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<catalase>

Contig404_f0d12a1.r1	1242	8.3e-126	8 703	sp P55305 CATA_E CATALASE A (SPORE-SPECIFIC CATALASE) >pir S68115catalase (EC 1.11.1.6) - Emericella nidulans >gi 109
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Contig1345_e4g06a1.fl	650	8.7e-125	470 916	sp P55305 CATA_E	CATALASE A (SPORE-SPECIFIC CATALASE)
clg07a1.r2	705	3.4e-108	66 482	>pir S68115catalase	(EC 1.11.1.6) - Emericella nidulans >gi 109
f0f12a1.r1	994	1.7e-99	10 612	sp P55305 CATA_EMENI	CATALASE A (SPORE-SPECIFIC CATALASE)
Contig1099_o8b12a1.fl	735	4.1e-72	78 533	>pir S68115catalase	(EC 1.11.1.6) - Emericella nidul
<super oxide dismutase>				sp P78619 CATB_EMENI	CATALASE B >gi 1737449 (U80672) catalase
Contig1507_g4h09a1.fl	609	8.9e-59	100 696	[Emericellanidulans]	
<epoxide hydrolase-+ water=glycol>				sp P78619 CATB_E	CATALASE B >gi 1737449 (U80672) catalase
g3dl2a1.r1	274	8.1e-23	74 580	[Emericellanidulans]	
				sp P00447 SODM_Y	SUPEROXIDE DISMUTASE PRECURSOR (MN)
				>pir DSBYNsuperoxide	dismutase (EC 1.15.1.1) (Mn) precursor - ye
				gi 1465805	(U64852) coded for by C. elegans cDNA cm17d4; Similar
					to epoxidehydrolase. [Caenorhabditis

4. salt tolerance (1)

<HALOTOLERANCE PROTEIN HAL2>

g9e05a1.fl	295	1.9e-25	127 531	gi 1109672	(U33283) 3'(2'),5-diphosphonucleoside 3'(2')
				phosphohydrolase[Oryza sativa]	>prf 2204308A=homolog of HALOTOLERANCE
					PROTEIN HAL2 of S. cerevisiae, homology is same
g9e05a1.r1	179	1.3e-12	221 448	sp P32179 HAL2_YEAST	HALOTOLERANCE PROTEIN HAL2 >pir S35318 HAL2 protein
					-yeast (Saccharomyces cerevisiae) >gi

5. starvation response (1)

<MAK16 PROTEIN-moves cytoplasmic proteins to vacuole-autophagocytosis>

c3f08a1.r1	249	5.9e-30	19 264	sp P40344 AUT1_YEAST	AUTOPHAGOCYTOSIS PROTEIN AUT1 >pir S45130
					hypotheticalprotein YNR007c - yeast (Saccharomy

B. Cell signalling, signal transduction

1. Kinases phosphatases and second messengers

a. PHOSPHATASES (15)

<PROTEIN PHOSPHATASE>

m5b10a1.r1	860	2.7e-85	37 648	gi 2429085	(U59418) protein phosphatase 2A B'alpha3 regulatory
				subunit [Musmusculus]	
o5a03a1.r1	672	1.1e-77	25 447	gi 2290382	(U89985) serine/threonine protein phosphatase PPT1
				[Neurospora crassa]	
i2c04a1.r1	460	6.2e-43	27 497	sp Q09172 P2C2_SCHPO	PROTEIN PHOSPHATASE 2C HOMOLOG 2 (PP2C-2)
				>pir S54297	protein phosphatase 2C - fission yea
*Contig287_g8d11a1.r1	394	1.3e-34	16 612	gi 2459997	(AF012898) protein phosphatase Ssd1 homolog [Candida
				albicans]	
z6e07a1.r1	249	3.5e-20	87 380	pir S54298	protein phosphatase 2C - fission yeast

Contig1195_v3g09a1.f1 237 2.3e-18 8 217 (Schizosaccharomyces pombe)>gi|609658 (L34882) prot
gnl|PID|d1010595 (D63916) protein phosphatase 2A 65kD regulatory
subunit (Asubunit) [Schizosaccharomyces pombe]
c3g08a1.r1 156 1.3e-15 319 543 sp|P40371|P2C1_SCHPO PROTEIN PHOSPHATASE 2C HOMOLOG 1 (PP2C-1)
>pir||A56058phosphoprotein phosphatase (EC 3.1.3
Contig941_o3b04a1.f1 173 2e-10 324 791 sp|P40152|YNV7_Y HYPOTHETICAL 37.2 KD PROTEIN IN ALG9-RAP1
INTERGENICREGION >pir||S50713 phosphoprotein phosphatase ho
Contig1224_c4a09a1.r1 143 1.6e-07 666 869 gnl|PID|d1016373 (D90827) Serine/Threonine protein phosphatase (EC
3.1.3.16).[Escherichia coli] >gi|1788143 (AE000278)
<ca dependent protein phosphatase>
olb08a1.r1 577 2.4e-55 199 597 gi|2645886 (AF034089) calcineurin subunit B [Neurospora crassa]
<PROTEIN-TYROSINE PHOSPHATASE>
Contig1586_o1f04a1.f1 305 5.8e-26 403 1011 sp|P34137|PTP1_D PROTEIN-TYROSINE PHOSPHATASE
1(PROTEIN-TYROSINE-PHOSPHATE PHOSPHOHYDROLASE 1) >gi|167862 (L07125)prot
<serine/threonine phosphatase>
Contig1494_m7c01a1.f1 718 2.9e-70 723 1238 sp|P78968|PPZ_SC SERINE/THREONINE PROTEIN PHOSPHATASE PP-Z
>gi|1763281(U73689) PPZ protein phosphatase [Schizosaccharo
r2h10a1.f1 622 4.5e-60 160 633 sp|P36614|PPE1_SCHPO SERINE/THREONINE PROTEIN PHOSPHATASE PPE1
(PHOSPHATASEESP1) >pir||A47727 cell shape contro
<PRL1/PRL2-LIKE PROTEIN>
Contig403_f0e04a1.r1 969 7.7e-97 24 728 sp|O13615|PRL1_S PRL1/PRL2-LIKE PROTEIN >gnl|PID|d1022249 (AB004535)
PRL1[Schizosaccharomyces pombe]
Contig260_i8d10a1.r1 413 6.1e-38 184 528 sp|O13615|PRL1_S PRL1/PRL2-LIKE PROTEIN >gnl|PID|d1022249 (AB004535)
PRL1[Schizosaccharomyces pombe]

b.Kinases (35)
<protein kinase>
Contig1226_g7h05a1.r1 668 6.1e-65 13 711 sp|P32490|MKK1_Y PROTEIN KINASE MKK1/SSP32 >pir||A48069 protein
kinaseMKK1 (EC 2.7.1.-) - yeast (Saccharomyces cerevis
Contig701_w6h11a1.f1 278 6.1e-23 416 739 gnl|PID|e1293569 (AL023634) protein kinase dsk1 [Schizosaccharomyces
pombe]
Contig1159_g3d04a1.r1 271 5.5e-20 54 635 gi|532798 (U13398) protein kinase [Saccharomyces cerevisiae]
<protein kinase C>
Contig281_g9b09a1.r1 981 3.8e-98 9 623 sp|Q00078|KPC1_A PROTEIN KINASE C-LIKE >pir||S61917 protein kinase
Chomolog PKCA - Aspergillus niger >gi|507900 (U1054
Contig1630_c4c06a1.f1 702 1.4e-68 81 920 sp|Q10138|YAS2_S HYPOTHETICAL 51.5 KD PROTEIN C3H8.02 IN CHROMOSOME
I>gi|1177660 (Z69086) unknown [Schizosaccharomyces
Contig1176_e7a01a1.r1 510 4.1e-47 341 658 sp|Q00078|KPC1_A PROTEIN KINASE C-LIKE >pir||S61917 protein kinase
Chomolog PKCA - Aspergillus niger >gi|507900 (U1054
<MAP kinase>
k8b03a1.f1 152 4.6e-10 55 162 gi|496307 (L26523) stem loop mutation suppressor [Saccharomyces
cerevisiae]-MAP kinase regulator
<MAP kinase HOG1>

Contig1648_g4a02a1.f1 821 3.7e-81 420 1109 sp|P32485|HOG1_Y MITOGEN-ACTIVATED PROTEIN KINASE HOG1 (MAP KINASE HOG1)(OSMOSENSING PROTEIN HOG1) >pir||S64950 protei

<CALMODULIN-DEPENDENT PROTEIN KINASE>

i3h06a1.r1 815 1.5e-80 23 493 sp|Q00771|KCC1_EMENI CALCIUM/CALMODULIN-DEPENDENT PROTEIN KINASE (CMPK)>pir||JN0323 Ca2+/calmodulin-dependent p

i7a07a1.r1 332 2.2e-29 158 367 sp|Q00771|KCC1_EMENI CALCIUM/CALMODULIN-DEPENDENT PROTEIN KINASE (CMPK)>pir||JN0323 Ca2+/calmodulin-dependent p

i3h06a1.f1 188 3.4e-12 342 497 sp|Q00771|KCC1_EMENI CALCIUM/CALMODULIN-DEPENDENT PROTEIN KINASE (CMPK)>pir||JN0323 Ca2+/calmodulin-dependent p

Contig29_d1f08a1.f1 145 9.6e-09 274 414 gi|2654181 (AF034963) calmodulin-dependent protein kinase; CgCMK [Glomerellacingulata]

<CAMP-DEPENDENT PROTEIN KINASE>

n8d03a1.r1 700 2.5e-68 32 502 gi|516040 (U12335) cAMP-dependent protein kinase catalytic subunit[Magnaporthe grisea]

Contig1318_i0a06a1.f1 683 1.5e-66 445 849 gi|3170248 (AF043231) cAMP-dependent protein kinase regulatory subunit[Emericella nidulans]

f2h07a1.r1 383 8.6e-34 15 605 sp|P11792|SCH9_YEAST CAMP-DEPENDENT PROTEIN KINASE SCH9 >pir||S48986 probableprotein kinase SCH9 (EC 2.7.1.-) -

<SERINE/THREONINE-PROTEIN KINASE, casein kinase acts on acidic proteins>

Contig1695_d2d12a1.f1 1127 1.4e-113 727 1581 sp|P25333|KCR8_Y PROBABLE SERINE/THREONINE-PROTEIN KINASE YCR8W>pir||OKBY8W probable protein kinase YCR008w (EC 2.7.1.

Contig1717_c5c12a1.r1 757 2.2e-74 40 987 sp|P38623|RCK2_Y SERINE/THREONINE-PROTEIN KINASE RCK2 (CAM KINASE-LIKEPROTEIN KINASE CLK1) >gi|733594 (U23464) Cam kin

Contig1440_r2c01a1.f1 488 5.7e-74 712 1227 sp|Q01389|BCK1_Y SERINE/THREONINE PROTEIN KINASE BCK1/SLK1/SSP31>pir||S20117 protein kinase BCK1 (EC 2.7.1.-) - yeast(

k5d01a1.r1 602 5.9e-58 16 510 gi|2911462 (AF046923) serine/threonine protein kinase [Colletotrichumtrifolii]

Contig1673_g3g11a1.r1 554 7.1e-53 290 1252 sp|O14019|KDPG_S PROBABLE SERINE/THREONINE-PROTEIN KINASE C29A4.16>gnl|PID|e325359 (Z97210) protein kinase [Schizosacc

Contig881_r4f03a1.f1 502 2.4e-47 28 600 sp|Q07538|PRP4_S SERINE/THREONINE-PROTEIN KINASE PRP4 >gi|1857026 (L10739)serine/threonine kinase [Schizosaccharomyces

m6f09a1.r1 454 4.1e-41 42 413 sp|Q08217|KOE5_YEAST PROBABLE SERINE/THREONINE-PROTEIN KINASE YOL045W>pir||S66730 hypothetical protein YOL045w

i8h08a1.r1 403 1.2e-36 149 490 sp|P33886|WIS1_SCHPO SERINE/THREONINE PROTEIN KINASE WIS1 >pir||S18648 protein kinase wis1 (EC2.7.1.-) - fission yeast (Schizosa

v1h01a1.r1 313 4.9e-26 207 587 sp|P32361|IRE1_YEAST SERINE/THREONINE-PROTEIN KINASE IRE1 PRECURSOR>pir||A47541 protein kinase IRE1-required for iniositol uptake (EC 2.7.1.-

u4h01a1.f1 250 2.7e-19 108 506 sp|P32361|IRE1_YEAST SERINE/THREONINE-PROTEIN KINASE IRE1 PRECURSOR>pir||A47541 protein kinase IRE1 (EC 2.7.1.-

n1a03a1.r1 110 1.7e-05 102 269 gnl|PID|d1033019 (AB014506) non receptor serine/threonine kinase [Dugesiajaponica]

<serine/threonine-specific protein kinase KIN2>

j5c10a1.r1 151 1.2e-17 10 168 sp|P13186|KIN2_YEAST PROTEIN KINASE KIN2 >gi|171789 (M69018) protein kinase
2[Saccharomyces cerevisiae]=serine/threonine-specific protein kinase KIN2,EC
2.7.1

<casein kinase-ser/thr protein kinase, acts on acidic proteins>

m7a09a1.f1 233 7.4e-19 272 499 sp|P40232|KC2B_SCHPO CASEIN KINASE II BETA CHAIN (CK II) >gi|452290
(X74274)casein kinase II beta subunit [Schi
z7e04a1.f1 137 2.9e-07 496 603 sp|Q08466|KC22_ARATH CASEIN KINASE II, ALPHA CHAIN 2 (CK II)
>pir||S31099casein kinase II (EC 2.7.1.-) alpha-ty

<histidine kinase>

Contig528_c8b06a1.r1 397 1.9e-35 66 539 gi|1679757 (U77605) two-component histidine kinase CHK-1
[Glomerellacingulata] >gi|1679760 (U77606) two-componen
c5e08a1.r1 241 1.1e-30 374 709 sp|P39928|SLN1_YEAST OSOMOLARITY TWO-COMPONENT SYSTEM PROTEIN
SLN1>pir||S48387 SLN1 protein - yeast (Saccharomy
j7e12a1.r1 275 7.3e-22 2 241 gi|1262210 (U50264) Nik-1 histidine kinase [Neurospora crassa]
g2e07a1.r1 220 3.5e-16 104 508 gn1|PID|d1018731 (D90910) sensory transduction histidine kinase
[Synechocystis sp.]
Contig1187_g2a10a1.f1 146 1.2e-06 245 601 gi|2313493 (AE000555) histidine kinase (cheA) [Helicobacter
pylori]
g2c06a1.f1 149 1.6e-06 209 523 gi|3243089 (U69886) histidine kinase [Candida albicans]

<nitrogen permease reactivator-on for nitrogenous transport systems/s-t kinase>

Contig555_o5c08a1.r1 336 9.1e-29 4 420 sp|P22211|NPR1_Y NITROGEN PERMEASE REACTIVATOR PROTEIN
>pir||S63138probable protein kinase NPR1 (EC 2.7.1.-) - yeast (

c.cAMP (1)

<adenyl cyclase>

<ADENYLYL CYCLASE-ASSOCIATED PROTEIN-CAP protein, binds cAMP to allow activation>

j9f01a1.f1 258 8.1e-21 179 706 sp|P17555|CAP_YEAST ADENYLYL CYCLASE-ASSOCIATED PROTEIN (CAP)
>pir||A34896adenylate cyclase-associated protein
j9f01a1.r1 218 2e-16 125 481 sp|P36621|CAP_SCHPO ADENYLYL CYCLASE-ASSOCIATED PROTEIN (CAP)
>pir||A60047adenylate cyclase-associated protein

2. G protein (24)

<GTP-binding protein>

Contig73_m3f12a1.r1 725 6.5e-80 93 602 sp|P52886|SARA_A GTP-BINDING PROTEIN SARA >gi|1061034 (Z67742)
sarA[Aspergillus niger]
nih10a1.r1 767 1.8e-75 20 604 sp|P53742|YN8U_YEAST HYPOTHETICAL GTP-BINDING PROTEIN IN POP2-HOL1
INTERGENICREGION >pir||S63384 hypothetical p
Contig833_w4d03a1.r1 764 3.5e-75 7 489 sp|P32835|GSP1_Y GTP-BINDING NUCLEAR PROTEIN GSP1/CNR1
>pir||S35504GTP-binding protein GSP1 - yeast (Saccharomyces cer
Contig1665_13g11a1.f1 736 3.8e-72 201 800 sp|P36586|YPT5_S YPT1-RELATED PROTEIN 5 >pir||S34729 GTP-binding

o5f05a1.r1 699 3.1e-68 7 489 proteinypt5 - fission yeast (Schizosaccharomyces pombe
 sp|P22129|R11B_DISOM RAS-RELATED PROTEIN RAB-11B (ORA3)
 >pir||C38625GTP-binding protein ora3 - electric ray
 *Contig1458_g6b11a1.f1 546 5.1e-52 611 1159 sp|P01122|RHO_AP RAS-LIKE GTP-BINDING PROTEIN RHO >pir||TVGAAC
 transformngprotein rho - California sea hare >gi|15580
 f0h02a1.r1 473 2.7e-44 180 611 pir||I38176 ragA - human >pir||I84474 Raga (ras-related,
 alternatively splicedGTPase A) - rat >gi|1063
 h1a06a1.f1 433 4.9e-40 9 359 gnl|PID|e353254 (Z99753) rho protein GTP-binding [Schizosaccharomyces
 pombe]
 *Contig383_f5a04a1.f1 245 3.3e-39 502 843 gi|173183 (M33315) GTP-binding protein (VPS1) [Saccharomyces
 cerevisiae]
 Contig1227_g7a07a1.f1 398 2.4e-36 295 576 sp|P36017|YP51_Y GTP-BINDING PROTEIN YPT51/VPS21 >pir||S43399
 GTP-bindingprotein VPS21 - yeast (Saccharomyces cerevisi
 c3f01a1.r1 355 8.4e-32 19 342 sp|P32235|GTP1_SCHPO GTP-BINDING PROTEIN 1 >pir||JT0741 GTP-binding protein
 1- fission yeast (Schizosaccharomyc
 y6a01a1.r1 170 2e-25 512 649 pir||S71233 GTP-binding protein 2 - Arabidopsis thaliana
 >pir||S71585 Rab2homolog GTP-binding protein
 j9c11a1.f1 244 2.8e-19 226 615 sp|P40010|YEJ6_YEAST HYPOTHETICAL GTP-BINDING PROTEIN IN PMI40-PAC2
 INTERGENICREGION >pir||S50464 hypothetical
 *Contig199_i3a04a1.f1 198 1.3e-14 288 593 gnl|PID|e1294581 (AJ006412) putative GTP-binding protein [Homo sapiens]
 y6a01a1.f1 189 3.2e-14 128 442 pir||E42148 GTP-binding protein rab14 - rat
 m8d10a1.r1 148 2e-08 417 785 pir||A53778 GTP-binding protein A-ras - Emericella nidulans
 >gi|458024(U03023) Ras-like protein [Asper

 <GUANINE NUCLEOTIDE-BINDING PROTEIN>
 Contig163_j0e04a1.r1 253 5.5e-21 126 287 sp|Q01369|GBLP_N GUANINE NUCLEOTIDE-BINDING PROTEIN BETA
 SUBUNIT-LIKEPROTEIN (CROSS-PATHWAY CONTROL WD-REPEAT PROTEIN

 <RHO1 GDP-GTP EXCHANGE PROTEIN:GUANINE-NUCLEOTIDE
 g4e12a1.r1 278 3.7e-22 32 511 sp|P51862|ROM2_YEAST RHO1 GDP-GTP EXCHANGE PROTEIN 2 >pir||S51389 ROM2
 protein- yeast (Saccharomyces cerevisiae
 Contig1206_g4e12a1.f1 207 1.6e-12 624 836 sp|P51862|ROM2_Y RHO1 GDP-GTP EXCHANGE PROTEIN 2 >pir||S51389 ROM2
 protein- yeast (Saccharomyces cerevisiae) >gi|60940

 <rho-gdp dissociation inhibitor-prevents cycling of GDP with GTP of rho protein family>
 b0c03a1.r1 256 2.4e-21 58 378 gnl|PID|e334111 (Z98533) rho-gdp dissociation inhibitor
 [Schizosaccharomycespombe]
 b0c03a1.f1 239 1.7e-19 224 478 gnl|PID|e334111 (Z98533) rho-gdp dissociation inhibitor regulates rho
 protein needed for actin reorganization[Schizosaccharomycespombe]

 <RHO2 PROTEIN>
 Contig1386_e9f01a1.r1 723 8.5e-71 405 971 sp|Q10133|RHO2_S RHO2 PROTEIN >pir||JC4045 Rho2 protein - fission
 yeast(Schizosaccharomyces pombe) >gnl|PID|d1007956 (

 <GTPASE-ACTIVATING PROTEIN-neg regulator of Ras1, play antagonistic role with rho-gdp dissociation inhibitor>
 o4e01a1.r1 379 1.9e-33 22 480 sp|P33277|GAP1_SCHPO GTPASE-ACTIVATING PROTEIN >pir||A40258
 RASGTPase-activating protein sar1 - fission yeast (

 <RAS-2 PROTEIN-GTP-BINDING>

Contig1415_o9d09a1.r1 829 5.1e-82 129 662 sp|Q09914|RHO1_S RHO1 PROTEIN >pir||JC4044 Rho1 protein - fission yeast (Schizosaccharomyces pombe) >pir||S62576 hypoth

Contig1216_g8h09a1.r1 407 2.7e-37 8 571 sp|Q01387|RAS2_N RAS-2 PROTEIN >gnl|PID|d1004223 (D16137) NC-ras-2 protein [Neurospora crassa]

C. Transmembrane transport

1. secretion (6)

<secretion>

c7e05a1.r1 237 4e-18 32 595 gnl|PID|e1316739 (AL031324) subunit of the final step of the secretory pathway [Schizosaccharomyces pombe]

c7e05a1.f1 200 7.4e-13 89 616 gnl|PID|e1316739 (AL031324) subunit of the final step of the secretory pathway [Schizosaccharomyces pombe]

d2f12a1.r1 136 1.3e-05 486 659 sp|Q06245|SC10_YEAST EXOCYST COMPLEX COMPONENT SEC10 >pir||S68482 probable membrane protein YLR166c - yeast (Sac

<KEXIN-proteinase secretion>

m7g09a1.r1 341 8.2e-39 99 578 sp|O13359|KEX2_CANAL KEXIN PRECURSOR (KEX2 PROTEASE) >gi|2511732 (AF022372) proteinase secretion [Candida albicans]

<VESICULAR-FUSION PROTEIN SEC17>

Contig1446_g7e01a1.r1 349 1.4e-45 82 681 gi|1711132 (U79186) sec17-like protein [Coprinus cinereus]

<SECRETORY COMPONENT PROTEIN SHR3>

Contig165_j0c07a1.f1 171 2.5e-12 349 675 sp|Q02774|SHR3_Y SECRETORY COMPONENT PROTEIN SHR3 >gi|172572 (L01264) secretory component [Saccharomyces cerevisiae]

2. exoenzymes (5)

<exoenzyme>

e0c10a1.r1 720 1.5e-70 26 610 gi|2340046 (L48074) secreted dipeptidyl peptidase [Aspergillus fumigatus]

<dipeptidyl peptidase-exoenzyme>

i8f01a1.r1 691 2.1e-67 20 595 gi|2340046 (L48074) secreted dipeptidyl peptidase [Aspergillus fumigatus]

Contig679_u4e06a1.r1 480 1.6e-44 9 350 gi|2351700 (U87950) dipeptidyl-peptidase IV [Aspergillus fumigatus]

*Contig729_w7f07a1.f1 383 9e-34 24 584 gnl|PID|d1025528 (D89340) dipeptidyl peptidase [Rattus norvegicus]

Contig1312_c9a01a1.f1 121 1e-05 134 298 gnl|PID|e1254390 (AJ002369) prolyl dipeptidyl peptidase [Aspergillus oryzae]

3. transport (85)

a. sugar transport

<sugar transport>

o0f08a1.r1 435 2.7e-40 21 416 gi|2306977 (AF010145) hexose transporter [Aspergillus parasiticus]

s3g03a1.f1	360	5.1e-32	55 561	sp P39932 STL1_YEAST	SUGAR TRANSPORTER STL1 >pir s69591 sugar transportprotein STP1 - yeast (Saccharomyces cer
*Contig1230_k0h11a1.f1	329	1.4e-28	15 1067	gi 409547	(L07492) sugar transport protein [Saccharomyces cerevisiae]
d5a05a1.r1	310	1.8e-26	68 646	gi 409547	(L07492) sugar transport protein [Saccharomyces cerevisiae]
c6g07a1.r1	289	3.3e-24	11 640	gi 409547	(L07492) sugar transport protein [Saccharomyces cerevisiae]
o0f08a1.f1	263	2.3e-21	184 447	gi 2306977	(AF010145) hexose transporter [Aspergillus parasiticus]
<GLUCOSE TRANSPORTER>					
*Contig1735_d5f08a1.r1	719	2.4e-70	353 1351	sp Q12300 RGT2_Y	HIGH-AFFINITY GLUCOSE TRANSPORTER RGT2 >pir s67684probable membrane protein YDL138w - yeast (Sacchar
m7a06a1.f1	197	4e-14	206 490	sp P49374 HGT1_KLULA	HIGH-AFFINITY GLUCOSE TRANSPORTER >gi 726336 (U22525)high affinity glucose transporter [Kl
Contig57_o0b10a1.r1	137	1.2e-07	253 414	sp P32465 HXT1_Y	LOW-AFFINITY GLUCOSE TRANSPORTER HXT1 >pir s38798 hexosetransport protein HXT1 - yeast (Saccharomyce
Contig924_o0b10a1.f1	121	9e-06	232 408	sp Q12300 RGT2_Y	HIGH-AFFINITY GLUCOSE TRANSPORTER RGT2 >pir s67684probable membrane protein YDL138w - yeast (Sacchar
<GALACTOSE TRANSPORTER>					
r7d01a1.r1	245	1.5e-19	25 717	dbj AB007638_9	(AB007638) metabolite transport protein [Bacillus subtilis]>gnl PID e1182602 (Z99107) simi
<inositol transport>					
Contig1074_f0a04a1.r1	329	7e-29	9 926	gnl PID e353127	(Z99708) myo-inositol transport protein homolog [Arabidopsisthaliana]
o5h09a1.r1	208	8.6e-16	27 506	gnl PID d1014610	(D89252) similar to Saccharomyces cerevisiae myo-inositoltransporter 1
b. multidrug resistance					
<multidrug resistance>					
r1g01a1.f1	763	4.8e-74	3 581	gi 2673951	(U62933) multidrug resistance protein 1 [Aspergillus fumigatus]>gi 2673953 (U62934) multid
j9b01a1.r1	603	9.5e-57	57 479	gnl PID d1032208	(AB010442) PMR1 [Penicillium digitatum]
h8h04a1.r1	599	2.6e-56	39 467	gnl PID d1032208	(AB010442) PMR1 MULTIDRUG RESISTANCE[Penicillium digitatum]
r2c11a1.f1	389	4.7e-51	49 381	gi 2673955	(U62935) multidrug resistance protein 2 [Aspergillus fumigatus]>gi 2673957 (U62936) multid
q0g01a1.f1	539	6e-50	2 454	gnl PID d1032208	(AB010442) PMR1 [Penicillium digitatum]
o8f05a1.f1	371	4.7e-32	157 666	gi 2668553	(U62929) multidrug resistance protein 1 [Filobasidiella neoformans]>gi 2668555 (U62930) mu
o6b10a1.r1	232	6.1e-18	37 528	gnl PID e1313752	(Y14703) multidrug resistance protein [Candida albicans]
<multidrug transporter>					

h0d01a1.f1 605 5.5e-57 44 673 gn1|PID|e219956 (Z68904) ATP-binding cassette multidrug transporter[*Emericella nidulans*]

y4h01a1.f1 229 1.8e-15 332 541 gn1|PID|e219956 (Z68904) ATP-binding cassette multidrug transporter[*Emericella nidulans*]

<oxytetracycline exporter>
l0e10a1.r1 122 4.7e-06 309 476 gi|3108177 (AF061335) oxytetracycline exporter [*Streptomyces rimosus*]

<cycloheximide resistance protein>
*Contig1115_r2h04a1.f1 174 1.3e-11 62 571 pir||JC1173 cycloheximide resistance protein - yeast (*Candida maltosa*)

alf03f2.f1 161 5.2e-09 33 773 pir||JC1173 cycloheximide resistance protein - yeast (*Candida maltosa*)

<HOL1 PROTEIN-DRUG RESISTANCE TRANSLOCASE FAMILY, MAJOR FACILITATOR FAMILY>
Contig1295_m8d08a1.f1 251 6.8e-20 166 648 gi|825501 (L42348) HOL1 [*Saccharomyces cerevisiae*]

c. nuclear membrane transport
<nuclear pore membrane protein>
Contig1041_d2b04a1.r1 415 8.7e-37 33 884 sp|P39685|P152_Y NUCLEAR ENVELOPE PORE MEMBRANE PROTEIN POM152 (P150)>pir||A53824 nuclear pore membrane protein POM152

<NUCLEAR TRANSPORT FACTOR 2>
Contig472_o0c11a1.f1 394 6e-36 292 660 sp|P33331|NTF2_Y NUCLEAR TRANSPORT FACTOR 2 (NTF-2) (NUCLEAR TRANSPORTFACTOR P10) >pir||S50467 hypothetical protein YE

d.cation transport-ATPase, or major facilitator superfamily
<cation transport>
z5c08a1.r1 539 7e-50 12 587 sp|P32660|ATC5_YEAST PROBABLE CALCIUM-TRANSPORTING ATPASE 5 >pir||S50669hypothetical protein YER166w - yeast (S

e6h10a1.r1 324 3.3e-27 11 472 sp|P54678|ATC1_DICDI CATION-TRANSPORTING ATPASE PAT1 >pir||S57726 PAT1 protein- slime mold (*Dictyostelium disco*

d3a10a1.r1 175 2.5e-09 27 278 sp|P40527|ATC7_YEAST PROBABLE CALCIUM-TRANSPORTING ATPASE 7 >pir||S48431probable membrane protein YIL048w - yea

<CATION-TRANSPORTING ATPASE>
k0c11a1.r1 440 1.3e-39 37 417 sp|P54678|ATC1_DICDI CATION-TRANSPORTING ATPASE PAT1 >pir||S57726 PAT1 protein- slime mold (*Dictyostelium disco*

<CALCIUM-TRANSPORTING ATPASE>
Contig642_c0a04a1.f1 238 5.4e-18 95 349 sp|P39986|ATC6_Y PROBABLE CALCIUM-TRANSPORTING ATPASE 6 >pir||S50428hypothetical protein YEL031w - yeast (*Saccharomyce*

m8b04a1.f1 103 4.1e-05 7 195 pir||A35731 Ca²⁺-transporting ATPase (EC 3.6.1.38), cardiac and slow skeletal muscle - rabbit (fragment

<sodium transport>
Contig1736_i8e08a1.f1 1073 7e-108 44 952 gi|1438947 (U61840) sodium transport ATPase FST [*Fusarium solani* f. sp. pisi]

<sulfate transporter>

hlc01a1.f1	300	6.1e-25	15 590	sp P53394 SULX_YEAST PUTATIVE SULFATE TRANSPORTER YPR003C >pir S52816probable membrane protein YPR003c - yeast
<cobalt transporter>				
Contig1391_n0d02a1.f1	247	7.8e-20	208 720	sp P32798 COT1_Y COBALT UPTAKE PROTEIN COT1 >pir S58327 COT1 protein -yeast (Saccharomyces cerevisiae) >gi 940847 (X9
<COPPER TRANSPORT>				
Contig430_e0f09a1.r1	195	7.6e-15	144 341	sp Q06686 CTR3_Y COPPER TRANSPORT PROTEIN CTR3 (COPPER TRANSPORTER 3)>pir S59377 probable membrane protein YLR411w -
<zinc cadmium resistance>				
Contig221_i0b06a1.r1	460	6.4e-43	86 589	sp P20107 ZRC1_Y ZINC/CADMIUM RESISTANCE PROTEIN >pir S56057 heavy metal ion resistance protein ZRC1 - yeast (Saccharo
o5d12a1.r1	232	2e-18	69 419	gnl PID e334142 (Z98559) probable zinc cadmium resistance protein[Schizosaccharomyces pombe]
<manganese resistance>				
Contig1525_glg10a1.r1	370	2.3e-33	9 359	gnl PID e1169877 (AJ001272) manganese resistance 1 protein [Saccharomycescerevisiae]
m6d08a1.f1	287	2.4e-23	193 585	sp P35724 MNR2_YEAST MANGANESE RESISTANCE PROTEIN >pir S37886 hypotheticalprotein YKL064w - yeast (Saccharomyc
Contig1478_a0c02a1.f1	178	2.4e-12	315 482	gnl PID e1169877 (AJ001272) manganese resistance 1 protein [Saccharomycescerevisiae]
e. Anion transport				
<arsenite translocating ATPase-anion transport, resistance to arsenite, antimonite, arsenate>				
c0h09a1.f1	441	6.7e-41	103 594	gi 2905657 (AF047469) arsenite translocating ATPase [Homo sapiens]
c0h09a1.r1	133	1.3e-07	325 486	gi 1616741 (U60276) HASNA-I [Homo sapiens]
<phosphate transporter>				
*Contig1457_j9a02a1.f1	571	1.1e-54	124 693	gnl PID d1032546 (AB016066) mitochondrial phosphate transporter [Arabidopsisthaliana]
m2g02a1.f1	545	6.6e-52	61 636	gnl PID d1032546 (AB016066) mitochondrial phosphate transporter [Arabidopsisthaliana]
Contig1511_d1f10a1.f1	507	6.5e-48	278 880	pir S60949 phosphate transport protein, mitochondrial - yeast (Saccharomycescerevisiae) >gi 1050774 (X92441) YOR
Contig91_l3g04a1.r1	319	5.7e-28	300 596	gnl PID d1032543 (AB016063) mitochondrial phosphate transporter [Glycine max]
<tartrate transport>				
*Contig258_m8b10a1.f1	316	2.3e-27	349 1344	gi 805291 (U25634) putative tartrate transporter; inducible by tartrate;Method: conceptual translation supplied
mlc01a1.r1	262	1.4e-21	143 667	gi 805291 (U25634) putative tartrate transporter; inducible by tartrate;Method: conceptual translati
<CHOLINE TRANSPORT>				
r4a12a1.r1	313	1e-26	76 798	sp P19807 HNM1_YEAST CHOLINE TRANSPORT PROTEIN >pir S11175 choline transportprotein - yeast (Saccharomyces cer

r4a12a1.f1	125	0.00019	425 763	sp P19807 HNM1_YEAST CHOLINE TRANSPORT PROTEIN >pir S11175 choline transportprotein - yeast (Saccharomyces cer
<allantoate transport>				
Contig880_r5f08a1.f1	355	1.6e-31	135 929	sp P15365 DAL5_Y ALLANTOATE PERMEASE >pir A28671 allantoate transportprotein - yeast (Saccharomyces cerevisiae) >gi 2
g7c12a1.r1	325	4.1e-28	18 767	sp P15365 DAL5_YEAST ALLANTOATE PERMEASE >pir A28671 allantoate transportprotein - yeast (Saccharomyces cerevi
d4e02a1.r1	180	1.3e-10	13 639	sp P15365 DAL5_YEAST ALLANTOATE PERMEASE >pir A28671 allantoate transportprotein - yeast (Saccharomyces cerevi
f. Protein, amino acid transport				
<PROTEIN TRANSPORT PROTEIN>				
z4b10a1.r1	685	9.1e-67	11 649	sp P15303 SC23_YEAST PROTEIN TRANSPORT PROTEIN SEC23 >pir BVBY23 proteintransport protein SEC23 - yeast (Sacch
a0d10a1.f1	402	9.1e-37	133 489	sp Q04491 SC13_YEAST PROTEIN TRANSPORT PROTEIN SEC13 >pir A45442 SEC13protein - yeast (Saccharomyces cerevisia
Contig1341_e4b02a1.f1	127	0.0002	289 735	sp P40357 SEC9_Y PROTEIN TRANSPORT PROTEIN SEC9 >pir A55100 SEC9 protein- yeast (Saccharomyces cerevisiae) >gi 508620
<EXPORT PROTEIN NCE2-protein transport, w/o signal sequence>				
Contig1262_c2c07a1.f1	217	3.5e-17	36 473	sp Q12207 NCE2_Y NON-CLASSICAL EXPORT PROTEIN NCE2 >pir S69036 NCE2protein - yeast (Saccharomyces cerevisiae) >gi 106
<PEPTIDE TRANSPORTER>				
Contig973_m6b08a1.f1	337	3.2e-29	130 654	sp P46031 PTR2_A PEPTIDE TRANSPORTER PTR2-A >gi 575427 (U01171) similar toS. cerevisiae PTR2 gene, GenBank Accession N
m0h08a1.r1	200	2.5e-14	14 466	sp P46030 PTR2_CANAL PEPTIDE TRANSPORTER PTR2 >gi 806693 (U09781) peptidetransporter [Candida albicans]
<AMINO ACID TRANSPORTER>				
*Contig1165_e0f01a1.r1	307	3.2e-26	18 587	sp P51906 EAT3_M EXCITATORY AMINO ACID TRANSPORTER 3 (SODIUM-DEPENDENTGLUTAMATE/ASPARTATE TRANSPORTER 3) (EXCITATORY A
<AMINO-ACID PERMEASE>				
l5b08a1.f1	276	9.8e-23	130 420	sp Q09887 YAHB_SCHPO HYPOTHETICAL AMINO-ACID PERMEASE C8A4.11 >pir S62527hypothetical protein SPAC8A4.11 - fis
w7b01a1.f1	185	8.8e-13	198 497	sp P38971 ALP1_YEAST BASIC AMINO-ACID PERMEASE >pir S60912 probable transportprotein ALP1 - yeast (Saccharomyc
p0h04a1.r1	154	2.1e-09	132 461	sp P36029 YKR4_YEAST HYPOTHETICAL AMINO-ACID PERMEASE IN STE3-GIN10 INTERGENICREGION >pir S38004 probable tran
<ARGININE PERMEASE>				
w7b01a1.r1	438	1.4e-40	6 530	sp P04817 CAN1_YEAST ARGININE PERMEASE >pir QRBYPR arginine transport protein- yeast (Saccharomyces cerevisiae
<importin beta-4 subunit, nuclear protein import>				
Contig1370_c4a02a1.f1	432	7.2e-39	47 760	gnl PID e1295815 (AL023780) putative importin beta-4 subunit[Schizosaccharomyces pombe]

g.mitochondrial transport

<MITOCHONDRIAL PROTEIN IMPORT PROTEIN 2>

Contig1606_e9d06a1.f1 588 1.8e-56 252 698 sp|P39515|IM17_Y MITOCHONDRIAL IMPORT INNER MEMBRANE TRANSLOCASE
SUBUNITTIM17 (MITOCHONDRIAL PROTEIN IMPORT PROTEIN 2)

<MITOCHONDRIAL IMPORT RECEPTOR SUBUNIT TOM22-translocation of cytosolic proteins into mitochondria>

f2f07a1.r1 185 1e-13 200 535 sp|Q07335|OM22_NEUCR MITOCHONDRIAL IMPORT RECEPTOR SUBUNIT
TOM22(MITOCHONDRIAL 22 KD OUTER MEMBRANE PROTEIN) =N. crassa

<mitochondrial transport protein amc-1>

Contig1668_c4b09a1.f1 418 1.9e-38 171 521 gi|1621438 (U71603) mitochondrial transport protein amc-1
[Emericellanidulans]

g5b11a1.r1 272 5.3e-23 363 542 gi|1621438 (U71603) mitochondrial transport protein amc-1
[Emericellanidulans]

<MITOCHONDRIAL 2-OXOGLUTARATE/MALATE CARRIER>

r5e09a1.r1 353 1.5e-31 86 703 pir||S65040 2-oxoglutarate/malate translocator (clone OMT134),
mitochondrialmembrane - proso -millet

<benzodiazepine receptor-TRANSPORT OF PORPHYRINS AND HEME, mitochondria>

v3c06a1.r1 173 1.7e-12 63 482 pir||A53405 peripheral-type benzodiazepine receptor 1
isoquinoline-bindingprotein - mouse =MITOCHONDRIAL

<ADP,ATP CARRIER PROTEIN>

Contig1208_s9c05a1.f1 781 5e-77 72 626 sp|P02723|ADT_NE ADP,ATP CARRIER PROTEIN (ADP/ATP TRANSLOCASE)
(ADENINENUCLEOTIDE TRANSLOCATOR) (ANT) >pir||XWNC ADP,A

Contig1293_i8a04a1.r1 580 1.2e-55 258 650 sp|Q09188|ADT_SC ADP,ATP CARRIER PROTEIN (ADP/ATP TRANSLOCASE)
(ADENINENUCLEOTIDE TRANSLOCATOR) (ANT) >gnl|PID|e186767

h.ABC transporter family

<ABC transporter>

Contig1663_d5h04a1.f1 1385 6.3e-141 311 2029 sp|P36619|PMD1_S LEPTOMYCIN B RESISTANCE PROTEIN PMD1
>pir||S20548leptomycin B resistance protein - fission yeast(Schi

Contig1432_c3a08a1.r1 765 5.4e-74 2 883 gi|2625138 (AF032443) ABC1 transporter; ABC-type ATPase
[Magnaporthe grisea]

Contig1558_c9a02a1.f1 709 2.6e-69 363 893 sp|P40024|YEM6_Y PROBABLE ATP-DEPENDENT TRANSPORTER YER036C
>pir||S50539hypothetical protein YER036c - yeast (Saccharo

c9a02a1.r1 705 6.8e-69 13 540 sp|P40024|YEM6_YEAST PROBABLE ATP-DEPENDENT TRANSPORTER YER036C
>pir||S50539hypothetical protein YER036c - yeas

*Contig1477_s3f01a1.f1 527 1.4e-48 244 792 gi|2625138 (AF032443) ABC1 transporter; ABC-type ATPase
[Magnaporthe grisea]

f0d04a1.r1 500 1.1e-45 10 600 gi|2625138 (AF032443) ABC1 transporter; ABC-type ATPase
[Magnaporthe grisea]

Contig1000_c9e04a1.f1 372 3.4e-32 6 440 gnl|PID|e1316128 (AL031307) leptomycin B resistance protein, ABC
transporter[Schizosaccharomyces pombe]

Contig428_e4a06a1.r1 309 6.4e-26 57 707 gnl|PID|e1285355 (AL022299) ABC transporter [Schizosaccharomyces pombe]

*Contig1647_c3a02a1.f1 239 4.8e-16 900 1337 gi|2622773 (AE000923) ABC transporter [Methanobacterium
thermoautotrophicum]

d3g08a1.f1 169 1.1e-10 155 412 sp|P25371|ADP1_YEAST PROBABLE ATP-DEPENDENT PERMEASE PRECURSOR
>pir||S19421ATP-dependent permease ADP1 precurso

<ATP-DEPENDENT PERMEASE>
Contig464_d3g08a1.rl 412 1.1e-36 235 729 sp|P25371|ADP1_Y PROBABLE ATP-DEPENDENT PERMEASE PRECURSOR
>pir||S19421ATP-dependent permease ADP1 precursor - yeast (
*Contig1339_g3c12a1.rl 255 1.1e-19 4 258 sp|P51533|PDRA_Y ATP-DEPENDENT PERMEASE PDR10 >pir||S55517
probabletransport protein PDR10 - yeast (Saccharomyces cere

i.other

<transport protein>

Contig1361_c5g12a1.f1 506 8.4e-48 360 908 gi|409547 (L07492) sugar transport protein [Saccharomyces cerevisiae]

<aquaporin>

c5f02a1.rl 284 2.8e-24 172 744 sp|P53386|AQPL_YEAST AQUAPORIN-LIKE PROTEIN YPR192W >pir||S58822
probablemembrane protein YPR192w - yeast (Sacc

D. Classes of Enzymes-No pathway specified

1. Oxidoreductases (24)

<OXIDOREDUCTASE>

Contig884_y8f01a1.f1 682 2e-66 101 964 gi|2407193 (AF017151) oxidoreductase [Aspergillus parasiticus]
Contig1472_e0e10a1.rl 668 5.5e-65 48 917 sp|O07575|YHDF_B HYPOTHETICAL OXIDOREDUCTASE IN CITA-SSPB
INTERGENICREGION >gnl|PID|e1191878 (Y14082) hypothetical pro
Contig1579_d5d03a1.f1 387 3.2e-35 82 492 sp|P11943|ACPM_N ACYL CARRIER PROTEIN, MITOCHONDRIAL PRECURSOR
(ACP)(NADH-UBIQUINONE OXIDOREDUCTASE 9.6 KD SUBUNIT) >p
Contig1776_alg04f2.f1 376 5.1e-34 37 660 sp|Q09851|YAEB_S HYPOTHETICAL OXIDOREDUCTASE C23D3.11 IN CHROMOSOME
I>pir||S62502 hypothetical protein SPAC23D3.11 - f
Contig465_d3f01a1.f1 335 1.1e-29 176 703 sp|Q39172|P1_ARA PROBABLE NADP-DEPENDENT OXIDOREDUCTASE P1
>pir||S57611zeta-crystallin homolog - Arabidopsis thaliana
Contig1418_c8g11a1.f1 307 1.1e-26 584 1117 sp|P76113|YNCE_E PUTATIVE NADP-DEPENDENT OXIDOREDUCTASE IN
TEHB-RHSEINTERGENIC REGION >gnl|PID|d1015800 (D90784) Possi
d3f01a1.rl 297 1.2e-25 194 652 dbj||D86417_36 (D86417) YfmJ [Bacillus subtilis] >gnl|PID|e1182735
(Z99108)similar to quinone oxidoreduct=PROBABLE NADP-DEPENDENT
OXIDOREDUCTASE
*Contig1393_k9c01a1.f1 247 3.7e-20 365 913 sp|P25145|YINL_L HYPOTHETICAL OXIDOREDUCTASE IN INLA 5'REGION
(ORFA)>gi|149673 (M67471) ORFA [Listeria monocytogenes]
Contig1296_g5h07a1.f1 203 1.6e-15 266 760 gnl|PID|e1287847 (AL022603) putative NADPH quinone oxidoreductase
[Arabidopsisthaliana]
Contig1046_c0e04a1.f1 163 2.1e-11 120 569 sp|P42317|YXJF_B HYPOTHETICAL OXIDOREDUCTASE IN PEPT-KATB
INTERGENICREGION >gnl|PID|d1012374 (D83026) homologous to ma
Contig1119_d5c07a1.rl 156 2.9e-10 124 627 gi|3293547 (AF072709) putative oxidoreductase [Streptomyces lividans]

<MONOOXYGENASE>
g8g03a1.r1 271 5.9e-22 30 650 sp|P55487|Y4ID_RHISN PROBABLE MONOOXYGENASE Y4ID >gi|2182441 (AE000078) Y4id[Rhizobium sp. NGR234]
p0f05a1.r1 181 1.8e-12 179 430 sp|P17549|CP53_ASPNG BENZOATE 4-MONOOXYGENASE (BENZOATE-PARA-HYDROXYLASE)>pir||S12015 benzoate 4-monooxygenase

<cytochrome P450>
i8e02a1.r1 436 2.3e-40 9 482 gnl|PID|e339903 (Z98974) putative cytochrome p450 [Schizosaccharomyces pombe]
f5b05a1.f1 238 1.3e-18 211 729 pir||S57337 trichodiene oxygenase 4 - fungus (Fusarium sporotrichioides)>gi|837032 (U22462)= tri4 gene is p450 monooxygenase of trichothecene biosyn
g6d12a1.f1 160 3.5e-08 186 476 sp|P48416|CP10_LYMST CYTOCHROME P450 X >pir||JX0225 cytochrome P450 CYP10 -great pond snail >bbs|115322 (S46130)

<CR(VI) REDUCTASE-FLAVIN OXIDOREDUCTASE family>
i2h01a1.f1 187 8e-14 21 452 sp|P96977|CHRR_PSESP CR(VI) REDUCTASE >gnl|PID|d1012488 (D83142) Cr(VI)reductase [Pseudomonas sp.]

<AMINE OXIDASE-OXIDATIVE DEAMINATION of AMINES>
k0g09a1.f1 516 6.9e-49 37 522 sp|Q12556|AMO1_ASPNG COPPER AMINE OXIDASE 1 >gi|1401157 (U31869) copper amineoxidase [Aspergillus niger]
k0g09a1.r1 310 3.8e-26 136 396 sp|Q12556|AMO1_ASPNG COPPER AMINE OXIDASE 1 >gi|1401157 (U31869) copper amineoxidase,HISTAMINE OXIDASE [Aspergillus niger]

<SEXUAL DIFFERENTIATION PROCESS PROTEIN-expressed during sexual diff in S. pombe>
m0e12a1.r1 113 3e-05 157 414 sp|P40902|ISP7_SCHPO SEXUAL DIFFERENTIATION PROCESS PROTEIN ISP7 >pir||S45496isp7 protein - fission yeast (Schi

<fructosyl amine:oxygen oxidoreductase>
n5a03a1.r1 561 1.3e-53 33 692 gi|2661130 (AF035700) fructosyl amine:oxygen oxidoreductase [Aspergillusfumigatus]

<chlorocatechol 1,2-dioxygenase-degradation>
k8a07a1.f1 211 1.6e-16 96 410 gnl|PID|d1013794 (D86544) hydroxyquinol-1, 2-dioxygenase [Ralstonia pickettii]
Contig489_o0g05a1.f1 159 4.5e-10 2 301 gi|2318013 (AF011355) isotrichodermin C-15 hydroxylase [Fusariumsporotrichioides]

<HYDROXYACID DEHYDROGENASE>
Contig1209_g7g12a1.f1 286 1.8e-24 275 604 sp|P30799|DDH_ZY 2-HYDROXYACID DEHYDROGENASE HOMOLOG >pir||D40649D-2-hydroxy-acid dehydrogenase (EC 1.1.99.6) - Zymomo

<SOL1 Protein>
Contig1090_o6d10a1.r1 502 2.3e-47 173 841 sp|P50278|SOL1_Y SOL1 PROTEIN >pir||S62015 SOL1 protein - yeast(Saccharomyces cerevisiae) >gi|1163192 (U43608) Sol1p[S

2. Transferases (1)
<ORNITHINE AMINOTRANSFERASE>
Contig1762_c1a08a1.f1 2172 2.4e-224 299 1660 sp|Q92413|OAT_EM ORNITHINE AMINOTRANSFERASE

(ORNITHINE--OXO-ACIDAMINOTRANSFERASE) >gi|1658173 (U74303) ornithine trans

3. Hydrolases (1)
<alkaline phosphatase>
j0g12a1.r1

369 5.3e-33 13 447 sp|P11491|PPB_YEAST REPRESSIBLE ALKALINE PHOSPHATASE PRECURSOR
>pir||s69648alkaline phosphatase (EC 3.1.3.1) -

4. Lyases (0)

5. Isomerases (0)

6. Ligases (0)

7. Synthetases (2)

<S-adenosylmethionine synthetase>

z6d05a1.r1 401 1.1e-36 232 675 sp|P48466|METK_NEUCR S-ADENOSYLMETHIONINE SYNTHETASE
(METHIONINEADENOSYLTRANSFERASE) (ADOMET SYNTHETASE) >pir||
Contig1039_s4b01a1.f1 264 4.2e-22 365 544 sp|P48466|METK_N S-ADENOSYLMETHIONINE SYNTHETASE
(METHIONINEADENOSYLTRANSFERASE) (ADOMET SYNTHETASE) >pir||s65800 meth

E. Non-enzymatic classes (not in defined pathways)

1. Zinc finger motif-DNA binding (11)

<zinc finger protein>

n3g08a1.r1 570 1.4e-54 63 566 gnl|PID|e254304 (X99094) zinc finger protein [Ascobolus immersus]
z7f05a1.f1 331 3e-29 127 603 gi|1438877 (U41287) zinc finger protein [Mus musculus]
x9f12a1.r1 185 3.4e-13 215 436 gnl|PID|e223435 (X95455) RING zinc finger protein [Gallus
gallus]>prf||2211437A RING finger protein [Gallu
Contig517_c8g02a1.f1 201 4.8e-13 281 997 gnl|PID|e1291640 (AL023288) Zinc finger protein [Schizosaccharomyces
pombe]
v3f10a1.f1 174 1e-11 209 532 sp|P38682|GLO3_YEAST ZINC FINGER PROTEIN GLO3 >pir||s50625 GLO3 protein
-yeast [Saccharomyces cerevisiae] >gi|6
z1h04a1.f1 157 8.3e-11 294 557 gnl|PID|d1021201 (D45213) zinc finger protein [Homo sapiens]
Contig1196_e9a09a1.f1 175 4.5e-10 318 626 sp|P34670|YO14_C HYPOTHETICAL ZINC FINGER PROTEIN ZK686.4 IN
CHROMOSOMEIII >pir||s44909 ZK686.4 protein - Caenorhabdit
z7d04a1.f1 154 1.8e-09 199 348 gnl|PID|e1291655 (AL023290) zinc finger C3HC4 type [Schizosaccharomyces
pombe]
*Contig749_y3d09a1.f1 143 2.1e-07 488 676 gi|3033395 (AC004238) putative zinc-finger protein [Arabidopsis
thaliana]
o0f10a1.r1 120 7.6e-06 78 191 gnl|PID|e1316877 (AL031349) zinc-finger protein [Schizosaccharomyces
pombe]
<KIN17 protein-crossreacts w recA antibody>
n8e10a1.r1 225 5.2e-18 25 426 prf||1713233A recA crossreacting protein KIN17 [Mus musculus]

2. Leucine zipper motif (1)

<leucine zipper>

Contig1642_c7b12a1.f1 140 6.3e-07 524 637 gnl|PID|e253252 (X99215) leucine zipper [Aspergillus niger]

VI. Unclassified (significant homolog but function uncertain in Aspergillus nidulans) (132)

<uncertain function>

g2c02a1.r1	908	2.3e-90	55 588	gi 3411262	(AF080599) Medusa [Emericella nidulans]
Contig1231_r5c11a1.f1	827	8.3e-82	10 921	gnl PID d1004374	(D16355) crml-N1 protein [Schizosaccharomyces pombe]=CHROMOSOME REGION MAINTENANCE PROTEIN 1
o6g08a1.r1	813	2.3e-80	57 533	gi 168081	(M59935) unidentified gene; ORF [Emericella nidulans]
i3e01a1.r1	778	1.4e-76	14 487	sp P03710 VMCB_LAMBDA	PORTAL PROTEIN (GPB) (MINOR CAPSID PROTEIN B)>pir VHBPBL minor capsid protein B - phage =possible contaminant from Lambda
n0f09a1.r1	777	1.7e-76	11 454	gi 1755051	(U56696) palFp [Emericella nidulans]=pH response
o4a08a1.r1	771	7.4e-76	6 443	sp P03765 Y146_LAMBDA	HYPOTHETICAL NIN REGION PROTEIN ORF-146 >pir QXBP4Lhypothetical protein C-146 (nin region=lambda contaminant?
Contig1548_d3a07a1.f1	548	1e-75	698 1291	sp P39743 R167_Y	REDUCED VIABILITY UPON STARVATION PROTEIN 167>pir S40887 RVS167 protein - yeast (Saccharomyces cerev
Contig1773_d1d09a1.f1	742	8.5e-73	118 1080	gi 2352898	(AF012091) cystein rich protein [Metarhizium anisopliae]
Contig1718_c8g07a1.f1	735	3.1e-71	322 1182	pir S62011	PHO85 protein - yeast (Saccharomyces cerevisiae) >gi 1163103(U43503) Lph16p [Saccharomyces cerevisiae
x7h06a1.r1	681	2.4e-66	5 607	gi 563253	(L32177) guanine nucleotide regulatory protein [Cryphonectriaparasitica]
Contig434_e0d10a1.f1	654	1.8e-63	226 639	sp P19815 SC1D_E	CONIDIUM-SPECIFIC PROTEIN >pir S12113 Spoc1-C1D protein- Emericella nidulans >gi 2419 (X54668) Spoc1
Contig466_d3e06a1.f1	645	1.6e-62	2 373	gi 215150	(J02459) O (DNA replication;299) [Bacteriophage lambda]
m0e10a1.r1	647	6.3e-62	22 459	pir D45029	crml+ protein - fission yeast (Schizosaccharomyces pombe)=CHROMOSOME REGION MAINTENANCE PROTEIN
Contig541_c7d04a1.r1	355	1.1e-61	31 498	pir S51256	probable membrane protein YDR105c - yeast (Saccharomycescerevisiae) >gi 633641 (Z47746) unknown [Sacc
rlh07a1.r1	593	5.2e-57	164 667	sp Q12499 NOP5_YEAST	NUCLEOLAR PROTEIN NOP5 >pir S58322 hypothetical proteinYOR310c - yeast (Saccharomyces cer
c3d07a1.r1	561	1.3e-53	58 642	sp P10962 MK16_YEAST	MAK16 PROTEIN >pir BVBYK6 MAK16 protein - yeast(Saccharomyces cerevisiae) >gi 171880= may be concerned with nuclear events of cell cycle
z7g06a1.f1	534	9.1e-51	114 602	pir S71153	het-c4 protein - Podospora anserina >gi 537939 (L36210) het-c[Podospora anserina]
Contig1455_c9f12a1.f1	523	1.2e-49	180 650	gnl PID e1285104	(AJ001732) rAsp f 4 [Aspergillus fumigatus]
u4a09a1.r1	475	1.7e-44	191 604	gi 1870215	(AC000133) ORF [Emericella nidulans]
w7c04a1.r1	465	1.8e-43	10 504	pir JC4516	protein kinase (EC 2.7.1.37) - fission yeast

o5b02a1.r1	462	4e-43	13 459	(Schizosaccharomycespombe) >gnl PID d1008838 sp P40107 GOG5_YEAST VANADATE RESISTANCE PROTEIN GOG5/VRG4/VAN2 >pir S50238vanadate resistance protein VAN2 ==REGULATION OF THE PHOSPHORYLATION OF A NUMBER OF PROTEINS
l3b09a1.r1	461	5.2e-43	142 594	sp P33300 SUR1_YEAST SUR1 PROTEIN >pir S41798 SUR1 protein - yeast(Saccharomyces cerevisiae) >gi 311163 (M9664
k5g12a1.r1	459	7.4e-43	121 525	sp Q39963 ER1_HEVBR ETHYLENE-INDUCIBLE PROTEIN HEVER >pir S60047ethylene-responsive protein 1 - Para rubber t
Contig537_c8a02a1.r1	431	7.2e-40	252 569	gnl PID e1250610 (AJ223315) rAsp f 7 [Aspergillus fumigatus
v8h01a1.r1	405	4.4e-37	8 829	sp P38789 SSF1_YEAST SSF1 PROTEIN >pir S46700 SSF1 protein - yeast(Saccharomyces cerevisiae) >gi 487948 (U0006=possible role in mating
c4c08a1.r1	366	5.3e-33	169 603	gnl PID d1013767 (D86381) Ran/sp11 binding protein [Schizosaccharomyces pombe]=may regulate transport of protein across nuclear membrane as part of cell cycle
Contig911_y8b01a1.f1	365	7.2e-33	216 857	sp Q13610 PWP1_H PERIODIC TRYPTOPHAN PROTEIN 1 HOMOLOG (KERATINOCYTEPROTEIN IEF SSP 9502) >gi 177765 (L07758) IEF SSP
Contig173_i8h09a1.f1	364	9.4e-33	205 474	gi 348156 (L16844) yps-3 [Histoplasma capsulatum]=yeast-phase-specific gene
Contig686_o9f10a1.f1	241	7.2e-32	454 642	pir S49326 Nascent polypeptide associated complex alpha chain - human>gi 556642 (X80909) Nascent polypeptide ass
m7f02a1.f1	354	1.1e-31	29 622	sp P25621 YCR8_YEAST PUTATIVE TRANSPORTER YCR28C >pir S19439 probablemembrane protein YCR028c - yeast (Sacchar
m7f02a1.r1	348	5.9e-31	236 607	gi 2981103 (AF052688) putative transmembrane transporter Lizlp[Schizosaccharomyces pombe] >gnl PID e1
Contig1553_rlg04a1.f1	344	1.2e-30	324 836	gi 603050 (U18061) CAP20 [Glomerella cingulata]
Contig69_m3d06a1.r1	351	3.4e-30	76 576	gi 1261823 (L77234) glycine rich protein [Neurospora crassa]
y7h02a1.f1	342	8.6e-30	83 691	sp P90587 WD66_PHYPO 66 KD STRESS PROTEIN (P66) >gi 1835727 (U86011) 66-kDastress protein p66 [Physarum polycep
b0e11a1.r1	335	1.1e-29	13 270	sp P78712 ARP3_NEUCR ACTIN-LIKE PROTEIN 3 >gi 1718497 (U79737) actin-relatedprotein 3 [Neurospora crassa]
i7f03a1.f1	333	1.8e-29	151 627	sp Q10494 YDG7_SCHPO PROBABLE OXIDOREDUCTASE C26F1.07 IN CHROMOSOME I>gnl PID e241770 (Z73100) unknown [Schizos
Contig435_e0c12a1.r1	315	1.1e-27	486 740	gnl PID e1285104 (AJ001732) rAsp f 4 [Aspergillus fumigatus]
Contig1793_c3c10a1.f1	310	4.8e-27	88 306	sp P10713 CONX_N CONIDIATION-SPECIFIC PROTEIN 10 >pir A31849conidiation-specific protein - Neurospora crassa >gi 1687
Contig420_n8a04a1.f1	305	4.2e-26	247 744	sp Q10097 YAOI_S PUTATIVE TRANSPORTER C11D3.18C >gi 1107907 (Z68166)unknown [Schizosaccharomyces pombe]
c7e10a1.r1	292	4.2e-25	363 758	gi 1326076 (U28150) adrenoleukodystrophy related protein (Homo sapiens)
w9d08a1.r1	299	5e-25	84 440	gnl PID e1263921 (AL022104) kinase-binding protein 1 [Schizosaccharomycespombe]
y9h11a1.r1	290	7e-25	56 532	sp P53859 YNX2_YEAST HYPOTHETICAL 31.6 KD PROTEIN IN SIN4-URE2 INTERGENICREGION >pir S63198 hypothetical prote

i0c11a1.r1	288	1e-24	11 454	gi 1845332	(U60450) putative ATPase [Gigaspora rosea]
Contig951_c8b09a1.f1	176	5.3e-24	203 418	sp Q02336 ADA2_Y	POTENTIAL TRANSCRIPTIONAL ADAPTOR >pir A43252
r7h01a1.r1	292	9.4e-24	412 726	gnl PID e325416	(Z97211) probable involvement in ergosterol synthesis [Schizosaccharomyces pombe]
v3g01a1.r1	270	8.6e-23	128 562	gnl PID d1031084	(AP000004) 387aa long hypothetical amidohydrolase [Pyrococcus horikoshii]
c0g09a1.f1	159	8.9e-23	369 575	sp Q92377 MD12_SCHPO	MITOCHONDRIAL INHERITANCE COMPONENT MDM12 >gi 1655884(U64674) required for mitochondrial inheritance in budding and fission yeast
w9a03a1.r1	279	1.6e-22	40 513	sp P26674 STE6_SCHPO	STE6 PROTEIN >pir S28098 ste6 protein - fission yeast [Schizosaccharomyces pombe] >gi 5101
e7f02a1.r1	259	1.3e-21	202 489	gnl PID e1175783	(AJ002894) OsGRP2 [Oryza sativa]=RNA binding protein
glg12a1.r1	264	7.3e-21	42 509	pir S55945	STE23 protein - yeast [Saccharomyces cerevisiae]
c5f04a1.r1	247	2.3e-20	67 423	>gi 625109(U19729) Ste23p	similar to insulin protease
i7f03a1.r1	247	2.4e-20	42 305	sp P32495 NHP2_YEAST	HIGH MOBILITY GROUP-LIKE NUCLEAR PROTEIN 2 >pir S67767 high mobility group-like protein NH
c6c10a1.r1	251	6e-20	14 460	sp Q12458 YPR1_YEAST	PUTATIVE REDUCTASE 1 >pir S61163 YPR1 protein - yeast [Saccharomyces cerevisiae] >gi 84918
Contig913_c6h06a1.r1	244	8.7e-20	76 456	gnl PID e339034	(Z98762) hypothetical acetyl hydrolase [Schizosaccharomyces pombe]=enzyme involved in antibiotic bialaphos biosynthesis
z6h10a1.r1	182	8.9e-20	186 542	sp P41890 SCN1_S	SCN1 PROTEIN >pir B55164 scn1 protein - fission yeast [Schizosaccharomyces pombe] >gnl PID d1007203 (
f5g01a1.f1	232	2.3e-18	154 525	sp Q09698 YA27_SCHPO	HYPOTHETICAL 68.8 KD PROTEIN C2F7.07C IN CHROMOSOME I >pir S58151 hypothetical protein SPA
l0f04a1.f1	226	3.8e-18	252 443	sp P53693 RDS1_SCHPO	RDS1 PROTEIN >pir S58477 rds1 protein - fission yeast [Schizosaccharomyces pombe] >gnl PID
t2c06a1.r1	230	8.9e-18	36 440	gnl PID e1216790	(AL021046) probable involvement in transcription initiation [Schizosaccharomyces pombe]
o8h04a1.f1	230	1.3e-17	269 706	gi 3249066	(AC004473) Similar to S. cerevisiae SIK1P protein gb 984964. ESTsgb F15433 and gb AA395158
h1h05a1.r1	219	2.1e-17	74 547	gi 517205	(U09352) 67 kDa Myosin-crossreactive streptococcal antigen [Streptococcus pyogenes]
g0a04a1.f1	221	2.2e-17	98 529	gnl PID e1231246	(AJ001261) NIPSNAP2 protein [Mus musculus]
c6d11a1.r1	221	6.2e-17	118 780	gnl PID e1293292	(AL023592) rna binding protein [Schizosaccharomyces pombe]
w8g04a1.r1	219	2.1e-16	19 576	gnl PID e1314296	(AL031182) putative lipoprotein [Streptomyces coelicolor]
Contig1276_j7b11a1.f1	215	2.6e-16	112 477	gi 531469	(U12973) renal osmotic stress-induced Na-Cl organic solute cotransporter [Rattus norvegicus]
g4d03a1.r1	205	2.1e-15	38 562	sp P32783 ABD1_Y	ABD1 PROTEIN >pir S41782 ABD1 protein - yeast [Saccharomyces cerevisiae] >gi 170966 (L12000) ABD1 [Sa
				sp Q15392 DIMH_HUMAN	DIMINUTO-LIKE PROTEIN >gnl PID d1003311 (D13643)

Contig118_k0e09a1.f1	211	4.3e-15	84 287	KIAA0018[Homo sapiens] gi 2804455 (AF043699) similar to a human orf (GB:D13642) and human UV-damagedDNA binding factor (GB:U32986) in s
z5e10a1.r1	206	6.9e-15	23 616	sp P53946 ARP5_YEAST ACTIN-LIKE PROTEIN ARP5 >pir S58718 probable nuclearprotein YNL059c - yeast (Saccharomyce
m0f05a1.f1	120	7.5e-15	68 196	pir S33788 luciferase - southern Russian firefly >bbs 133113 (S61961)luciferase [Luciola mingrelica=E
x9d07a1.r1	197	2.4e-14	13 486	sp P32832 NPL6_YEAST NPL6 PROTEIN >pir S30792 NPL6 protein - yeast(Saccharomyces cerevisiae) >gi 172050 (M9843=possible involvement in nuclear protein localisation
hlh05a1.f1	196	2.4e-14	352 801	gnl PID e1231231 (AJ001258) NIPSNAP1 protein [Homo sapiens]
Contig1121_u4c11a1.r1	208	6.1e-14	527 931	sp P21560 CBP3_Y CBP3 PROTEIN PRECURSOR >pir A34290 membrane protein CBP3- yeast (Saccharomyces cerevisiae) >gi 17117
Contig222_i0b03a1.f1	184	4.8e-13	273 587	sp P41890 SCN1_S SCN1 PROTEIN >pir B55164 scn1 protein - fission yeast(Schizosaccharomyces pombe) >gnl PID d1007203 (
i3g06a1.r1	191	7.1e-13	29 433	sp P40340 TBP7_YEAST TAT-BINDING HOMOLOG 7 >pir S64603 YTA7 protein - yeast(Saccharomyces cerevisiae) >gnl PID
r7f01a1.r1	185	7.8e-13	389 766	sp P40445 YIQ6_YEAST PUTATIVE TRANSPORTER YIL166C >pir S50361 probablemembrane protein YIL166c - yeast (Saccha
Contig693_x8g03a1.r1	184	1.1e-12	160 561	gnl PID d1012479 (D83125) secretory component [Sarcophaga peregrina]
d0e03a1.f1	174	1.4e-12	193 435	sp P36149 YK48_YEAST HYPOTHETICAL 22.1 KD PROTEIN IN CCP1-MET1 INTERGENICREGION >pir S38144 =bet3 homolog-
g4c09a1.r1	186	2e-12	31 453	sp P15904 PCR_AVESA PROTOCHLOROPHYLLIDE REDUCTASE (PCR)(NADPH-PROTOCHLOROPHYLLIDE OXIDOREDUCTASE) >pir S08406electron transport in chlorophyll biosynthesis?
f0h04a1.r1	166	2.4e-11	219 554	gnl PID d1002364 (D11111) chloroplast 33 kd ribonucleoprotein (cp33)[Nicotiana sylvestris] >gi 311952 (X583
g3f09a1.r1	174	2.6e-11	30 422	sp P40528 SYG1_YEAST SYG1 PROTEIN >pir S49931 SYG1 protein - yeast(Saccharomyces cerevisiae) >gi 600001 (Z4686
c1f03a1.f1	169	6.6e-11	109 564	gi 2367392 (U82513) random slug cDNA25 protein [Dictyostelium discoideum]
g0b09a1.r1	166	7.3e-11	58 354	gnl PID e1316378 (AJ010169) Ariadne-2 protein [Drosophila melanogaster]
z6h07a1.r1	183	7.6e-11	50 538	gi 1098491 (U12141) Ynl2515p [Saccharomyces cerevisiae]
n3b09a1.r1	163	1.2e-10	365 589	sp Q02336 ADA2_YEAST POTENTIAL TRANSCRIPTIONAL ADAPTOR >pir A43252 probabletranscriptional adaptor ADA2 - yeas
g8b01a1.r1	184	1.4e-10	25 339	gi 2109297 (U97696) cyclooxygenase-2 [Oryctolagus cuniculus]required for prostiglandin synthesis
Contig1817_alh05c9.r1	180	9.6e-10	752 1237	sp Q03465 SON1_Y NUCLEAR PROTEIN SON1 (UB FUSION DEGRADATION PROTEIN 5)>pir S41986 nuclear protein SON1 - yeast (Sacc
hlg12a1.r1	154	2.3e-09	109 276	gnl PID d1018112 (D90905) rehydrin [Synechocystis sp.]
Contig1217_n2g11a1.r1	141	3.9e-09	215 418	gi 3168840 (U88711) copper homeostasis factor [Arabidopsis thaliana]
r1e08a1.r1	142	7.2e-09	335 547	gnl PID e1316908 (AL031350) putative dehydrogenase [Streptomyces

i0a07a1.f1	142	8.2e-09	126	503	coelicolor] gnl PID d1016518 (D90835) H-NS-repressed protein, 30K [Escherichia coli]
c5d12a1.r1	144	3.6e-07	24	158	sp P19541 YP33_YEAST PUTATIVE TRANSCRIPTIONAL REGULATORY PROTEIN IN MKK2-COX11INTERGENIC REGION >pir s69051 hy
v8g01a1.r1	132	6.8e-07	7	150	sp P52977 LON_CAUCR ATP-DEPENDENT PROTEASE LA >gi 1667399 (U56652) lonprotease [Caulobacter crescentus]-DNA methylation control
Contig1298_j4e02a1.f1	118	1e-06	152	307	pir A54523 histidine-rich protein - Plasmodium lophurae (fragment) >gi 552196(M15317) histidine-rich protein [P1
w7c04a1.f1	142	1.6e-06	430	570	pir JC4516 protein kinase (EC 2.7.1.37) - fission yeast (Schizosaccharomycespombe) >gnl PID d1008838
z6h04a1.f1	138	2.2e-06	44	349	gnl PID e1252031 (AL021899) hypothetical protein Rv2052c [Mycobacteriumtuberculosis]
r2d07a1.r1	138	2.7e-06	427	681	sp P54069 BE46_SCHPO BEM46 PROTEIN >gi 987287 (U29892) temperature sensitivesuppressor of Saccharomyces cerevisi
Contig1532_d2g07a1.r1	135	3.9e-05	48	299	gi 172672 (M13629) sporulation protein [Saccharomyces cerevisiae]
Contig1109_u4a07a1.r1	126	4.1e-05	357	626	gnl PID e236571 (X96977) cell wall anchoring signal [Enterococcus faecalis]
v4g04a1.r1	127	8.9e-05	337	585	gi 1572821 (U70856) weak similarity to rat cytosolic acyl coenzyme A thioesterhydrolase (GB:U49694)C. elegans
Contig412_e9e05a1.r1	120	0.00024	309	452	sp P53081 NIF3_Y NGG1-INTERACTING FACTOR 3 >pir s64243 hypotheticalprotein YGL221c - yeast (Saccharomyces cerevisiae)
<bacteriorhodopsin>					
Contig1753_c8f10a1.f1	180	2.1e-11	367	1116	sp P02945 BACR_H BACTERIORHODOPSIN PRECURSOR (BR) >pir RAHSBbacteriorhodopsin precursor - Halobacterium halobium >pir
<LEUKOTRIENE biosynthesis>					
Contig246_hlg06a1.r1	723	9.1e-71	3	680	sp Q10740 LKHA_Y PROBABLE LEUKOTRIENE A-4 HYDROLASE (LTA-4 HYDROLASE)(LEUKOTRIENE A(4) HYDROLASE) >pir s61099 leukotr
Contig247_hlg06a1.f1	172	2.9e-11	193	342	sp Q10740 LKHA_Y PROBABLE LEUKOTRIENE A-4 HYDROLASE (LTA-4 HYDROLASE)(LEUKOTRIENE A(4) HYDROLASE) >pir s61099 leukotr
<bleomycin hydrolase>					
Contig476_d3b04a1.f1	499	4.8e-47	51	884	sp Q13867 BLMH_H BLEOMYCIN HYDROLASE (BLM HYDROLASE) (BMH)>gnl PID e205512 (X92106) bleomycin hydrolase [Homo sapiens]
<transposase>					
*Contig1404_g5h09a1.f1	294	7e-24	503	898	pir s60179 pol polyprotein homolog - fungus (Fusarium oxysporum)retrotransposon skippy >gi 510697 (L34658) pol p
w4c11a1.r1	201	2.9e-15	6	155	bbs 175409 (S80872) putative Tc1-mariner class transposase/IS630-Tc1 homolog[Aspergillus niger, chlor
g9e10a1.f1	201	4.6e-14	11	421	gnl PID e1273348 (AL022140) LTR retrotransposon like protein [Arabidopsisisthaliana]
Contig1184_g6g01a1.r1	189	1.2e-12	271	741	pir s32437 pol polyprotein - Volvox carteri f. nagariensis retrotransposonOssex >gi 288597 (X69552) gag,protease

<prohibitin> y3b11a1.r1	425	1e-68	39 386	gi 2582388	(AF022225) prohibitin [<i>Pneumocystis carinii</i>]
<DIPHTHINE SYNTHASE> Contig1021_s9d11a1.f1	531	7.3e-87	351 839	sp P32469 DPH5_Y	DIPHTHINE SYNTHASE (DIPHTAMIDE BIOSYNTHESIS METHYLTRANSFERASE) >pir S30890 methyltransferase DPH5 - y
<Spoc1-C1c protein> m5f12a1.r1	652	3.1e-63	61 510	pir S27412	Spoc1-C1c protein - <i>Emericella nidulans</i> >gi 168091 (M83571)Spoc1-C1c [<i>Emericella nidulans</i>]
Contig1034_m5f12a1.f1	527	5.2e-50	127 444	pir S27412	Spoc1-C1c protein - <i>Emericella nidulans</i> >gi 168091 (M83571)Spoc1-C1c [<i>Emericella nidulans</i>]
<PEPTIDASE> Contig1724_c1f11a1.f1	630	6.6e-61	72 857	sp P43590 YFH6_Y	HYPOTHETICAL 61.8 KD PEPTIDASE IN MPR1-GCN20 INTERGENICREGION >pir S56261 probable membrane protein
Contig1633_g0h01a1.f1	480	4.5e-45	93 707	gi 1763684	(U81483) pre-pro-penicillopepsin-JT2 [<i>Penicillium janthinellum</i>]=(PEPTIDASE A) >pdb 3APP Acid Proteinase
Contig400_f0g06a1.f1	280	3.7e-23	463 978	sp P43590 YFH6_Y	HYPOTHETICAL 61.8 KD PEPTIDASE IN MPR1-GCN20 INTERGENICREGION >pir S56261 probable membrane protein
Contig1040_c6c03a1.f1	270	8.1e-23	7 312	gi 1763684	(U81483) pre-pro-penicillopepsin-JT2 [<i>Penicillium janthinellum</i>](PEPTIDASE A) >pdb 3APP Acid Proteinase
Contig1328_d4b06a1.f1	270	8.2e-23	7 312	gi 1763684	(U81483) pre-pro-penicillopepsin-JT2 [<i>Penicillium janthinellum</i>]=(PEPTIDASE A) >pdb 3APP Acid Proteinase
Contig1580_c1f11a1.r2	264	1.9e-21	36 647	sp P43590 YFH6_Y	HYPOTHETICAL 61.8 KD PEPTIDASE IN MPR1-GCN20 INTERGENICREGION >pir S56261 probable membrane protein
<regulatory protein> Contig395_f1d08a1.r1	235	4.6e-19	50 439	pir A57145	regulatory protein prrC - <i>Rhodobacter sphaeroides</i> >gi 733128(U22347) membrane-anchored regulatory pro
Contig1020_c9h08a1.f1	165	2.6e-09	62 244	sp P28348 NIRA_E	NITROGEN ASSIMILATION TRANSCRIPTION FACTOR NIRA>pir A41697 nitrate assimilation regulatory protein n
Contig301_g7a11a1.f1	137	3.4e-06	390 593	pir A61382	phosphorylation regulatory protein HP-10 - human
<MAL3 PROTEIN> Contig1424_c8b11a1.f1	372	3.3e-49	166 516	sp Q10113 MAL3_S	MAL3 PROTEIN >gnl PID e213819 (Z68198) unknown[<i>Schizosaccharomyces pombe</i>] >gnl PID e282230 (Y09518) M
<GRR1 PROTEIN-GLUCOSE REPRESSION PATHWAY> Contig1516_c9h12a1.r1	734	2.9e-71	22 891	sp P24814 GRR1_Y	GRR1 PROTEIN >pir A41529 GRR1 protein - yeast(<i>Saccharomyces cerevisiae</i>) >gi 171617 (M59247) putative
<PIG8> Contig1742_g2b04a1.f1	326	9.5e-29	203 985	gi 1764133	(U81790) PIG8 [<i>Uromyces fabae</i>]
<PHO85 protein> Contig516_c8g07a1.r1	211	4.3e-15	37 402	pir S62011	PHO85 protein - yeast (<i>Saccharomyces cerevisiae</i>) >gi 1163103(U43503) Lph1p [<i>Saccharomyces cerevisiae</i>]
<Argonaute protein> Contig664_o8h09a1.f1	274	6.8e-22	215 715	gi 2149640	(U91995) Argonaute protein [<i>Arabidopsis thaliana</i>]

<BC-2 protein-putative breast adenocarcinoma marker>

Contig705_p0c12a1.f1 322 2.6e-28 325 729 gi|2828147 (AF042384) BC-2 protein [Homo sapiens]

<POS5 protein>

Contig787_s8a06a1.f1 321 3.5e-28 345 704 pir||s65200 POS5 protein - yeast (Saccharomyces cerevisiae)
>gnl|PID|e247057(Z73544) ORF YPL188w [Saccharomyces c

<G10 PROTEIN>

Contig979_j9h07a1.f1 384 4.4e-48 122 454 sp|P12805|G10_XE G10 PROTEIN >pir||s05955 G10 protein - African clawed frog>gi|64704 (X15243) G10 protein (AA 1-144) [

<COLD SHOCK PROTEIN>

Contig1802_d1e12a1.f1 146 1e-09 147 338 sp|P95459|CSPA_P MAJOR COLD SHOCK PROTEIN CSPA >gi|1778825 (U82822) majorcold shock protein Cspa [Pseudomonas aerugino

<GLUCOSE-REPRESSIBLE GENE PROTEIN>

Contig1809_c1a06a1.f1 199 2.4e-15 68 280 sp|P22151|GRG1_N GLUCOSE-REPRESSIBLE GENE PROTEIN >gi|3014 (X14801) grglgene product [Neurospora crassa]

Contig1843_a5a07a1.f1 177 5.6e-13 87 293 sp|P22151|GRG1_N GLUCOSE-REPRESSIBLE GENE PROTEIN >gi|3014 (X14801) grglgene product [Neurospora crassa]

Contig1859_ala06f2.f1 177 5.6e-13 138 344 sp|P22151|GRG1_N GLUCOSE-REPRESSIBLE GENE PROTEIN >gi|3014 (X14801) grglgene product [Neurospora crassa]

<EF hand protein>

Contig630_o8c05a1.f1 307 1.1e-26 210 599 gi|2459421 (AC002332) putative calcium-binding EF-hand protein [Arabidopsisthaliana]

VII. Unidentified (includes significant match with ORFs) (366)

<unknown function>

Contig557_c6b02a1.r1 1058 2.7e-106 1 1203 gnl|PID|d1022254 (AB004535) hypothetical protein YPR112c [Schizosaccharomycespombe]

Contig1626_c4c09a1.f1 945 2.5e-94 178 954 gnl|PID|d1014558 (D89200) similar to Saccharomyces cerevisiae reducedviability upon starvation protein 161, SWISS-PROT

Contig1719_f5d08a1.r1 937 1.9e-93 238 1266 sp|P53753|YN96_Y HYPOTHETICAL 121.1 KD PROTEIN IN BIO3-HXT17 INTERGENICREGION PRECURSOR >pir||s63399 probable membrane

Contig1798_c5e07a1.r1 927 2.1e-92 278 1207 pir||s67089 hypothetical protein YOR197w - yeast (Saccharomyces cerevisiae)>gnl|PID|e252390 (Z75105) ORF YOR197w

c6c01a1.r1 915 3.8e-91 19 837 sp|Q10178|YAV9_SCHPO HYPOTHETICAL 137.2 KD PROTEIN C27F1.09C IN CHROMOSOME I>gi|1182046 (Z69368) unknown [Schiz

n8b04a1.f1 893 8.9e-89 10 537 gi|1870209 (AC000133) ORF [Emericella nidulans]

Contig1587_i8b04a1.r1 838 5.1e-83 6 1124 sp|Q03655|YM64_Y HYPOTHETICAL 56.8 KD PROTEIN IN SCJ1-GUAL INTERGENICREGION PRECURSOR >pir||s55097 probable membrane p

Contig1379_c6c09a1.f1 543 2e-78 440 904 sp|P49954|YL85_Y HYPOTHETICAL 32.5 KD PROTEIN YLR351C

Contig1506_c7b08a1.f1 374 1.3e-73 695 1141 >pir||s51459hypothetical protein YLR351c - yeast (Saccharomyces

Contig1506_c7b08a1.f1 374 1.3e-73 695 1141 gnl|PID|e1287784 (AL022600) hypothetical protein [Schizosaccharomyces pombe]

h0h04a1.f1 748 4.6e-73 37 534 sp|Q04336|YM54_YEAST HYPOTHETICAL 126.6 KD PROTEIN IN RPL39-VTI1

Contig1545_h4b06a1.f1	737	3e-72	89 1252	gnl PID e1285394	INTERGENICREGION >pir S50925 hypothetical pro (AL022305) hypothetical protein [Schizosaccharomyces pombe]
Contig1401_f0g09a1.r1	725	5.2e-71	30 887	pir S59389	probable membrane protein YLR243w - yeast (Saccharomyces cerevisiae) >gi 662338 (U20865) Ylr243wp [Sac
Contig1568_c5h09a1.f1	695	7.5e-68	188 910	sp Q08193 YOD0_Y	HYPOTHETICAL 51.9 KD PROTEIN IN MSE1-LAG2 INTERGENICREGION PRECURSOR >pir S66713 hypothetical protei
Contig1729_a5h10a1.f1	681	2.4e-66	298 1077	sp P53252 YG2J_Y	HYPOTHETICAL 38.3 KD PROTEIN IN RPL16B-PDC6 INTERGENICREGION >pir S64381 hypothetical protein YGR086
Contig530_o4f02a1.r1	681	2.4e-66	9 746	sp P40055 YER2_Y	HYPOTHETICAL 62.3 KD PROTEIN IN PTP3-ILV1 INTERGENICREGION >pir S50585 hypothetical protein YER082c
Contig329_g4g01a1.r1	670	3.6e-65	33 917	sp Q09885 YAH9_S	HYPOTHETICAL 43.0 KD PROTEIN C8A4.09C IN CHROMOSOME I>pir S62525 hypothetical protein SPAC8A4.09c -
Contig1384_c3c09a1.r1	649	6e-63	127 732	sp O13917 YDWD_S	HYPOTHETICAL 23.6 KD PROTEIN C23C11.13C IN CHROMOSOME I>gnl PID e334141 (Z98559) SPAC23C11.13c; len:2
c8a01a1.r1	622	4.5e-60	23 718	sp Q92359 YDHE_SCHPO	HYPOTHETICAL 73.3 KD PROTEIN C6G9.14 IN CHROMOSOME I>gnl PID e276617 (Z81317) serine rich
Contig883_r4f07a1.f1	400	8.5e-60	169 546	sp Q92342 YDI4_S	HYPOTHETICAL 50.4 KD PROTEIN C1F8.04C IN CHROMOSOME I>gnl PID e276494 (Z81312) unknown [Schizosacchar
Contig1565_r4b01a1.f1	614	3.1e-59	281 1081	gnl PID d1019605	(D90917) hypothetical protein [Synechocystis sp.] x8f06a1.r1
x8f06a1.r1	606	2.2e-58	13 552	gnl PID e1294549	(AL023706) hypothetical protein [Schizosaccharomyces pombe]
m1d02a1.r1	603	4.6e-58	48 650	pir S67033	hypothetical protein YOR145c - yeast (Saccharomyces cerevisiae)>gi 1293706 (U55020) O3513p
o8g08a1.r1	597	1.9e-57	16 552	gnl PID e334145	(Z98559) SPAC23C11.17; len:485aa, similar eg. to YPR125W,Q06493, chromosome xvi orf, (454a=S. pombe
y4g01a1.r1	595	3e-57	19 627	gnl PID e334163	(Y14554) pSI-7 protein [Cladosporium fulvum]
v7c12a1.r1	594	4.3e-57	13 621	gnl PID e330194	(Z93386) R11H6.1 [Caenorhabditis elegans]
n8d02a1.r1	584	4.6e-56	6 614	gnl PID e334344	(Z98598) hypothetical protein [Schizosaccharomyces pombe]
i7e12a1.f1	580	1.3e-55	120 683	pir S61980	hypothetical protein YPL086c - yeast (Saccharomyces cerevisiae)>gi 1151240 (U43281) Lpg22p
w4d09a1.r1	582	6.1e-55	8 523	sp Q10251 YD23_SCHPO	HYPOTHETICAL 119.9 KD PROTEIN C56F8.03 IN CHROMOSOME I>gi 1204225 (Z69728) unknown [Schizo
Contig1844_c3c01a1.f1	569	4.3e-54	29 1360	sp Q10327 YD72_S	HYPOTHETICAL 97.1 KD PROTEIN C32A11.02C IN CHROMOSOME I>gi 1213266 (Z69796) unknown [Schizosaccharomy
f1g08a1.r1	565	5e-54	24 455	sp P25586 YCF9_YEAST	HYPOTHETICAL 37.2 KD PROTEIN IN CHA1-PRD1 INTERGENICREGION >pir S19389 hypothetical protein
Contig1530_a5d09a1.f1	562	1e-53	412 915	sp P53224 YG1Q_Y	HYPOTHETICAL 25.2 KD PROTEIN IN ACB1-K8S1 INTERGENICREGION >pir S64329 probable membrane protein YGR
Contig778_y4e04a1.r1	556	1e-52	118 690	pir S74280	hypothetical protein YCL054w - yeast (Saccharomyces cerevisiae)>gnl PID e309034 (X59720) YCL054w, len
o1f05a1.f1	347	1.3e-52	15 293	sp Q10325 YD6D_SCHPO	HYPOTHETICAL 47.3 KD PROTEIN C17G8.13C IN CHROMOSOME

d3b03a1.r1	530	2.4e-50	80 658	I>gi 1213262 (Z69795) unknown [Schizo gnl PID e1249764 (AL021730) hypothetical protein [Schizosaccharomyces pombe]
y4h02a1.r1	522	1.8e-49	31 510	gnl PID e1256479 (AL022071) hypothetical protein [Schizosaccharomyces pombe]
c6h03a1.r1	515	8.8e-49	18 578	gnl PID e1287786 (AL022600) hypothetical protein [Schizosaccharomyces pombe]
o8g06a1.r1	511	2e-48	12 563	sp P39941 YE10_YEAST HYPOTHETICAL 56.5 KD PROTEIN IN HXT8 5'REGION AND IN PAU65'REGION >pir S50519 hypothetica
t2c07a1.r1	509	4e-48	9 437	pir S58091 probable membrane protein YDR091c - yeast (Saccharomyces cerevisiae) >gi 914875 (Z50111) un
m5a07a1.r1	409	5.7e-48	82 450	sp P43547 YFF6_YEAST HYPOTHETICAL 23.9 KD PROTEIN IN THI5-AGP3 INTERGENICREGION >pir S56199 hypothetical prote
i3a04a1.r1	516	8.3e-48	134 487	sp Q10251 YD23_SCHPO HYPOTHETICAL 119.9 KD PROTEIN C56F8.03 IN CHROMOSOME I>gi 1204225 (Z69728) unknown [Schizo
j9b03a1.r1	519	8.4e-48	15 479	pir S70099 hypothetical protein YDR334w - yeast (Saccharomyces cerevisiae)>gi 1230661 (U51032) Ydr334
Contig1381_c6d02a1.r1	500	3.4e-47	68 592	sp P47096 YJZ5_Y HYPOTHETICAL 20.2 KD PROTEIN IN MER2-CPR7 INTERGENICREGION >pir S57043 hypothetical protein YJR025c
m7e10a1.f1	498	5.9e-47	4 447	gnl PID e343814 (Z99126) hypothetical protein [Schizosaccharomyces pombe]
b0e08a1.f1	503	3.4e-46	11 487	gnl PID e291018 (Y08997) 146kDa nuclear protein [Xenopus laevis]
Contig1771_c9f07a1.f1	491	3.4e-46	116 1075	sp O13716 YDZ9_S HYPOTHETICAL 44.5 KD PROTEIN C14C4.09 IN CHROMOSOME I>gnl PID e334267 (Z98596) hypothetical protein [
Contig536_c8a03a1.f1	491	3.4e-46	629 1030	pir B26955 hypothetical protein - yeast (Yarrowia lipolytica) (fragment)
m2g01a1.f1	481	3.8e-45	197 715	pir S67622 hypothetical protein YDL086w - yeast (Saccharomyces cerevisiae)>gnl PID e253022 (Z74134) O
m5a07a1.f1	474	2.1e-44	165 725	sp P42884 YN71_YEAST HYPOTHETICAL 42.0 KD PROTEIN IN THI12-RPD3 INTERGENICREGION >pir S51335 probable aryl-alc
Contig604_g2h07a1.f1	471	4.3e-44	108 842	sp Q09686 YA14_S HYPOTHETICAL 28.0 KD PROTEIN C13C5.04 IN CHROMOSOME I>pir S58096 hypothetical protein SPAC13C5.04 -
n8e12a1.r1	465	2e-43	130 699	pir S67695 hypothetical protein YDL147w - yeast (Saccharomyces cerevisiae)>gnl PID e242702 (X97751) D
10b09a1.r1	460	1.4e-42	122 511	pir S61717 probable membrane protein YOL060c - yeast (Saccharomyces cerevisiae) >gi 984180 (X91067) O1
Contig1428_c8c10a1.f1	451	5.3e-42	221 673	pir S69049 hypothetical protein YPL135w - yeast (Saccharomyces cerevisiae)>gi 1244779 (U43703) Lpi10p [Saccharom
y4d02a1.r1	451	5.9e-42	37 504	pir S69699 hypothetical protein YDR415c - yeast (Saccharomyces cerevisiae)>gi 927713 (U33007) Ydr415c
Contig851_z3b12a1.f1	433	4.7e-40	183 611	gi 2583079 (AF026816) putative oncogene protein [Homo sapiens]
Contig1405_c9b05a1.f1	432	6e-40	130 792	sp P53290 YG3T_Y HYPOTHETICAL 38.6 KD PROTEIN IN TIF4631-KRE11 INTERGENICREGION >pir S64474 probable membrane protein

Contig1838_c4g07a1.f1	428	1.3e-39	523 972	gnl PID e1283575	(AJ224865) IgE-binding protein [Aspergillus fumigatus]
zle04a1.f1	428	2.9e-39	103 561	sp Q09909 YAJ9_SCHPO	HYPOTHETICAL 74.4 KD PROTEIN C30D11.09 IN CHROMOSOME
Contig207_i2a02a1.f1	318	3.6e-39	290 697	I>pir s62567	hypothetical protein SP
Contig134_m6a03a1.f1	424	3.8e-39	219 938	gi 2583216	(AF029913) unknown [Cochliobolus heterostrophus]
o5f09a1.r1	421	8.7e-39	14 388	>gi 2598190(AF027687)	unknown [Cochliobolus heterost
c6g01a1.r1	416	3.1e-38	335 832	gnl PID d1014592	(D89234) similar to Saccharomyces cerevisiae ORF
Contig390_n3d12a1.r1	415	3.9e-38	116 565	YGR205W,EMBL Accession Number Z72990	[Schizosaccharo
rla10a1.r1	415	3.9e-38	138 659	sp Q03529 YM8I_YEAST	HYPOTHETICAL 44.9 KD PROTEIN IN URA10-NRC1
v7c12a1.f1	414	5e-38	136 639	INTERGENICREGION >pir s54484	probable membrane
Contig1485_c1d10a1.f1	412	7.2e-38	22 525	sp O14171 YE54_SCHPO	HYPOTHETICAL 30.2 KD PROTEIN C4D7.04C IN CHROMOSOME
Contig1207_g7h02a1.r1	407	2.5e-37	200 769	I>gnl PID e334311 (Z98602)	hypothetical
Contig1490_c3d05a1.r1	407	2.6e-37	283 864	sp Q10319 YD67_S	HYPOTHETICAL 24.9 KD PROTEIN C17G8.07 IN CHROMOSOME
e9b10a1.r1	407	2.8e-37	64 729	I>gi 1213256 (Z69795)	unknown [Schizosaccharomyce
p0h07a1.r1	411	1.8e-36	36 506	gnl PID e339276	(Z98850) hypothetical dehydrogenase
Contig1659_c7f11a1.f1	399	1.8e-36	128 763	[Schizosaccharomycespombe] >gnl PID e1314269	(AL031180
Contig856_x3c10a1.r1	398	2.3e-36	66 476	sp P43616 YFL4_YEAST	HYPOTHETICAL 52.9 KD PROTEIN IN SAP155-YMR31
c7a12a1.r1	398	6.8e-36	37 666	INTERGENICREGION >pir s56299	hypothetical pr
Contig777_y4d05a1.f1	401	1.3e-35	308 670	sp Q03677 YMO9_Y	HYPOTHETICAL 20.9 KD PROTEIN IN PLB1-HXT2
z1c01a1.f1	392	1e-34	4 588	INTERGENICREGION >pir s53039	hypothetical protein YMR009w
Contig594_c4d02a1.r1	391	1.3e-34	26 742	sp P38805 YHO8_Y	HYPOTHETICAL 35.1 KD PROTEIN IN NAM8-GAR1
o6b05a1.r1	380	2e-34	20 523	INTERGENICREGION >pir s46718	hypothetical protein YHR088w
e4e10a1.r1	387	8.2e-34	37 600	sp Q07821 YL27_Y	HYPOTHETICAL 27.7 KD PROTEIN IN PRP19-HSP104
				INTERGENICREGION >pir s64778	hypothetical protein YLL02
				sp O14057 YEB8_SCHPO	HYPOTHETICAL 58.0 KD PROTEIN C2C6.08 IN CHROMOSOME
				I>gnl PID e339287 (Z98887)	hypothetical
				pir s52525	probable membrane protein YPL006w - yeast
				(Saccharomycescerevisiae) >gi 683784 (Z48483)	un
				sp O13716 YDZ9_S	HYPOTHETICAL 44.5 KD PROTEIN C14C4.09 IN CHROMOSOME
				I>gnl PID e334267 (Z98596)	hypothetical protein [
				sp O14155 YE72_S	HYPOTHETICAL 15.9 KD PROTEIN C4A8.02C IN CHROMOSOME
				I>gnl PID e338958 (Z98762)	hypothetical protein [
				sp P40009 YEJ5_YEAST	HYPOTHETICAL 71.9 KD PROTEIN IN PMI40-PAC2
				INTERGENICREGION >pir s50463	hypothetical prot
				sp P42839 YN61_Y	HYPOTHETICAL 102.5 KD PROTEIN IN KRE1-HXT14
				INTERGENICREGION >pir s51293	probable membrane protein Y
				gnl PID e340027	(Z98980) hypothetical protein [Schizosaccharomyces
				pombe]	
				sp P40164 YNU1_Y	HYPOTHETICAL 98.1 KD PROTEIN IN SPX19-GCR2
				INTERGENICREGION >pir s50730	hypothetical protein YNL201c
				sp O14249 YE63_SCHPO	HYPOTHETICAL 48.7 KD PROTEIN C6G10.03C IN CHROMOSOME
				I>gnl PID e334323 (Z98603)	hypothetic
				sp Q10164 YAU9_SCHPO	HYPOTHETICAL 143.6 KD PROTEIN C26A3.09C IN CHROMOSOME
				I>gnl PID e220681 (Z69240)	hypotheti

Contig1629_a5b07a1.f1	365	7.2e-33	298 945	gi 2649154	(AE001006) membrane protein [Archaeoglobus fulgidus]
g0d02a1.f1	362	1.5e-32	215 697	gi 2983324	(AE000705) hypothetical protein [Aquifex aeolicus]
n8d02a1.f1	365	2.9e-32	261 518	gnl PID e334344	(Z98598) hypothetical protein [Schizosaccharomyces pombe]
m8a03a1.f1	358	3.8e-32	131 409	sp P36156 YK56_YEAST	HYPOTHETICAL 43.3 KD PROTEIN IN SIS2-MTD1 INTERGENICREGION >pir S38153 hypothetical prote
o1g04a1.f1	358	4.3e-32	130 624	sp Q09923 YAKC_SCHPO	HYPOTHETICAL 37.7 KD PROTEIN C1F7.12 IN CHROMOSOME I>pir s62584 hypothetical protein SPAC
Contig616_o6h08a1.r1	365	6e-32	128 532	sp Q09782 YA93_S	HYPOTHETICAL 85.7 KD PROTEIN C13G6.03 IN CHROMOSOME I>pir s62432 hypothetical protein SPAC13G6.3 - f
Contig984_c3e04a1.r1	356	7e-32	205 687	pir s52527	hypothetical protein YPL004c - yeast (Saccharomyces cerevisiae)>gi 683786 (Z48483) unknown [Saccharom
vlc10a1.r1	363	8.2e-32	24 524	pir s61717	probable membrane protein YOL060c - yeast (Saccharomycescerevisiae) >gi 984180 (X91067) 01
x8f04a1.f1	369	1e-31	270 593	sp Q04958 YMF9_YEAST	HYPOTHETICAL 187.1 KD PROTEIN IN OGG1-CNA2 INTERGENICREGION >pir s49802 probable membrane
j0b08a1.r1	362	1.1e-31	27 494	gnl PID e339160	(Z98850) hypothetical protein [Schizosaccharomyces pombe]>gnl PID e1314282 (AL031181) puta
Contig1095_o8e08a1.f1	356	1.1e-31	246 596	gnl PID e334108	(Z98532) hypothetical protein [Schizosaccharomyces pombe]
Contig1437_f5e03a1.f1	353	1.4e-31	26 826	gnl PID e1294540	(AL023705) hypothetical protein [Schizosaccharomyces pombe]
Contig1234_k8f08a1.f1	352	1.7e-31	306 1097	pir s57377	probable membrane protein YOL092w - yeast (Saccharomycescerevisiae) >gi 600466 (X83121) orf 00929 gen
Contig598_c4a07a1.f1	363	2.3e-31	482 889	sp P38144 YB95_Y	HYPOTHETICAL 131.1 KD HELICASE IN ALG7-ENP1 INTERGENICREGION >gnl PID e304681 (Z36114) ORF YBR245c [s
h1b12a1.r1	356	1.4e-30	41 385	pir s65236	probable membrane protein YPL217c - yeast (Saccharomycescerevisiae) >gnl PID e246934 (Z735
w5a03a1.r1	343	1.6e-30	53 481	pir JC4256	hypothetical 32.0k protein - Neurospora crassa >gi 773386 (L40806)open reading frame [Neur
m5e02a1.f1	342	1.9e-30	212 655	gnl PID d1025722	(AB010900) YNL123w homolog [Schizosaccharomyces pombe]
Contig441_n8e06a1.f1	225	2.2e-30	557 901	sp Q10063 YAM8_S	HYPOTHETICAL 53.9 KD PROTEIN C1F5.08C IN CHROMOSOME I>gi 1103735 (Z68136) unknown [Schizosaccharomyce
n5d03a1.r1	338	5.4e-30	197 673	sp P38286 YB09_YEAST	HYPOTHETICAL 38.7 KD PROTEIN IN RPB5-CDC28 INTERGENICREGION >pir s46030 probable membrane
w9b04a1.r1	337	7.1e-30	74 628	sp P47137 YJ66_YEAST	PROBABLE OXIDOREDUCTASE YJR096W >pir s57117 aldehydereductase homolog YJR096w - yeast (Sa
Contig107_m5d10a1.f1	335	1.2e-29	188 856	sp P36091 YKE6_Y	HYPOTHETICAL 49.6 KD PROTEIN IN ELM1-PRI2 INTERGENICREGION >pir s37867 hypothetical protein YKL046c
i8e07a1.r1	335	1.2e-29	55 408	pir s67038	hypothetical protein YOR150w - yeast (Saccharomyces cerevisiae)>gi 1293710 (U55020) O3530p
i0e05a1.r1	334	1.4e-29	11 397	sp P32623 UTR2_YEAST	UTR2 PROTEIN (UNKNOWN TRANSCRIPT 2 PROTEIN)
Contig643_o8f01a1.f1	252	1.4e-29	588 863	pir s51434	hypothetical protein YLR189c - yeast (Saccharomyces

w9c09a1.r1	333	1.8e-29	7 489	cerevisiae)>gi 577215 (U17246) Ylr189cp [Saccharo gnl PID e349610 (Z99262) hypothetical protein [Schizosaccharomyces pombe]
i3c03a1.r1	330	3.8e-29	16 483	sp P43567 YFD0_YEAST HYPOTHETICAL 41.9 KD PROTEIN IN HAC1-CAK1 INTERGENICREGION >pir S56224 hypothetical prote
g6g08a1.r1	329	5.1e-29	50 694	sp Q10478 YDF6_SCHPO HYPOTHETICAL 51.8 KD PROTEIN C17C9.06 IN CHROMOSOME I>gnl PID e241976 (Z73099) hypothetical protein [Schizosaccharomyces pombe]
f0g01a1.r1	337	5.4e-29	10 606	sp Q09850 YAEA_SCHPO HYPOTHETICAL 77.9 KD PROTEIN C23D3.10C IN CHROMOSOME I>pir S62501 hypothetical protein SP
i3h07a1.r1	336	6.9e-29	29 466	sp O13965 YE45_SCHPO HYPOTHETICAL 79.3 KD PROTEIN C24C9.05C IN CHROMOSOME I>gnl PID e334374 (Z98601) hypothetical protein
f5c03a1.f1	325	1.2e-28	100 498	gnl PID d1018329 (D90907) hypothetical protein [Synechocystis sp.]
v3b07a1.r1	324	1.6e-28	149 514	sp P53219 YG1L_YEAST HYPOTHETICAL 38.5 KD PROTEIN IN ERV1-GLS2 INTERGENICREGION >pir S64322 probable membrane protein YDR109c - yeast
Contig1338_gld12a1.r1	330	3e-28	406 750	pir S52675 (Saccharomycescerevisiae) >gi 747884 (Z48758) unknown [Sacc charomyces]
Contig985_j9f11a1.f1	320	4.5e-28	397 774	gnl PID e1263895 (AL022103) hypothetical protein [Schizosaccharomyces pombe]
j7c01a1.r1	329	6.7e-28	38 289	gnl PID e1292820 (AJ005963) 100 kDa protein [Ajellomyces capsulatus]
r5d12a1.r1	316	1.1e-27	199 537	gnl PID e339959 (Z98980) transcription factor [Schizosaccharomyces pombe]
q0d09a1.r1	315	1.5e-27	47 379	sp Q03691 YM56_YEAST HYPOTHETICAL 28.9 KD PROTEIN IN CLN1-RAD14 INTERGENICREGION
Contig611_c3a12a1.f1	324	1.6e-27	125 544	sp P97739 ECE1_C ENDOTHELIN-CONVERTING ENZYME 1 (ECE-1) >bbs 178962(S82653) endothelin converting enzyme, ECE [guinea pig]
c3e08a1.r1	313	2.6e-27	53 613	sp P75791 YBIU_ECOLI HYPOTHETICAL 47.3 KD PROTEIN IN OMPX-MOEB INTERGENICREGION >gi 1787042 (AE000184) f421; Th
Contig605_c3g12a1.r1	312	2.9e-27	141 641	sp P38260 YBVI_Y HYPOTHETICAL 32.6 KD PROTEIN IN VPS15-YMC2 INTERGENICREGION >pir S48266 hypothetical protein YBR101c (X69881) ORF2 [Saccharomyces cerevisiae]
Contig145_j5a05a1.f1	316	5.5e-27	220 684	gi 4088
c5h08a1.r1	317	7.1e-27	90 572	sp P39992 YEC3_YEAST HYPOTHETICAL 78.3 KD PROTEIN IN RIP1-URA3 INTERGENICREGION >pir S50436 hypothetical prote
Contig457_d4d11a1.f1	309	1.1e-26	50 604	sp Q10478 YDF6_S HYPOTHETICAL 51.8 KD PROTEIN C17C9.06 IN CHROMOSOME I>gnl PID e241976 (Z73099) hypothetical protein [Schizosaccharomyces pombe]
z3a10a1.r1	201	1.1e-26	9 326	sp P53326 YG5L_YEAST HYPOTHETICAL 81.2 KD PROTEIN IN MES1-FOL2 INTERGENICREGION >pir S64599 probable membrane protein YDR284c - yeast
Contig1778_alg03c9.r1	312	1.2e-26	480 956	pir S70114 (Saccharomycescerevisiae) >gi 1332640 (U51031) Ydr284cp [Sa ccharomyces]
o6e11a1.r1	303	2.8e-26	70 531	gnl PID e1295796 (AL023776) hypothetical protein [Schizosaccharomyces pombe]
Contig114_l3a06a1.r1	302	3.1e-26	34 435	gnl PID e332207 (AJ000977) hypothetical protein [Rhodobacter sphaeroides]
Contig211_i0g12a1.r1	302	3.3e-26	18 455	gi 2266911 (AE001274) L4171.5 [Leishmania major]

Contig1309_j7h09a1.f1	301	4.5e-26	344 598	gi 2707187	(U94183) unknown [Glomerella cingulata]
Contig1200_g9g02a1.f1	310	6.5e-26	101 508	sp Q04500 YMJ3_Y	HYPOTHETICAL 103.0 KD PROTEIN IN RAD10-PRS4
Contig503_c9g10a1.f1	299	7.2e-26	197 649	gnl PID e236467	INTERGENICREGION >pir S49634 hypothetical protein YML093 (Z71178) B0024.12 [Caenorhabditis elegans]
Contig994_j9a1a1.f1	299	7.4e-26	498 794	gi 3094014	(AF060862) unknown [Homo sapiens]
p0f10a1.r1	302	8.1e-26	11 466	sp P38821 YHR3_YEAST	HYPOTHETICAL 54.2 KD PROTEIN IN CDC12-ORC6
Contig1598_mlf01a1.r1	297	1.1e-25	515 1165	gnl PID e321532	INTERGENICREGION >pir S48955 hypothetical prot (Y13635) Vip1 protein [Schizosaccharomyces pombe]>gnl PID e1202248 (AL009197) hypothetical protein[Sc
Contig563_c5h03a1.f1	304	2.8e-25	438 656	gnl PID d1022254	(AB004535) hypothetical protein YPR112c [Schizosaccharomyces pombe]
n3c09a1.r1	292	4.1e-25	101 325	gnl PID e275716	(Z81071) F28F8.3 [Caenorhabditis elegans]
v3a12a1.r1	291	5e-25	5 328	gnl PID e323034	(Z97052) hypothetical protein [Schizosaccharomyces pombe]
r7f02a1.r1	303	5.3e-25	232 744	gi 1197061	(L36344) ORF; putative [Saccharomyces cerevisiae]
Contig1407_c4h07a1.r1	296	5.4e-25	9 533	sp P53189 YGC8_Y	HYPOTHETICAL 56.4 KD PROTEIN IN RPL32-CWH41
Contig1097_g9g10a1.r1	290	6.4e-25	116 808	gnl PID e1291650	INTERGENICREGION PRECURSOR >pir S64030 probable membrane (AL023290) hypothetical protein [Schizosaccharomyces pombe]
Contig629_c1b03a1.f1	300	6.8e-25	246 725	gnl PID e1313487	(AL031154) hypothetical protein [Schizosaccharomyces pombe]
z6a10a1.r1	290	1.5e-24	4 417	sp P53285 YG3H_YEAST	HYPOTHETICAL 54.5 KD PROTEIN IN CBF2-SKN1
10f05a1.f1	285	1.9e-24	103 474	pir S61978	INTERGENICREGION >pir S64450 probable membrane hypothetical protein YPL088w - yeast (Saccharomyces cerevisiae)>gi 1151238 (U43281) Lpg20p
Contig1236_g9d02a1.f1	285	2.3e-24	3 497	sp P38278 YBZ1_Y	HYPOTHETICAL 38.5 KD PROTEIN IN IRA1-MAK5
n0f01a1.f1	285	2.3e-24	181 450	gnl PID e1285101	INTERGENICREGION >pir S46010 hypothetical protein YBR141c (AJ002026) rAsp f 13 [Aspergillus fumigatus]=human allergen
g7h07a1.r1	284	3.1e-24	222 686	sp Q04179 YD40_YEAST	HYPOTHETICAL 42.3 KD PROTEIN IN YTA2-DIT1
Contig613_c2f06a1.f1	191	3.5e-24	149 400	pir S59765	INTERGENICREGION >pir S69683 hypothetical prote hypothetical protein YPR100w - yeast (Saccharomyces cerevisiae)>gi 914971 (U32445) Note that there is
v3e02a1.r1	282	4.8e-24	303 566	gnl PID e331452	(Z98056) hyypothetical protein [Schizosaccharomyces pombe]
Contig1342_d4c01a1.f1	281	5.9e-24	354 863	pir S67183	hypothetical protein YOR281c - yeast (Saccharomyces cerevisiae)>gnl PID e189406 (X89633) hypothetical
Contig262_a0e01a1.f1	281	6.1e-24	172 573	sp Q40784 AAPC_P	POSSIBLE APOSPORY-ASSOCIATED PROTEIN C
o8e11a1.f1	288	1.7e-23	10 312	>gi 549984(U13148)	possible apospory-associated protein [Penni
m5e02a1.r1	275	2.7e-23	27 482	sp O14053 YEB4_SCHPO	HYPOTHETICAL 100.6 KD TRP-ASP REPEATS CONTAINING
Contig827_y4b09a1.f1	279	4.3e-23	209 631	gnl PID d1025722	PROTEINC2C6.04C IN CHROMOSOME I >gnl PID (AB010900) YNL123w homolog [Schizosaccharomyces pombe]
				gi 1067085	(Z47357) ZK1128.1 [Caenorhabditis elegans]

m7b03a1.r1	273	4.3e-23	38 364	pir s61970	hypothetical protein YPL096w - yeast (Saccharomyces cerevisiae)>gi 1151230 (U43281) Lpg12p
k9d02a1.f1	272	5.4e-23	287 574	sp P39519 YBDF_YEAST	HYPOTHETICAL PROTEIN IN BDF1 5'REGION (ORF1)
13a06a1.f1	272	5.5e-23	249 521	>gi 547573(Z18944) ORF1	[Saccharomyces cerevi
Contig525_c8c02a1.f1	280	6.4e-23	215 1033	sp P36156 YK56_YEAST	HYPOTHETICAL 43.3 KD PROTEIN IN SIS2-MTD1 INTERGENICREGION >pir s38153 hypothetical prote
q0e02a1.r1	283	6.7e-23	27 521	gnl PID e1251101	(AL021838) hypothetical protein [Schizosaccharomyces pombe]
w8g01a1.f1	278	9.1e-23	118 405	gnl PID d1010417	(D63484) The KIAA0150 gene product is novel. [Homo sapiens]
o6h03a1.r1	268	1.4e-22	20 547	gi 736313	(Z48756) unknown [Saccharomyces cerevisiae]
h1a09a1.f1	267	1.8e-22	198 626	sp P40402 YZEC_BACSU	HYPOTHETICAL 41.8 KD PROTEIN (ORFM)
r1d10a1.r1	266	2.2e-22	338 661	sp P47095 YJZ4_YEAST	HYPOTHETICAL 27.4 KD PROTEIN IN MER2-CPR7 INTERGENICREGION >pir s57042 hypothetical prote
d5b06a1.r1	266	2.3e-22	39 338	gi 2408032	(Z99162) hypothetical protein [Schizosaccharomyces pombe]
h1d10a1.f1	265	7.5e-22	1 546	gnl PID e1216801	(AL021046) SPAC3G9.15c; len:230aa; similarity: to YLR051C,Q12035, unclassified protein, (2
m2f08a1.f1	272	8.9e-22	98 484	sp P35728 YKF9_YEAST	HYPOTHETICAL 49.6 KD PROTEIN IN FBA1-TOA2 INTERGENICREGION >pir s37881 hypothetical prote
Contig1253_g7a02a1.r1	260	8.9e-22	70 303	gi 1019710	(L47993) ORF YJR091c [Saccharomyces cerevisiae]
13e10a1.r1	164	9.3e-22	34 309	pir s72314	hypothetical protein YHR004c-a - yeast (Saccharomyces cerevisiae)>gnl PID e273884 (Z80875) Mrs11p [Sa
z5a06a1.r1	272	9.9e-22	248 577	pir s61140	probable membrane protein YPR156c - yeast (Saccharomyces cerevisiae) >gi 849164 (U28371) si
c0h04a1.r1	266	1e-21	195 476	sp Q09764 YA7B_SCHPO	HYPOTHETICAL 107.1 KD PROTEIN C24H6.11C IN CHROMOSOME I>pir s62413 hypothetical protein 8
p0e05a1.r1	278	1.2e-21	208 501	sp Q04991 YM68_YEAST	HYPOTHETICAL 56.2 KD PROTEIN IN ERG8-UBP8 INTERGENICREGION >pir s57589 probable membrane
w4b10a1.r1	275	1.2e-21	11 520	sp Q10064 YAMB_SCHPO	HYPOTHETICAL 420.8 KD PROTEIN C1F5.11C IN CHROMOSOME I>gi 1103738 (Z68136) unknown [Schizo
y4c07a1.r1	258	1.7e-21	99 476	sp P38737 YHD0_YEAST	HYPOTHETICAL 210.4 KD PROTEIN IN GUT1-RIM1 INTERGENICREGION >pir s48938 hypothetical prot
Contig208_j7f01a1.f1	257	2.1e-21	136 465	sp Q09839 YADE_SCHPO	HYPOTHETICAL 40.0 KD PROTEIN C4G8.14C IN CHROMOSOME I>pir s62491 hypothetical protein SPA
Contig809_r8d09a1.r1	270	3.6e-21	287 541	sp P87132 YDM1_S	HYPOTHETICAL PROTEIN C57A7.01 IN CHROMOSOME I>gnl PID e316110 (Z95396) unknown [Schizosaccharomyces p
s9e01a1.r1	260	3.6e-21	118 597	gnl PID e1314595	(AL031187) putative protein [Arabidopsis thaliana]
Contig1380_d5f07a1.r1	257	5e-21	271 837	sp Q04371 YMR7_YEAST	HYPOTHETICAL 54.1 KD PROTEIN IN PEX12-TAP42 INTERGENICREGION >pir s54029 hypothetical pro
x7g05a1.r1	249	1.5e-20	90 599	sp P40087 DDI1_Y	DNA-DAMAGE INDUCIBLE PROTEIN DDI1 >pir s50646hypothetical protein YER143w - yeast (Saccharomyces cer
				pir s66832	hypothetical protein YOL135c - yeast (Saccharomyces

y6h11a1.r1	254	1.6e-20	181 558	cerevisiae>gnl PID e252305 (Z74877) O sp Q09844 YAE3_SCHPO HYPOTHETICAL 54.3 KD PROTEIN C23D3.03C IN CHROMOSOME I>pir S62494 hypothetical protein SP
r7d03a1.r1	255	1.9e-20	56 679	gi 1707074 (U80450) M01E11.2 [Caenorhabditis elegans]
Contig201_i2g01a1.r1	258	3.4e-20	10 348	pir S69079 hypothetical protein YPR097w - yeast (Saccharomyces cerevisiae)>gi 1230699 (U51033) P9513.1 gene prod
Contig699_t2g12a1.r1	247	4e-20	176 622	gnl PID e349695 (Z99296) hypothetical protein [Schizosaccharomyces pombe]
Contig41_c4d04a1.r1	154	7.9e-20	271 408	pir S72314 hypothetical protein YHR004c-a - yeast (Saccharomyces cerevisiae)>gnl PID e273884 (Z80875) Mrs11p [Sa
r7g09a1.r1	240	1.3e-19	217 651	gi 927403 (Z50177) F46G10.3 [Caenorhabditis elegans]
Contig658_b0d01a1.f1	239	1.6e-19	132 395	gi 832882 (L42454) EF-hand protein [Schizosaccharomyces pombe]
Contig135_j9d07a1.f1	238	2.1e-19	289 552	sp o13868 YE12_s HYPOTHETICAL 11.8 KD PROTEIN C1B3.02C IN CHROMOSOME I>gnl PID e334341 (Z98598) hypothetical protein [
c4h05a1.r1	249	2.4e-19	263 550	gnl PID e1292820 (AJ005963) 100 kDa protein [Ajellomyces capsulatus]
d3a01a1.r1	244	4.3e-19	82 642	sp P53962 YND5_YEAST HYPOTHETICAL 43.8 KD PROTEIN IN NCE3-HHT2 INTERGENICREGION >pir S62957 hypothetical prote
Contig623_c1f06a1.f1	239	7.5e-19	269 577	pir S61029 hypothetical protein YPL235w - yeast (Saccharomyces cerevisiae)>gi 1061254 (Z67751) putative protein
Contig995_c9c03a1.f1	243	1.2e-18	69 536	pir S55965 probable membrane protein YLR409c - yeast (Saccharomycescerevisiae) >gi 625119 (U19729) Ylr409cp [Sac
Contig306_g6f04a1.f1	230	1.5e-18	13 426	pir S67607 probable membrane protein YDL072c - yeast (Saccharomycescerevisiae) >gnl PID e253013 (Z74120) ORF YDL
Contig1026_n3g10a1.r1	229	2.1e-18	101 316	gnl PID d1014541 (D89182) similar to Saccharomyces cerevisiae Lpg10p, SWISS-PROT Accession Number U43281 [Schizosacchar
z7g05a1.f1	228	2.3e-18	232 540	sp P38068 YBM4_YEAST HYPOTHETICAL 22.6 KD PROTEIN IN IPP1-TTP1 INTERGENICREGION >pir S45869 glutaredoxin homol
rla10a1.f1	228	2.9e-18	53 424	sp P53839 YN14_YEAST HYPOTHETICAL 38.8 KD PROTEIN IN MET2-SEC2 INTERGENICREGION >pir S63248 hypothetical prote
c6f08a1.r1	227	3.3e-18	65 277	gnl PID e213997 (Z68218) K01H12.1 [Caenorhabditis elegans]
Contig609_c3f10a1.f1	235	6e-18	194 697	sp Q09782 YA93_s HYPOTHETICAL 85.7 KD PROTEIN C13G6.03 IN CHROMOSOME I>pir S62432 hypothetical protein SPAC13G6.3 - f
Contig1522_d3b06a1.f1	224	6.9e-18	563 949	sp P40513 MA33_Y MITOCHONDRIAL ACIDIC PROTEIN MAM33 PRECURSOR >pir S48409hypothetical protein YIL070c - yeast (Saccha
Contig79_l3c11a1.r1	231	9.3e-18	107 532	sp Q09799 YAA5_s HYPOTHETICAL 69.5 KD PROTEIN C22G7.05 IN CHROMOSOME I>pir S62449 hypothetical protein SPAC22G7.05 -
Contig1746_c7g11a1.f1	222	1e-17	431 619	pir S66926 hypothetical protein YOR052c - yeast (Saccharomyces cerevisiae)>gnl PID e251973 (Z74960) ORF YOR052c
o6c10a1.r1	222	1e-17	10 408	sp Q07651 YD22_YEAST HYPOTHETICAL 34.1 KD PROTEIN IN CDC13-GCS1 INTERGENICREGION >pir S67785 probable membrane
j0h11a1.f1	222	1.1e-17	105 446	gnl PID d1019435 (D90916) hypothetical protein [Synechocystis sp.]
Contig891_w9e07a1.f1	219	2.2e-17	301 507	sp o14056 YEB7_s HYPOTHETICAL 11.3 KD PROTEIN C2C6.07 IN CHROMOSOME I>gnl PID e339194 (Z98887) hypothetical protein [S

Contig1694_dlc06a1.fl	222	2.5e-17	10 489	gnl PID e1293607	(AL023635) hypothetical protein MLCB1243.36 [Mycobacteriumleprae]
g3e01a1.r1	226	6.3e-17	25 522	gnl PID d1019541	(D90917) hypothetical protein [Synechocystis sp.]
Contig491_dif11a1.r1	210	1.9e-16	305 568	pir S54502	probable membrane protein YPR028w - yeast (Saccharomycescerevisiae) >gi 809593 (Z49274) unknown [Sacc
gld07a1.fl	212	2e-16	71 469	gnl PID e293680	(Z84498) hypothetical protein Rv1928c [Mycobacteriumtuberculosis]
Contig368_g2d09a1.fl	212	2.2e-16	364 822	gnl PID e1132792	(Z99163) hypothetical protein [Schizosaccharomyces pombe]
Contig845_r7g08a1.fl	208	3e-16	31 549	sp P54168 YPGQ_B	HYPOTHETICAL 23.1 KD PROTEIN IN BSAA-ILVD INTERGENICREGION >gi 1256633 (L77246) putative [Bacillus su
Contig454_n8h01a1.fl	208	3.4e-16	313 570	gi 949850	(Z50795) R166.3 [Caenorhabditis elegans]
m6d09a1.fl	221	3.6e-16	94 576	pir S65157	hypothetical protein YPL146c - yeast (Saccharomyces cerevisiae) >gi 1244771 (U43703) Lpi2p
u4h11a1.r1	219	4.4e-16	67 507	sp Q09731 YA4E_SCHPO	HYPOTHETICAL 107.3 KD TRP-ASP REPEATS CONTAINING PROTEINC31A2.14 IN CHROMOSOME I >pir S58
Contig1683_v7b02a1.fl	208	6.7e-16	586 1107	sp P53721 YN89_Y	HYPOTHETICAL 25.3 KD PROTEIN IN TIM23-ARE2 INTERGENICREGION >pir S63349 probable membrane protein YN
r2g07a1.r1	220	6.8e-16	28 426	sp P47045 YJF4_YEAST	HYPOTHETICAL 54.2 KD PROTEIN IN BTN1-PEP8 INTERGENICREGION >pir S56826 hypothetical prote
i0e06a1.fl	206	1.1e-15	255 515	gnl PID e1292638	(AL023534) hypothetical protein [Schizosaccharomyces pombe]
c5h03a1.r1	217	1.2e-15	28 576	gnl PID e1250351	(AL021768) putative protein [Arabidopsis thaliana]
m8h10a1.fl	202	1.2e-15	188 373	sp o01578 YXX3_CAEEL	HYPOTHETICAL 16.3 KD PROTEIN F53F10.3 IN CHROMOSOME I >gi 1943771 (U97191) similar to the r
Contig407_f0c01a1.r1	202	1.3e-15	2 181	sp P34227 YBG4_Y	HYPOTHETICAL 29.5 KD PROTEIN IN SEF1-KIP1 INTERGENICREGION >pir S39825 hypothetical protein YBL064c
o0d01a1.r1	202	1.4e-15	241 450	gnl PID e349291	(Z99281) Y57G11C.13 [Caenorhabditis elegans]
k5b08a1.r1	211	1.8e-15	16 234	sp P53968 YNC7_YEAST	HYPOTHETICAL 76.3 KD ZINC FINGER PROTEIN IN KTR5-UME3INTERGENIC REGION >pir S62939 hypoth
g7a11a1.r1	207	2.4e-15	149 496	pir S66939	hypothetical protein YOR056c - yeast (Saccharomyces cerevisiae) >gnl PID e251975 (Z74964) o
o5a08a1.r1	210	2.6e-15	114 461	sp P53326 YG5L_YEAST	HYPOTHETICAL 81.2 KD PROTEIN IN MES1-FOL2 INTERGENICREGION >pir S64599 probable membrane
j5g10a1.r1	199	3e-15	75 323	gnl PID d1018464	(D90908) hypothetical protein [Synechocystis sp.]
m8d11a1.fl	198	3.8e-15	239 718	sp Q10433 YDD9_SCHPO	HYPOTHETICAL 24.7 KD PROTEIN C1B9.09C IN CHROMOSOME I >gnl PID e235401 (Z70720) unknown [Sc
Contig386_f4b12a1.r1	219	3.9e-15	131 442	sp P36114 YKZ8_Y	HYPOTHETICAL 81.8 KD PROTEIN IN YPT52-DBP7 INTERGENICREGION >pir S38077 hypothetical protein YKR018c
clg08a1.r2	227	5.2e-15	36 353	gnl PID e315881	(Z95396) unknown [Schizosaccharomyces pombe]
Contig1554_c7c05a1.fl	204	5.7e-15	557 796	gnl PID e1251086	(AL021837) hypothetical protein [Schizosaccharomyces pombe]
c2f07a1.fl	195	7.6e-15	144 410	gi 809578	(Z49273) unknown [Saccharomyces cerevisiae]

f5c08a1.r1	205	1e-14	18 263	gnl PID e1250023 pombe]	(AL021747) hypothetical protein [Schizosaccharomyces
Contig1108_g9e08a1.r1	194	1e-14	55 234	gnl PID e275630	(Z81038) C25A1.6 [Caenorhabditis elegans]
g4f12a1.r1	193	1.2e-14	311 586	gi 3378330	(AF079317) unknown [Sphingomonas aromaticivorans]
Contig408_e9h07a1.r1	200	1.4e-14	11 568	sp Q10367 YDBH_S I>gi 1220292 (Z70043)	HYPOTHETICAL 53.0 KD PROTEIN C22E12.17C IN CHROMOSOME unknown [Schizosaccharomy
Contig1311_o4b12a1.f1	196	1.5e-14	198 596	pir S61991 cerevisiae)>gi 1151003	hypothetical protein YOR007c - yeast (Saccharomyces (U43491) hypothetical prot
x1c08a1.r1	199	1.6e-14	220 630	gi 2622063 thermoautotrophicum]	(AE000870) conserved protein [Methanobacterium
r1d02a1.r1	201	2.6e-14	30 260	gnl PID e339911 pombe]	(Z98974) hypothetical protein [Schizosaccharomyces
15a05a1.f1	200	2.6e-14	287 529	gnl PID e1251101 pombe]	(AL021838) hypothetical protein [Schizosaccharomyces
Contig1474_h4a03a1.f1	199	2.6e-14	357 713	sp P40055 YER2_Y INTERGENICREGION >pir	HYPOTHETICAL 62.3 KD PROTEIN IN PTP3-ILV1 S50585 hypothetical protein YER082c
k0g06a1.r1	199	2.8e-14	5 385	gnl PID e1295804 pombe]	(AL023777) hypothetical protein [Schizosaccharomyces
m0e09a1.f1	193	3.6e-14	183 425	gnl PID e241985	(Z73100) unknown [Schizosaccharomyces pombe]
Contig578_c5a08a1.f1	207	4e-14	864 1136	gi 1658377	(U69170) unknown [Pichia pastoris]
Contig1078_j4f01a1.f1	188	4e-14	624 800	gnl PID e1256502 pombe]	(AL022072) hypothetical protein [Schizosaccharomyces
Contig1445_h8a07a1.f1	190	4.8e-14	465 767	pir S66764 cerevisiae)>gnl PID e251875 (Z74813)	hypothetical protein YOL071w - yeast (Saccharomyces ORF YOL071w
Contig670_alb05c9.r1	191	5.3e-14	297 524	sp P53290 YG3T_Y INTERGENICREGION >pir	HYPOTHETICAL 38.6 KD PROTEIN IN TIF4631-KRE11 S64474 probable membrane protein
x3e04a1.f1	194	5.4e-14	266 580	gi 2689890 burgdorferi]	(AE000792) conserved hypothetical protein [Borrelia
f1d11a1.r1	195	5.6e-14	18 410	gi 1173491	(U20390) ORF494 [Saccharomyces cerevisiae]
y6g07a1.r1	186	6.5e-14	13 450	sp Q04272 YMW7_YEAST INTERGENICREGION >pir	HYPOTHETICAL 25.6 KD PROTEIN IN ABF2-CHL12 S54452 hypothetical prot
r2c05a1.r1	209	9e-14	21 623	gnl PID e280810 Pristinamycin I synthase of Streptomyces	(Z82015) yukK [Bacillus subtilis]-similar to
g7b05a1.r1	192	1.2e-13	149 772	pir S53401 (Saccharomycescerevisiae) >gi 662138 (U20618)	probable membrane protein YLR324w - yeast Y1
Contig1787_c0c10a1.r1	193	1.3e-13	98 259	gnl PID e1188370	(Z82286) W02A2.g [Caenorhabditis elegans]
Contig1861_c4g05a1.f1	193	1.3e-13	394 555	gnl PID e1188370	(Z82286) W02A2.g [Caenorhabditis elegans]
e7c01a1.r1	205	1.4e-13	131 610	gnl PID e334014 pombe]	(Z98529) hypothetical protein [Schizosaccharomyces
g5b08a1.r1	203	1.4e-13	113 700	gnl PID e1315376 pombe]	(AL031261) hypothetical protein [Schizosaccharomyces
m7c06a1.r1	185	1.5e-13	274 609	sp P53337 YG5Y_YEAST INTERGENICREGION >pir	HYPOTHETICAL 35.0 KD PROTEIN IN BGL2-ZUO1 S64619 probable membrane

m0h07a1.r1	189	1.9e-13	144 446	sp O13909 YDW1_SCHPO HYPOTHETICAL 49.2 KD PROTEIN C23C11.01 IN CHROMOSOME I>gnl PID e334129 (Z98559) SPAC23C11.
i3e06a1.f1	188	2e-13	215 466	gnl PID d1032703 (AB016218) unknown: similar to human GA17 protein [Schizosaccharomyces pombe]
Contig1671_f5a07a1.r1	210	2.7e-13	726 1469	gnl PID e1292632 (AL023534) hypothetical protein [Schizosaccharomyces pombe]
g3b10a1.r1	136	2.9e-13	38 322	gi 2315350 (AF016439) No definition line found [Caenorhabditis elegans]
r5a04a1.r1	194	3.3e-13	96 407	sp P40531 YIE1_YEAST 36.7 KD PROTEIN IN CBR5-NOT3 INTERGENIC REGION>pir S49937 hypothetical protein YIL041w -
j0h09a1.r1	189	3.4e-13	20 328	sp P25351 YCR3_YEAST HYPOTHETICAL 69.2 KD PROTEIN IN HSP30-PMP1 INTERGENICREGION >pir S19434 probable transpor
Contig678_o9d08a1.r1	192	3.8e-13	302 610	sp P47029 YJI4_Y HYPOTHETICAL 117.2 KD PROTEIN IN EXO70-ARP4 INTERGENICREGION >gi 895905 (X88851) hypothetical protein
m6e06a1.r1	212	4.5e-13	48 404	pir S59841 probable membrane protein YPR184w - yeast (Saccharomycescerevisiae) >gi 786314 (U25842) Hi
z6g01a1.f1	178	4.8e-13	309 557	gnl PID e1198272 (Z99165) hypothetical protein [Schizosaccharomyces pombe]
y7g04a1.f1	202	5.7e-13	294 650	gnl PID e237905 (X97346) FCYX gene product [Saccharomyces cerevisiae]>gi 1381130 (U18813) Fcy22p: Puring-c
Contig1429_g6f08a1.f1	175	1e-12	184 423	sp P47111 YJ14_Y HYPOTHETICAL 15.7 KD PROTEIN IN NUP85-SSC1 INTERGENICREGION >pir S57063 probable membrane protein YJ
w4a10a1.f1	182	1.3e-12	109 507	sp Q09817 YAC3_SCHPO HYPOTHETICAL 56.6 KD PROTEIN C16C9.03 IN CHROMOSOME I>pir S62473 hypothetical protein SPA
Contig1643_o4g06a1.r1	173	1.6e-12	248 547	pir S67201 hypothetical protein YOR297c - yeast (Saccharomyces cerevisiae)>gnl PID e252135 (Z75205) ORF YOR297c
Contig446_d5d06a1.f1	184	1.9e-12	273 494	gnl PID e351296 (Z99532) hypothetical protein [Schizosaccharomyces pombe]
Contig1225_g3e01a1.f1	196	2e-12	315 686	gnl PID d1019541 (D90917) hypothetical protein [Synechocystis sp.]
Contig1044_s9f01a1.f1	183	2.1e-12	483 662	pir S37694 gene PC326 protein - mouse >gi 200241 (M95564) protein PC326 [Musmusculus]
j7c01a1.f1	183	2.8e-12	238 669	gnl PID e1292820 (AJ005963) 100 kDa protein [Ajellomyces capsulatus]
y6h06a1.r1	170	3.6e-12	85 276	gi 1350548 (L47609) heat shock-like protein [Picea glauca]
a0e01a1.r1	172	3.9e-12	158 406	sp Q03161 YMY9_YEAST HYPOTHETICAL 34.0 KD PROTEIN IN CTF13-YPK2 INTERGENICREGION >pir S55085 hypothetical prot
Contig724_u4g06a1.r1	169	4.4e-12	14 292	pir S59397 probable membrane protein YLR251w - yeast (Saccharomycescerevisiae) >gi 662333 (U20865) Ylr251wp [Sac
Contig1081_g9g02a1.r1	181	4.6e-12	5 415	sp Q04500 YMJ3_Y HYPOTHETICAL 103.0 KD PROTEIN IN RAD10-PRS4 INTERGENICREGION >pir S49634 hypothetical protein YML093
g6a10a1.f1	183	5.6e-12	294 548	gnl PID e1292587 (AL023518) hypothetical protein [Schizosaccharomyces pombe]
f2b12a1.r1	193	5.8e-12	95 490	gnl PID e1294546 (AL023706) hypothetical protein [Schizosaccharomyces pombe]

i0h03a1.r1	174	8.1e-12	111 449	gnl PID e310346	(Z93386) R11H6.2 [Caenorhabditis elegans]
Contig876_w4a08a1.f1	166	9.5e-12	270 596	gnl PID e324205	(Z97185) hypothetical protein [Schizosaccharomyces pombe]
Contig1189_h0b12a1.f1	177	1.1e-11	51 557	sp Q09686 YA14_S	HYPOTHETICAL 28.0 KD PROTEIN C13C5.04 IN CHROMOSOME
Contig663_a5e09a1.f1	171	1.2e-11	237 506	I>pir S58096	hypothetical protein SPAC13C5.04 -
s8a07a1.f1	183	1.4e-11	352 561	gnl PID e312773	(Z72840) ORF YGR054w [Saccharomyces cerevisiae]
k0f02a1.r1	163	3e-11	176 400	gnl PID e1285386	(AL022304) hypothetical protein [Schizosaccharomyces pombe]
Contig344_g3h06a1.f1	161	3e-11	275 550	sp P38838 YHT4_YEAST	HYPOTHETICAL 30.6 KD PROTEIN IN ACT5-YCK1 INTERGENICREGION >pir S48978
c6h09a1.r1	184	3.1e-11	98 436	sp P40156 YNV3_Y	HYPOTHETICAL 25.3 KD PROTEIN IN PEX17-MER1 INTERGENICREGION >pir S50718
h4c07a1.r1	171	3.1e-11	33 425	gnl PID e339146	hypothetical protein YNL213c (Z98849) hypothetical protein [Schizosaccharomyces pombe]
Contig641_c0c03a1.r1	172	5.1e-11	76 342	sp P38731 YHE0_YEAST	HYPOTHETICAL 70.9 KD PROTEIN IN CBP2 5'REGION>pir S48928
Contig1473_x5f05a1.r1	186	5.8e-11	200 820	gi 2149640	hypothetical protein YHL040c - y (U91995) Argonaute protein [Arabidopsis thaliana]
Contig695_o9h08a1.f1	158	6.1e-11	110 298	sp P38724 YHE7_Y	HYPOTHETICAL 71.6 KD PROTEIN IN CBP2 5'REGION>pir S48921
h0g07a1.r1	160	6.3e-11	38 376	gi 1397277	hypothetical protein YHL047c - yeast (Sacch (U61947) C06G3.11 gene product [Caenorhabditis elegans])
Contig388_f2d10a1.f1	164	7.7e-11	102 419	gi 2384956	(AF022985) No definition line found [Caenorhabditis elegans]
g7e08a1.r1	159	8.5e-11	355 711	gi 666912	(M93129) [Mycobacterium tuberculosis DNA sequence, complete cds.], gene products [Mycobacterium tuberc
Contig1443_d4b07a1.f1	187	8.6e-11	442 1059	gi 3128287	(AF010496) hypothetical protein [Rhodobacter capsulatus]
Contig775_x5a07a1.f1	160	9.6e-11	368 619	sp Q10327 YD72_S	HYPOTHETICAL 97.1 KD PROTEIN C32A11.02C IN CHROMOSOME
n8c01a1.r1	180	1e-10	21 326	I>gi 1213266	(Z69796) unknown [Schizosaccharomy
u4d06a1.f1	156	1.1e-10	344 508	gnl PID e123998	(X82490) unnamed protein product [Fusarium oxysporum]
d3d03a1.f1	174	1.5e-10	55 489	sp P48236 YG3L_YEAST	HYPOTHETICAL 51.6 KD PROTEIN IN RPL30B-RSR1 INTERGENICREGION >pir S60440
i3f03a1.r1	160	2e-10	78 458	I>gi 1103730	probable membran (Z68136) unknown [Schizos
m5f02a1.r1	177	2.2e-10	70 531	pir S67175	probable membrane protein YOR273c - yeast (Saccharomycescerevisiae) >gnl PID e189400
m7h11a1.f1	153	2.3e-10	273 461	sp P87151 YBOA_SCHPO	HYPOTHETICAL 20.9 KD PROTEIN C25H2.10C IN CHROMOSOME II>gnl PID e316124
h4a03a1.r1	162	2.4e-10	142 423	sp P40055 YER2_YEAST	(Z95397) unknowns. pombe HYPOTHETICAL 62.3 KD PROTEIN IN PTP3-ILV1 INTERGENICREGION >pir S50585

m3a11a1.r1	177	4.2e-10	124 285	gnl PID e1263973	(AL022117) hypothetical protein [Schizosaccharomyces pombe]
Contig766_v1c09a1.r1	162	5.9e-10	201 449	sp P38163 YBK6_Y	HYPOTHETICAL 111.7 KD PROTEIN IN PKC1
Contig746_p0h09a1.f1	148	7e-10	233 376	5'REGION>pir s45389	probable membrane protein YBL106c - yeast(
n2g05a1.r1	151	1.3e-09	79 318	gi 431953	(X76302) nucleic acid binding protein [Homo sapiens]
e9e04a1.r1	162	1.4e-09	182 526	sp O14256 YE6A_SCHPO	HYPOTHETICAL 22.4 KD PROTEIN C6G10.10C IN CHROMOSOME I>gnl PID e334330 (Z98603) hypothetic
m7c06a1.f1	155	1.5e-09	250 492	gi 3158469	(AF067216) No definition line found [Caenorhabditis elegans]
m0b08a1.r1	151	1.7e-09	12 161	sp Q06567 YL53_YEAST	HYPOTHETICAL 65.9 KD PROTEIN IN SSP120-HAP1 INTERGENICREGION >pir s59398 probable membran
Contig1356_d5f05a1.f1	146	1.9e-09	172 540	sp P38263 YBV5_YEAST	HYPOTHETICAL 41.2 KD PROTEIN IN YMC2-CMD1 INTERGENICREGION >pir s48270 hypothetical prote
13c11a1.f1	153	2.5e-09	231 440	sp O14068 YEA3_S	HYPOTHETICAL 13.9 KD PROTEIN C2E11.03C IN CHROMOSOME I>gnl PID e339159 (Z98850) hypothetical protein
Contig76_l3e08a1.f1	143	2.6e-09	243 467	sp P42846 YN48_YEAST	HYPOTHETICAL 68.7 KD PROTEIN IN STB1-MCK1 INTERGENICREGION >pir s51303 hypothetical prote
Contig1655_e9e11a1.f1	172	3e-09	698 1213	sp Q07549 YD23_Y	HYPOTHETICAL 15.7 KD PROTEIN IN UBP1-HNT1 INTERGENICREGION >pir s67666 probable membrane protein YDL
Contig308_g6e08a1.r1	168	3.7e-09	247 645	pir s61185	hypothetical protein YDR299w - yeast (Saccharomyces cerevisiae)>gi 849214 (U28374) Ydr299wp [Saccharo
n5a02a1.r1	167	3.9e-09	66 509	gi 2558956	(AF025475) Mascl [Ascobolus immersus]
Contig363_m2f12a1.f1	142	4.7e-09	327 590	sp P38787 YHM3_YEAST	HYPOTHETICAL 42.8 KD PROTEIN IN VMA22-RRP3 INTERGENICREGION >pir s46711 hypothetical prot
r4b10a1.f1	166	5.7e-09	327 656	sp Q09730 YA4D_S	HYPOTHETICAL 10.5 KD PROTEIN C31A2.13C IN CHROMOSOME I>pir s59647 hypothetical protein SPAC31A2.13c
j9f03a1.f1	159	7e-09	245 502	gnl PID e223969	(Z69793) R03A10.3 [Caenorhabditis elegans]
Contig696_p0a01a1.f1	139	7e-09	152 370	gnl PID d1022302	(AB004539) pi074 [Schizosaccharomyces pombe]>gnl PID e1250311 (AL021766) hypothetical prot
Contig393_flg11a1.f1	155	8.9e-09	442 681	gi 2621836	(AE000853) conserved protein [Methanobacterium thermoautotrophicum]
Contig277_g9e11a1.f1	153	1.4e-08	67 408	gnl PID d1026035	(AB011825) CSH3 [Schizosaccharomyces pombe]
Contig373_g2a02a1.r1	157	2e-08	72 473	>gnl PID e1263963(AL022117)	hypothetical protein [Schizos
j4h11a1.r1	135	2e-08	149 364	gnl PID e1292630	(AL023534) hypothetical protein [Schizosaccharomyces pombe]
x1g08a1.r1	141	2.2e-08	17 271	gnl PID d1011269	(D64004) hypothetical protein [Synechocystis sp.]
n3g12a1.r1	134	2.3e-08	122 319	sp Q10481 YDF9_SCHPO	HYPOTHETICAL 10.7 KD PROTEIN C17C9.09C IN CHROMOSOME I>gnl PID e241761 (Z73099) hypothetic
g7e07a1.r1	162	2.4e-08	191 427	sp P53938 YNI0_YEAST	HYPOTHETICAL 41.7 KD PROTEIN IN PMS1-TPM1 INTERGENICREGION >pir s53898 probable membrane
				pir s66709	probable membrane protein YOL026c - yeast (Saccharomycescerevisiae) >gnl PID e252264 (Z747
				gi 939724	(U30858) putative sensor kinase; regulatory protein

e0a10a1.f1	142	3.5e-08	182 472	gnl PID e1295823 (AL023780) hypothetical protein [Schizosaccharomyces pombe]
x8f04a1.r1	146	5.1e-08	75 473	sp Q04958 YMF9_YEAST HYPOTHETICAL 187.1 KD PROTEIN IN OGG1-CNA2 INTERGENICREGION >pir S49802 probable membrane
Contig385_n3c12a1.r1	157	5.8e-08	559 813	sp P53219 YG1L_Y HYPOTHETICAL 38.5 KD PROTEIN IN ERV1-GLS2 INTERGENICREGION >pir S64322 probable membrane protein YGR
Contig72_l3g10a1.f1	146	5.9e-08	134 463	gnl PID d1013422 (D85230) hypothetical protein [Plectonema boryanum]
t2f11a1.r1	136	6.6e-08	117 284	sp P35735 YKF1_YEAST HYPOTHETICAL 40.5 KD PROTEIN IN NUP120-CSE4 INTERGENICREGION >pir S37873 hypothetical pro
Contig1343_cld09a1.f1	150	7e-08	157 813	gi 2190955 (AF002247) ORF4; Putative transmembrane protein [Rhodococcuserythropolis]
g8b06a1.f1	155	9.7e-08	318 581	pir S66834 probable membrane protein YOL137w - yeast (Saccharomycescerevisiae) >gnl PID e252306 (Z748
z4a08a1.r1	137	1e-07	184 504	gnl PID d1017526 (D90900) hypothetical protein [Synechocystis sp.]
Contig935_l3a07a1.f1	132	1.5e-07	288 461	sp O13802 YE05_S HYPOTHETICAL 37.8 KD PROTEIN C17H9.05 IN CHROMOSOME I>gnl PID e334275 (Z98597) hypothetical protein [
Contig1444_clf02a1.f1	124	2.7e-07	398 493	pir S12206 hypothetical protein 2 (rRNA external transcribed spacer) - mouse
Contig497_d0c12a1.f1	152	3e-07	349 741	sp Q09844 YAE3_S HYPOTHETICAL 54.3 KD PROTEIN C23D3.03C IN CHROMOSOME I>pir S62494 hypothetical protein SPAC23D3.03c
Contig397_f0g10a1.f1	136	3.1e-07	201 329	gnl PID e340013 (Z98977) hypothetical protein [Schizosaccharomyces pombe]
y9d06a1.r1	128	3.3e-07	177 416	pir S69568 hypothetical protein YDR511w - yeast (Saccharomyces cerevisiae)>gi 927780 (U33057) Ydr511w
g9b06a1.f1	157	3.5e-07	197 562	gnl PID e1314595 (AL031187) putative protein [Arabidopsis thaliana]
Contig897_v7g01a1.f1	154	3.8e-07	618 1025	sp Q10218 YAYB_S HYPOTHETICAL 89.2 KD PROTEIN C4H3.11C IN CHROMOSOME I>gi 1184024 (Z69380) unknown [Schizosaccharomyce
m8a03a1.r1	145	4.2e-07	6 170	sp P42620 YQJG_ECOLI HYPOTHETICAL 37.4 KD PROTEIN IN EKUR-TDCC INTERGENICREGION (O328) >gi 606043 (U18997) ORF_
n8f02a1.f1	130	5.7e-07	168 485	sp O14011 YDP8_SCHPO HYPOTHETICAL 54.2 KD TRP-ASP REPEATS CONTAINING PROTEINC29A4.08C IN CHROMOSOME I >gnl PID
Contig1728_c7g04a1.f1	148	6e-07	283 705	gnl PID e339966 (Z98981) hypothetical protein [Schizosaccharomyces pombe]
i7e09a1.f1	143	9e-07	324 623	sp Q09875 YAGC_SCHPO HYPOTHETICAL 35.8 KD PROTEIN C12G12.12 IN CHROMOSOME I>pir S62543 hypothetical protein SP
Contig1783_c5d01a1.f1	152	1.2e-06	1149 1598	sp P39992 YEC3_Y HYPOTHETICAL 78.3 KD PROTEIN IN RIP1-URA3 INTERGENICREGION >pir S50436 hypothetical protein YEL023c
ald05f2.f1	141	1.3e-06	89 319	gnl PID e1251050 (AL021817) hypothetical protein [Schizosaccharomyces pombe]
Contig1414_m5g11a1.f1	137	1.4e-06	195 500	gnl PID d1031256 (AP000005) 223aa long hypothetical protein [Pyrococcusshorikoshii]
a0e05a1.r1	120	1.5e-06	136 456	sp Q10430 YDD5_SCHPO HYPOTHETICAL 28.8 KD PROTEIN C1B9.05C IN CHROMOSOME

j0a03a1.f1	128	1.7e-06	247 420	I>gnl PID e235479 (Z70720) unknown [Sc sp P38170 YBJ7_YEAST HYPOTHETICAL 83.0 KD PROTEIN IN ATP1-ROX3 INTERGENICREGION >pir s45403 hypothetical prote
Contig1685_j9f07a1.f1	139	2.1e-06	538 816	pir s61199 hypothetical protein YDR313c - yeast (Saccharomyces cerevisiae)>gi 849227 (U28374) YDR313C gene produ
c9d11a1.r1	151	2.2e-06	39 296	gnl PID d1010130 (D50929) The KIAA0139 gene product is related to mousecentrosomin B. [Homo sapiens] >gi 18
w5f12a1.r1	129	2.3e-06	3 236	sp P48231 YNI7_YEAST HYPOTHETICAL 132.5 KD PROTEIN IN TOP2-MKT1 INTERGENICREGION >pir s57535 probable membrane
c5g07a1.r1	142	2.6e-06	8 619	gnl PID e317345 (Z95620) unknown [Schizosaccharomyces pombe]
i0e06a1.r1	121	2.7e-06	274 447	gnl PID e1292638 (AL023534) hypothetical protein [Schizosaccharomyces pombe]
h1a09a1.r1	139	2.8e-06	222 386	sp P47095 YJZ4_YEAST HYPOTHETICAL 27.4 KD PROTEIN IN MER2-CPR7 INTERGENICREGION >pir s57042 hypothetical prote
Contig888_v3h04a1.f1	136	5e-06	419 505	gi 1870215 (AC000133) ORF [Emericella nidulans]
Contig1194_w6g09a1.f1	122	5.9e-06	481 642	gi 940146 (U30501) orf2; Method: conceptual translation supplied by author. [Thermotoga maritima]
n8d09a1.r1	136	6.9e-06	33 158	pir s67247 hypothetical protein YOR338w - yeast (Saccharomyces cerevisiae)>gnl PID e223200 (X95720) O
o8h04a1.r1	127	1.5e-05	84 374	gi 310604 (L19300) ORF3 [Staphylococcus aureus]
a0a09a1.f1	120	2.2e-05	67 444	gnl PID e1295819 (AL023780) hypothetical protein [Schizosaccharomyces pombe]
r4h12a1.r1	128	3.9e-05	76 318	gnl PID e1250039 (AL021748) hypothetical protein [Schizosaccharomyces pombe]
e7d05a1.f1	132	4.7e-05	334 522	pir s66834 probable membrane protein YOL137w - yeast (Saccharomycescerevisiae) >gnl PID e252306 (Z748
Contig1133_j7g02a1.f1	129	5.6e-05	296 556	gi 1109800 (U41528) C15C7.1 gene product [Caenorhabditis elegans]
Contig1706_c8b03a1.f1	124	5.8e-05	509 853	gnl PID e349611 (Z99262) hypothetical protein [Schizosaccharomyces pombe]
l3d09a1.r1	106	6.1e-05	127 279	gnl PID e324207 (Z97185) hypothetical protein [Schizosaccharomyces pombe]
Contig297_g7d08a1.r1	128	0.00011	621 815	sp Q09875 YAGC_S HYPOTHETICAL 35.8 KD PROTEIN C12G12.12 IN CHROMOSOME I>pir s62543 hypothetical protein SPAC12G12.12
w7f03a1.f1	128	0.00011	29 217	pir s67290 probable membrane protein YOR378w - yeast (Saccharomycescerevisiae) >gnl PID e252199 (Z752
t2h07a1.f1	109	0.00011	186 452	sp P53220 YG1M_YEAST HYPOTHETICAL 27.2 KD PROTEIN IN GLS2-RPL26B INTERGENICREGION >pir s64324 probable membran
Contig698_r5a07a1.f1	126	0.00013	694 939	sp P53267 YG2Y_Y HYPOTHETICAL 37.8 KD PROTEIN IN CLB6-SPT6 INTERGENICREGION >pir s64421 hypothetical protein YGR113w
Contig1769_e9g10a1.r1	133	0.00015	395 580	gnl PID e1293563 (AL023634) hypothetical protein [Schizosaccharomyces pombe]
d1b10a1.r1	134	0.00016	43 477	gnl PID e349693 (Z99296) hypothetical protein [Schizosaccharomyces pombe]

Contig280_g9c05a1.f1	122	0.00026	312 500	gnl PID e349324	(Z99264) T05F1A.2 [Caenorhabditis elegans]
				>gnl PID e1247195(Z81586)	T05F1.a [Caenorhabditis elegans]
Contig1164_g2a01a1.f1	118	0.00038	2 373	sp P40526 YIE9_Y	HYPOTHETICAL 30.3 KD PROTEIN IN GPP1-SYG1
				INTERGENICREGION >pir S48430	probable membrane protein YIL

VIII. No significant homolog

<NONE>

-949 Contigs

-1419 Singlets