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Scope and Method of Study: This report is devoted to the compilation of information concerning the discoveries and the potential for future work in biology using radiation sources as a tool. The fact has been stressed that nuclear or x-radiation is by no means the perfect research tool, and must be incorporated with other proven research media for maximun results.

Radioactive isotopes are used in tracer work and has given researchers much information concerning both plant and animal. Radioactive materials have been or will be used for producing chromosomal aberrations, gene mutations, and cellular destruction. Several examples are given in this report.

The present and future accomplishments and/or potentials, and the disadvantages of the uses of radioactive sources in biological research are covered.

ADVISER'S APPROVAL L. Herbert Bruneau

THE USES OF NUCLEAR RADIATION IN THE FIELD OF PROGRESSIVE BIOLOGY

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THE FIELD OF PROGRESSIVE

BIOLOGY

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PREFACE

In this report the term "progressive biology" will be used in reference to the use of any agent or agents to improve the living environment for the benefit of the majority of the human population. Since there are a number of agents capable of altering any particular cell, living or dead, and with the possibility of any agent acting in a variety of ways to produce the same or many different alterations, I have chosen only nuclear radiations in hopes of covering their uses and actions in a manner which befits the expectations of this report.

Nuclear energy is not new, but our knowledge of its presence and comprehension of its effects is relatively new. Our first knowledge of the biological effects of nuclear energy evolved out of its use to destroy life. Now we have hopes of using nuclear energy not only to sustain and improve life, but also to make life more pleasant.

Like all agents, nuclear radiations have their limitations and disadvantages. In the following chapters I will attempt to review the uses, possibilities, limitations, and disadvantages of this powerful agent in the field of progressive biology.

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CHAPTER I

PHYSICAL PROPERITIES OF NUCLEAR RADIATIONS

Nuclear energy evolves from the rearrangements within the interior of an atom.[1] The causes of these rearrangements are beyond the scope of this report. It might be noted that there is a binding force which holds the nucleus of the atom together. Any time there is a deletion or addition to the nucleus of the atom an unbalanced force is established and energy will be given off in some form at some interval of time later. We do not know what controls the time interval involved, however much is known about the forms in which the energy is given off. These forms may be particulate or electromagnetic in nature.

The particles which may be released from a nuclear rearrangement are of many types. Some are easy to detect, some are difficult to detect, some are very common, and some are very rare. This report is concerned with those particles which are common and which are posessed with sufficient energy to cause a rearrangement of cellular constituents. The most common of these radiations are naked helium atoms or alpha particles, naked hydrogen atoms or protons, electrons or beta particles, and neutrons. All of these radiations except the neutron carry an electric charge. The relative energy and penetrating power of each of these particles is dependent on the velocity, the mass, and the

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[1]Samuel Glasstone, Sourcebook on Atomic Energy (2d ed., New York, 1958), p. 63.

charge of that particle. The relative energy and penetrating power determine the overall effect of these particulate radiations upon any tissue.

As one would expect, the neutrons with their lack of charge are not hindered in their progress and are the most penetrating of the nuclear particles under discussion. The kinetic energy of neutrons will vary according to the amount of energy released when they are ejected from an atomic nucleus and according to the number of impacts they have made prior to reaching the locus under discussion. Neutrons have been categorized as either high energy or thermal (slow) neutrons. If a thermal neutron is captured by the nucleus of an atom, the nucleus will be in a high energy excited state which attains relative stability by the ejection of a proton or alpha particle or by emitting the excess energy as gamma radiation. The residual nucleus is not always completely stable, for it is frequently radioactive. The effect of fast neutrons is essentially due to the production of recoil protons.

The proton has the second greatest penetrating power of the particles under discussion. By virtue of its lack of an orbital electron it carries a positive charge. The proton produces ionization[2] along its entire traversity.

The electron or beta particle has shorter penetrance than the proton but produces a more dense ion field than the proton.

The alpha particle has less penetrance than any of the other particles under discussion and is not considered detrimental unless it is released

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[2]The ability to eject an orbital electron by virtue of an energy transfer to the electron.

within the body of an organism. It produces a very dense, very short ion field.

The electromagnetic waves produced, as a result of the rearrangement within the nucleus, are called gamma rays. They are much more penetrating than any of the particles. Their ionization potential is great. Secondary electromagnetic waves may be produced as excited orbital electrons drop back into their stable orbital path. The secondary electromagnetic waves may be of a biological ionizing nature or they may be non-ionizing.

The primary effect of ionization of any atom or molecule in a cell is to place that atom or molecule in a higher electromotive category. This may be beneficial, as in the case of photosynthesis, or harmful as in the case of common sunburn. The tendency of that atom or molecule, in the elevated electromotive category, is to reach some form of stability. In its quest for stability it may destroy the stability of other cellular constituents which in turn act in the same manner, but in a lesser degree, as the original ionized molecule. One can visualize a chain reaction as the excess energy is dissipated throughout the cell.

Each living cell has great power of regeneration. It has been estimated that from one to ten per cent of the protein of a cell must be altered to the point where it is non-functional before any observable change in the cell results.

It has been found that the sensitivity of a cell to radiation is dependent on many factors. The tissue from which these cells are derived, the stage of the life cycle in which irradiation occurs, and environmental factors influence the sensitivity of a cell. This information is diagramatically illustrated on charts 1, 2, 3, and 4.

It will be noted from chart 1a and 1b that the individual mammalian cells are much more sensitive that the protozoans. It is a known fact that at one time the earth was much more radioactive than it is at present. It is felt that there is a strong correlation between the geological age of origin and the degree of radiosensitivity of the organism. Our next assumption would be that radiosensitivity is genetically or chromosomally controlled.

Chromosomal aberrations[3] are much more easily produced in some organisms than in others, the lower forms being the most resistant. As would be guessed, there is a proportionality between the sensitivity of a cell and the number of chromosome breaks produced by a given dose of radiation.

It is felt that the most practicle long time effect of radiation on progressive biology will be gained in the field of genetics. With time, work, and patience, superior breeds of plants and animals could be developed; utilizing all the mutagenic devices at hand. No new mutation has been developed in the laboratory which could not have developed under natural conditions. It must be realized however that nuclear radiation is a natural condition.

Nuclear radiation has three primary modes of action on genetic material. These actions include the production of ionizing materials, the direct breakage of chemical bonds in genes, and chromosome breakage. One of the primary sources of ionizing substances is the direct result

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[3]Chromosomal alteration by loss of some of its component genes, by reduplication, translocation, or inversion.

Chart 1a

Radiation resistance of different animals and microorganisms.

Organism	Radiation dose necessary to kill (LD50)*
Mammalian types	200-1000 r
Guinea-pig	200-400 r
Swine	275 r
Dog	325 r
Goat	350 r
Monkey	500 r
Mouse	400 r
Rat	600-700 r
Hamster	700 r
Rabbit	800 r
Goldfish	700 r
Frog	700 r
Tortoise	1500 r
Newt	3000 r
Snail	10,000 r
Bacterium E. coli	10,000 r
Yeast	30,000 r
Drosophila adults	60,000 r
Amoeba	100,000 r
Paramecium	300,000 r

* The dose required to kill 50 per cent of the animals within 30 days following irradiation.
(from P. Alexander, Atomic Radiation and Life, (Baltimore, Md., 1957),p. 49)

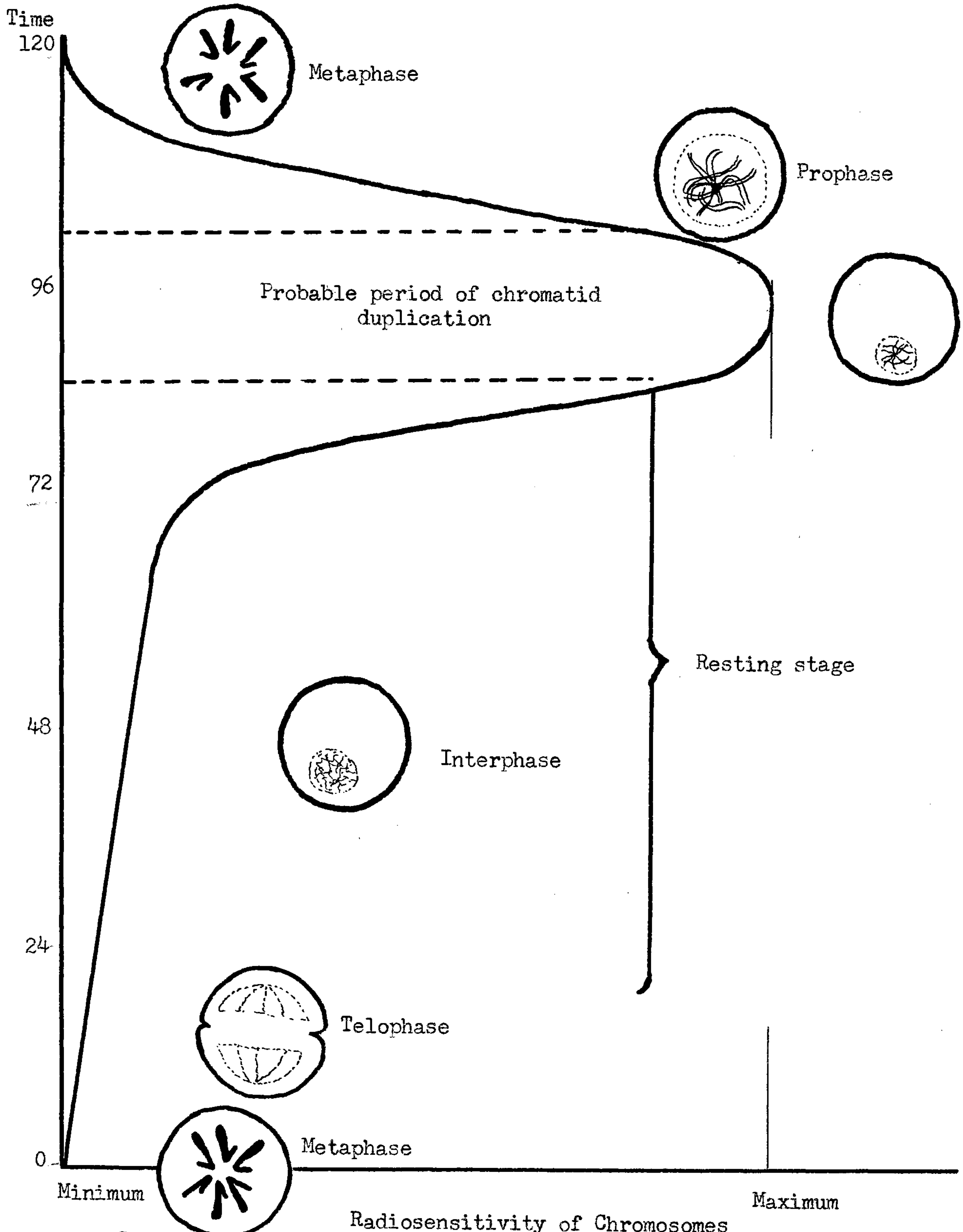
Chart 1b

Radiation resistance of human tissue, arranged in order according to decreasing sensitivity.

Lymphocytes
Granulocytes
Bone Marrow
Epithelium
Endothelium
Connective Tissue
Muscle Cells
Bone Cells
Nerve Cells

No information is currently available on the exact measure of radiosensitivity of the individual tissue types.
(from the lectures of M. Fleck, University of New Mexico, 1958)

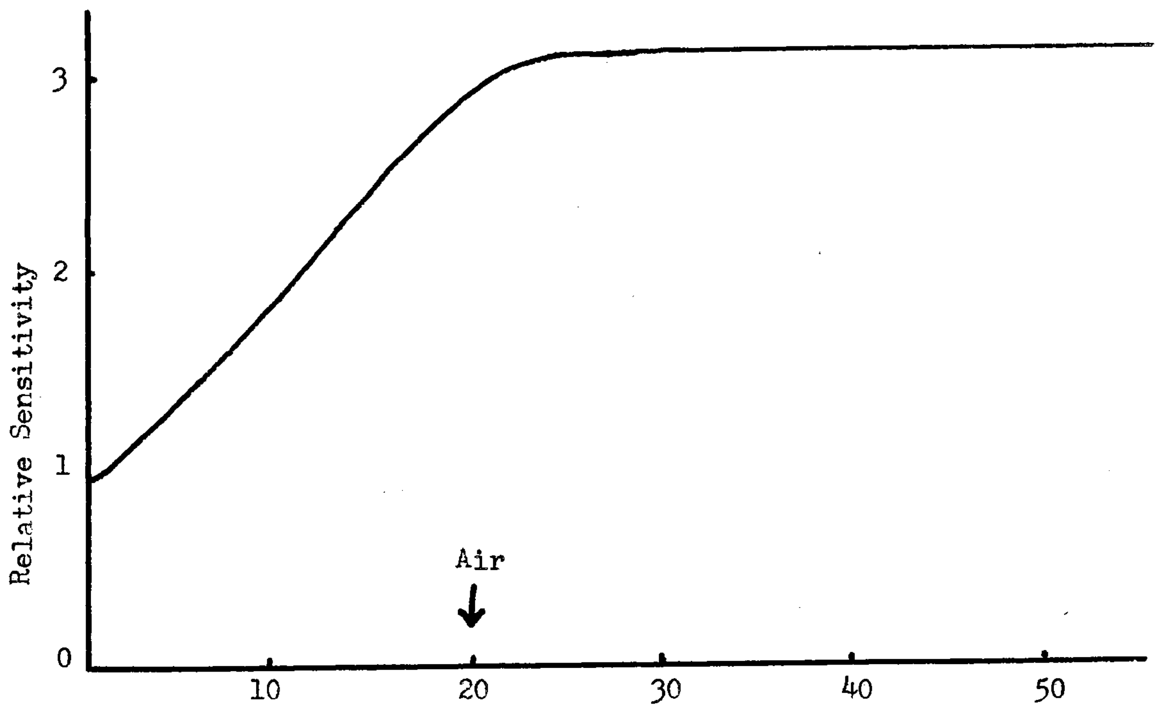
Chart 2



Diagramatic illustration showing the change in sensitivity of the the chromosomes to radiation during the mitotic cycle. Based on experiments with Tradescantia microspores.

(from P. Alexander, Atomic Radiation and Life, (Baltimore, Md., 1957) p. 54)

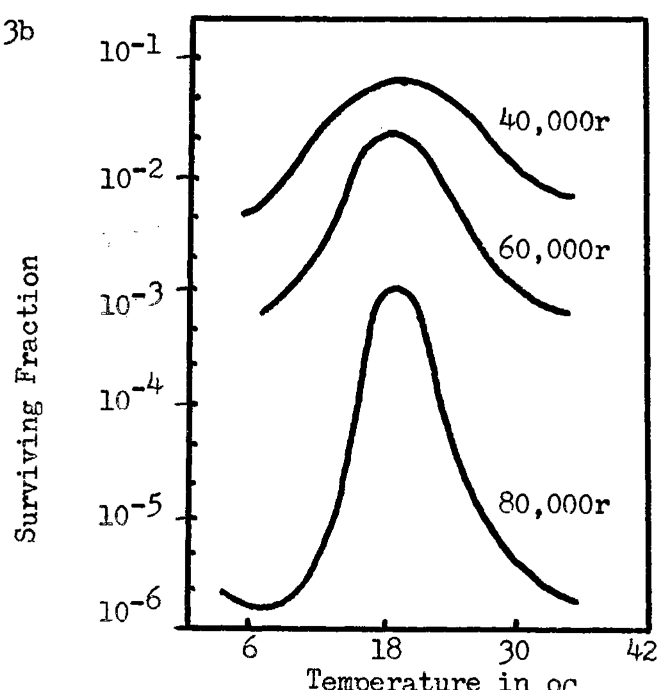
Chart 3a



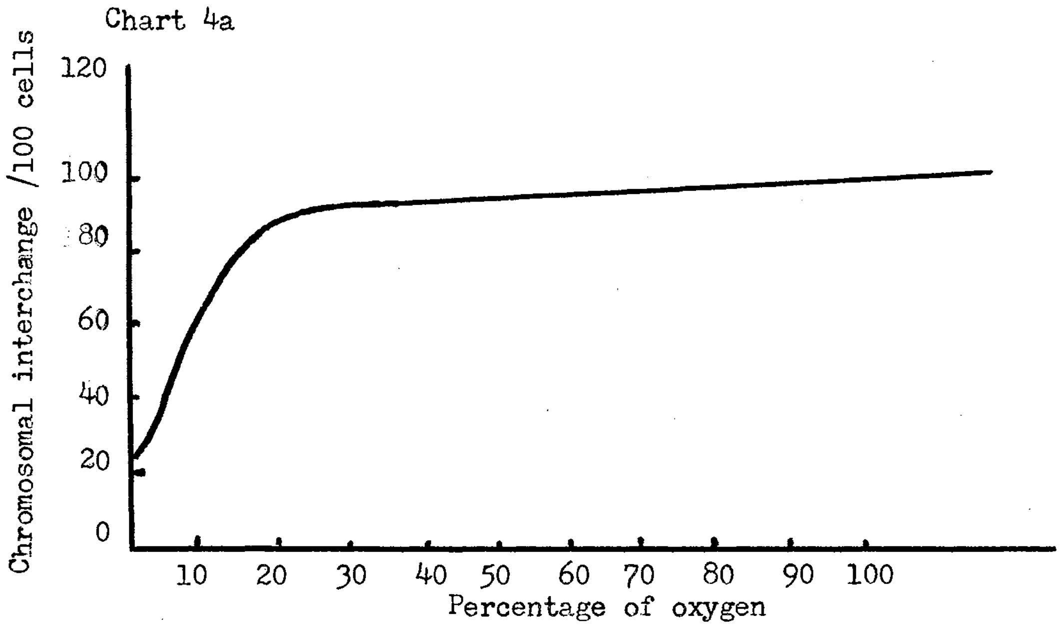
Percentage of oxygen in surrounding atmosphere

Relationship between the oxygen concentration in the atmosphere and the radiation sensitivity for different tissues.
(from P. Alexander, Atomic Radiation and Life, (Baltimore, Md., 1957) p. 167)

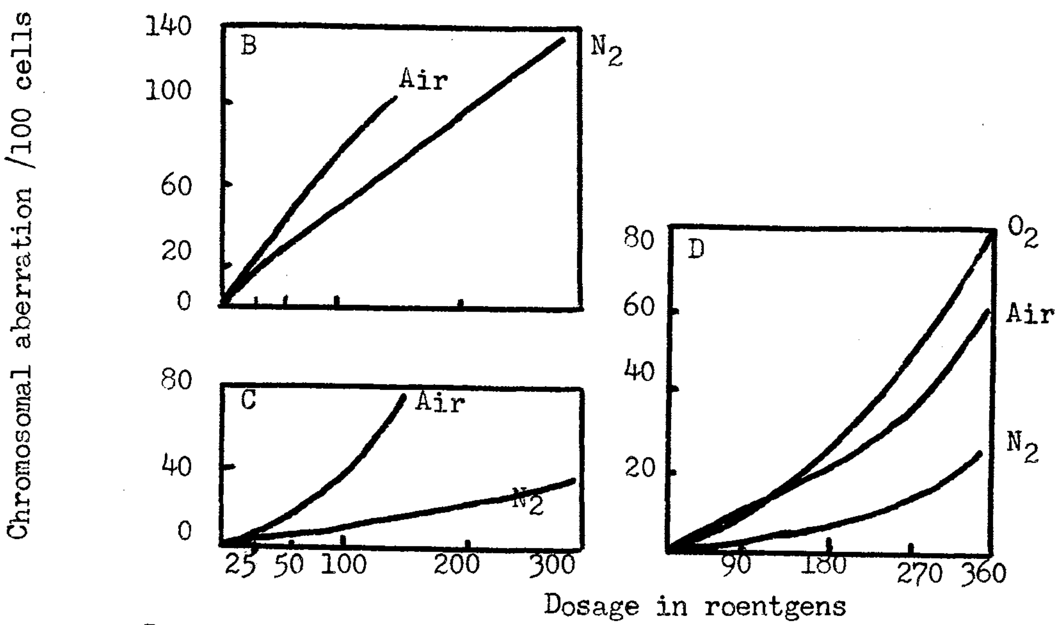
Chart 3b



Increase in survival of E. coli bacterium by storing at 18°C (64°F) after irradiation for a few hours.
(from P. Alexander, Atomic Radiation and Life, (Baltimore, Md.,1957) p. 174)



Relation between percentage of oxygen during exposure to x-rays and the frequency of chromosomal interchanges in Tradescantia microspores. Dosage 400 r at 50 r/min.



Relation of frequency of aberration in Tradescantia microspores to dosage when irradiation with X-rays is carried out in air and nitrogen. B, chromatid deletions; C, isochromatid deletions; D, chromosomal interchanges.

(from C. P. Swanson, Cytology and Cytogenetics, (Englewood Cliffs, N.J., 1957) p. 386-387) of irradiation of water. The principle chemical reactions in irradiated water are as follows:

$$2H_2O \longrightarrow H_2O_2 + H_2$$

$$H_2 \longrightarrow H + OH$$

Free hydrogen released in the above reaction may enter into the following chain reactions:

$$H + O_2 \longrightarrow HO_2$$

$$2HO_2 \longrightarrow H_2O_2 + O_2$$

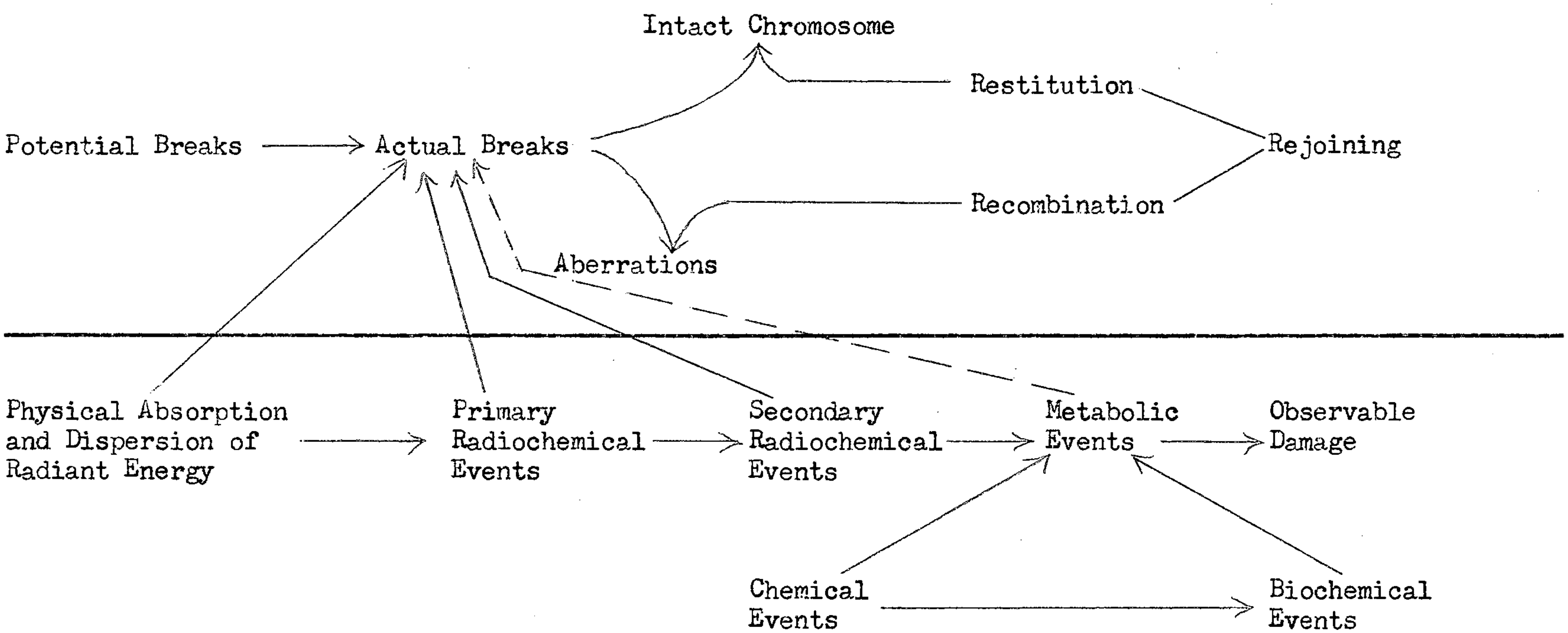
$$H + HO_2 \longrightarrow H_2O_2$$

Both HO_2 and H_2O_2 are very reactive compounds. The immediate chemical steps by which these reactants attack the chromosome and cause it to break are unknown. There is a proportionality between the amount of oxygen present and radiation damage. Chart 5 illustrates the possible relationship of chemical and physical events in the cell, arising from exposures to radiations and/or chemical mutagens; to structural events taking place in the chromosome. One out of every twenty breaks is a permanent break.[4]

If the source of nuclear radiation is concentrated, a proportional increase over natural or background radiation may be produced. In order to derive the maximum benefit from radiation in gene mutation one would desire to irradiate germ issue or primordial cells at a time when there would be the least number of chromosomal aberrations produced. If chromosomal aberration were effected, the gene mutation could be masked or lost. As is shown in chart 2, the optimal time for irradiation would

[4]Carl Swanson, Cytology and Cytogenetics, (Englwood Cliffs, N. J., 1957) p. 368.

Chart 5



The possible relationship of physical and chemical events in the cell, arising from exposures to radiations or to chemical mutagens, to structural events taking place in the chromosome. Solid arrows indicate possible radiation-chromosomal interactions; broken arrows indicate possible chemical mutagen-chromosomal interactions.

(from C. P. Swanson, Cytology and Cytogenetics, (Englewood Cliffs, N.J., 1957), p. 397.)

be during the metaphase stage. The difficulty lies in synchronizing the mitotic activities of the primordial germ cells. This is within the realm of possibility.

The production of chromosome aberrations is an important phase of genetics. If one can induce a multitude of different deletions in germ cells, the chromosomes posessing these deletions may be utilized in determining the loci of different genes in different organisms. They may also be used for a more rapid check on the phenotypic effects of mutations. Since most gene mutations are recessive in nature, a combination of a chromosome containing a mutated recessive gene with a homozygous chromosome with that gene loci deleted would give a rapid indication of the effects of the mutated gene. Thus if a person knew the locus of the gene which controlled the characteristic which he desired to change, and if he had access to gametes with that locus deleted, he could irradiate a wild type gamete and let it fertilize the gamete having the deletion. If the characteristic remained as before, the process would be reduplicated until some change occured which was favorable. While it is true that this is a trial and error venture, it is also true that it is an improvement in that the time required to induce mutations is greatly reduced.

CHAPTER II

RADIOACTIVE TRACERS

The application of radioactive isotopes to tracer work has been very promising. Although most of the successful work has been done with plants, there have been many instances where tracer work has been highly complementary to animal research.

In 1938 Schoenheimer and Rittenberg of the United States made an important discovery about food utilization by using radioactive isotopes. It had been universally believed that degradation changes in the living animal took place slowly, the purpose of food being largely to supply the currently required energy, while a small portion went to replace worn out tissue. As a result of experiments carried out with deuterium[5] and radioactive nitrogen as tracers, the idea of a near static tissue state was shown to be entirely wrong. It is now known that there is a continual interchange between the fats, proteins, and carbohydrates already present in the animal body and those ingested in the form of food.

Linseed oil was partially hydrogenated by means of deuterium, so as to yield a mixture of both saturated and slightly unsaturated fats, in which two of the hydrogen atoms attached to carbon were replaced by deuterium. The resulting deutero-fats, labeled in this manner were fed to animals. It was noted that only a small part of the deuterium was ex-

[5] radioactive hydrogen

creted for several days. The major portions of the deuterium was found to have been deposited in the fatty portions of the body. Even when the diet was very deficient in fat, and the total calorie supply was inadequate, so that the animal was drawing upon its reserves, the deutero-fat was mainly stored and not put to immediate use. After the natural diet of the animal had been resumed it was noted that the labeled deuterium began to disappear gradually, leaving the body in the form of water. However if the water included in the normal diet of other animals was enriched in deuterium so as to maintain a constant level of deuterium in the body fluids, the stored fat was found to gain deuterium at the same rate it was lost by the animals which had previously been fed the deutero-fats. These results indicated that reversible equilibria, involving fats and water, occur in the living organism. The saturated fats are desaturated by the removal of hydrogen or deuterium and at the same time the unsaturated fats are saturated by the addition of hydrogen or deuterium. Further investigations indicated that only singular unsaturated fats could be involved in this equilibrium. It is now known that the highly unsaturated fats must be supplied in the diet, the body being unable to synthesize them.

By using radioactive tracers much information has been gained about the utilization of minerals that could not have been determined without its use.[6] Many such examples are as follows:

Sulfur is needed by sheep for the production of two essential amino

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[6]Samuel Glasstone, Sourcebook on Atomic Energy, (2d ed., New York, 1958), p. 529.

acids in a wool molecule. By knowing the amount of sulfur needed, money can be saved in the production of mineral enriched foods for sheep.

Chickens require phosphorus and calcium in a certain proportion in egg production. By knowing this proportion, money can be saved in the production of chicken feeds.

It has been found that the proportion of phosphorus absorbed from a given soil and fertilizer depends on the nature of the plant and the period of growth. Tobacco and corn derive about 65 per cent of their phosphorus from fertilizer in the early stages of growth, but near maturity the proportion has dropped to 45 per cent for tobacco and only 15 per cent for corn. With potatoes 50 to 60 per cent of the phosphorus requirements are furnished by the fertilizer throughout the growth period at a constant rate.

It has been found that the ratio of phosphorus derived from the soil to that obtained from the fertilizer varies with the type of soil, its phosphorus content, the amount of fertilizer added, and the nature of the crop being grown. In general, the higher the amount of phosphorus in the soil, the larger is the proportion which the plants absorbs from that source.

It has been found that green manure was nearly as effective in supplying phosphorus as was super phosphate.

It has been found that during its growth period the corn plant's root system seeks phosphorus at a greater depth than that at which it is applied. Deeper location of phosphate would give better results. It has also been noted that foliage absorption of phosphorus prior to and during fruit development is 95 per cent efficient while root absorption during that same period is only 10 per cent efficient. Also in the case of corn, it was discovered that in the presence of certain soil chemicals calcium in fertilizer may become fixed in the soil and not available for absorption.

Peanuts absorb calcium more efficiently through their fruits. They also absorb calcium from immediately beneath the plant during the first seven weeks. This information is beneficial in fertilization practice.

The most efficient results in fertilizing apple trees with phosphorus and magnesium compounds occurs following the flowering season when applied to the verticle center of the leafing portion of the tree. Phosphorus and magnesium is needed in seed development, and if not supplied by an external source it is taken from essential parts of the tree. If it is applied to the verticle center of the leafing portion of the tree, the fertilizer is absorbed and translocated both upward and downward.

In working with radioactive iron, it was found that little of the labeled iron found its way into the hemoglobin of the blood. Most of the iron was passed from the body unused. In animals depleted of iron, a larger portion of this element is absorbed. It was also found that shortly after a loss of blood the immediate uptake of iron was not effected. Only after the hemoglobin begins to be replaced by drawing upon the bodies' supply of iron is there considerable absorption of iron from the food. Thus it was found that the extent to which iron is absorbed is dependent on the reserves of the element in the body. It was also found that iron is used over and over again. When red blood cells are destroyed by the liver, the iron is retained in the body and reincorporated into the hemoglobin of new red blood cells.

It has been known for many years that both animal and vegetable life are dependent on very minute quantities of certain elements, such as boron, cobalt, copper, manganese, molybdenum and zinc. These elements, being needed in minute quantities, are called micro-nutrients. It has been found that cattle fed on forage containing less than four parts cobalt in one hundred million of their diet will lose their appetite and actually starve to death, although adequate pasture is available. Other ruminants are similarly effected. By application of radioactive cobalt it has been found that cobalt is required by symbiotic bacterial coliforms in the production of vitamin B_{12}. By utilization of radioisotopes, research on micro-nutrients has evolved from a virtual impossibility to a practicality.

Using radioisotopes an accurate count has been made on the red blood cell count and the average life span of the red blood cells in the body.

Circulatory impairments are readily detected by use of radioactive isotopes.

Hyperthyroidism has been reduced by utilization of the iodine absorption characteristic of the thyroid gland. Radioactive iodine, which is a gamma ray emitter, is injected into the body. Tissue destruction results.

Information on the phenomenon of photosynthesis is currently evolving out of the use of radioactive isotope tracer work. This could well pave the way toward an economical source of energy.

Other instances in which radioactive elements have been used include research with cancerous tissue. It is known that cancerous tissue has a greater affinity for certain metals than do normal tissues. Different cell types have affinities for different metals. This knowledge is advantageous to medical science in two ways. First, it enables one to locate the tumor site. Secondly, after one knows the primary site of the tumor he can select a radioactive metal which has a low penetrating emission and a high affinity for that tissue, inject it into the body of the patient and destroy the cancerous cells. It is highly possible that the greatest use of radioactive isotopes in cancer treatment would be in conjunction with surgery, where the isotopes would be utilized to destroy any cancerous tissue inadvertently missed by surgery.

Brain surgery has been performed by focusing three converging beams of gamma radiation on a tumor. Neither of the individual beams is of sufficient intensity to do considerable damage, but when they are concentrated on a spot, cellular destruction results.

CHAPTER III

STERILIZATION OF FOOD AND COMMODITIES

The utilization of nuclear radiation in food sterilization and preservation[7] is one of high ranking importance since the cost of irradiation is low in comparison with the cost of refrigeration. In order to effect sterilization the radiation dose must approximate 2,000,000 reps.[8] Certain chemical reactions occur in foods following sterilization by irradiation. Since most of these reactions involve enzymes, it might be well to examine some of the occurences. Enzymes are chemicals produced by living cells for the purpose of controlling specific chemical reactions. Since enzymes are produced in quantity in every living cell, foods in the form of meats, fruits, and vegetables contain proportionally large amounts of these enzymes. Some of these enzymes such as those that bring about the tenderization of meats, are good. Some are not so good, like those that bring about the rancidity of fats. Unfortunately these enzymes are not as susceptible to radiation as we would like. It is fortunate that other means have been developed to destroy these enzymes such as a mild heat treatment or by blanching.

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[7] Radiation Sterilization of Foods, U. S. Government Printing Office, (Washington, 1955).

[8] roentgen equialent physical - absorption of 97 ergs of energy per gram of soft tissue or approximately the energy absorbed from exposure to one roentgen of X-rays.

It is also fortunate that most foods contain little of the harmful type enzymes.

The second group of reactions with which we are concerned involves off-flavors and off-odors generated as a result of nuclear radiation. When we look at the radiation process, we note that the rays disrupt the protoplasm of the bacteria and damage the cell, thus bringing about death. Since rays cause this extensive amount of damage to the bacteria, it would also be reasonable to expect that it would alter the food tissues to the same or to a greater degree. It is known that this is what happens. There is frequently a generation of off-odors and off-flavors, especially when doses above a million roentgen units are used.

The problem then is how to kill the bacteria and still not develop the side effects of off-odors and off-flavors. It has been found that if the food is irradiated in the frozen state or in the absence of oxygen, the amount of off-odor is minimized. If a food is irradiated in the presence of some inhibitors such as ascorbic acid, the amount of off-flavor and off-odor is reduced.

A third group of reactions which must be contended with is the destruction of vitamins in foods. It is known that only the B group is effected and of these, only B_{12} is markedly reduced in quality following intense irradiation.

If gamma or beta irradiation is used on foods there is no reason to be concerned about the transmutation[9] of stable elements into radioactive elements. While there will be a slight amount of transmutation, the

[9]the conversion of one element into another. The new element may be radioactive.

amount of radioactive material produced would be negligible.

Chart 6 lists some of the more common foods categorized according to the possibility of storing them without refrigeration[10] following irradiation. In addition to the practicality of irradiation of foods for storage without refrigeration, it is both practical and possible to extend the length of the shelf life of refrigerated perishables by irradiating foods prior to refrigeration.

Besides these two uses of food storage, there are still other possibilities of the application of radiation to foods and other commodities. These are the non-sterilizing radiation applications which have a rep range of 100,000 to 500,000.

One such application is the destruction of parasites in food. It has been estimated that 6 per cent of the meat obtained from pigs, which have been fed a diet of uncooked garbage, have trichina parasites. Small doses of radiation in the range of 10,000 to 50,000 reps are used to destroy this parasite. It is felt that irradiation of other potentially parasitic infested foods will yield the same results. With such low dosage of radiation, none of the deleterious effects of higher dosage of radiation are noted.

Another application deals with the destruction of insects in foods, clothing, and related commodities. Very low doses readily destroy weevil, moths, and other damaging insects.

The increased storage life of tubers may be increased by exposure to radiation. This is primarily due to the inhibition of sprouting. An inhibition of sugar formation in tubers is also effected. This is

[10] not to be confused with freezing.

Chart 6

Good	Fair	Poor
Bacon	Asparagus	Banana
Beans, green	Applejuice	Beans, lima
Beef liver	Applesauce	Cantaloup
Broccoli	Beefsteak	Celery
Brussel Sprouts	Bread	Cheese
Carrot	Cabbage	Frankfurters
Chicken	Cherries	Lemon Juice
Codfish cakes	Corn	Lettuce
Pork	Halibut	Milk
Pork sausage	Ham	Oranges
Waffles	Hamburger	Orange Juice
	Lamb chops	Squash
	Peas	Strawberry
	Spinach	Syrup
		Tomato Juice

Present estimates of promise of storage of foods without refrigeration following irradiation. The dose used was about 2,000,000 reps.

(From the hearing before the Subcommittee on Research and Development of the Joint Committee on Atomic Energy Congress of the United States, Eighty-Fourth Congress, First Session on Radiation Sterilization of Foods.)

of particular importance to manufacturers of potato chips. Normally potatoes stored at 50° F. have high values of sugar formation.

Some types of orange juices will develop a bitterness upon standing for a day. If the oranges, from which the juice is extracted, are irradiated prior to extraction of the juice this bitterness will not develop.

CHAPTER IV

CYTOLOGICAL AND GENETIC APPLICATIONS

There are many examples of practical applications of nuclear radiation on genetic material. A classic example is the case of screw-worm fly extermination on a small island in the Caribbean by Bushland and Hopkins in 1951.[11] It was known that the breeding habits of screw-worm flies was seasonal. When the population becomes drastically reduced, the mating season begins. An experiment was conducted in which thousands of male screw-worm flies were irradiated to the point where the germ cells contained many aberrations but remained functional. These flies were released among the wild flies at a time when the population was at its extreme low. The irradiated flies were more numerous than the wild flies, with the result that most of the fertilization occurred between normal females and the irradiated males. The male gametes contributed lethal deletions which acted as dominant lethal genes. Because of the large number of irradiated males released, proportionally few of the zygotes would be a result of the fusion of normal gametes. By repetition of this practice of dispersion of irradiated flies, the population of screw-worm flies was soon depleted.

It is felt that this approach would work equally well on any organism

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[11] the principle of the method was suggested by E. F. Knipling.

where the foci of natural breeding are sufficiently accessible to be adequately reached in the process of distributing the irradiated individuals.

In working with irradiated barley seeds it has been estimated that one in ten irradiated barley seeds contained a recessive mutation and only one out of every eight hundred mutations was found to be of some use in adapting barley better suited for the needs of man.[12] The same type mutations develop under natural conditions but under natural conditions the changes selected for multiplication are those which are advantageous for the species itself rather than for man. In spite of the fact that spontaneous mutations, like those produced by radiation, are in overwhelming majority detrimental, an advance in adaptation results by virtue of the selective multiplication of the very few gene mutations, and fewer multiplication of the very few gene mutations, and fewer chromosome structural changes, that happen to be helpful. Thus, it might be thought that the continual application of radiation would merely speed the advance, if other natural processes were allowed to take their course.

This advance can and does increase provided the necessary conditions exist. Of the necessary conditions the first is that the spontaneous mutation rate should not already be so high that when irradiation is applied mutations occur too frequently to allow an equilibrium elimination rate and/or a genetic load low enough to be tolerated by the population. A second condition is that the advantageous mutations should

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[12] Alexander Hollaender, Radiation Biology (Vol. 1, pt. 1, New York, 1954), p. 436.

multiply fast enough to replace the original type at a rate commensurate with their increased rate of origination. A third requirement is that the organism should not be at the limit of an evolutionary dead end, or in other words that there are possibilities of improvement. Opportunities for improvement will be present in greater abundance, allowing more of the mutations that occur to be helpful, if the population has been placed in an environment, and subjected to conditions of living, somewhat different from those previously natural to it; for it must already have become so highly adapted to its natural conditions as to make further progress difficult. Advance is also achieved more readily if the population is one which has to some extent lost its original ability to adapt due to genetic changes or recombinations. This regression may have come about through the prior establishment of some harmful mutations, the effects of which can now be overcome by reverse or counteracting mutations. This regression is most likely to have occurred as a result of derivation from one, or from a few more or less inbred lines, or from relatively few progenitors.

It is felt that mutations advantageous to man can be developed in parasitic microorganisms.[13] This involves the development of strains whose virulence has been so decreased that they may be used as live vaccines as in the manner of cowpox. The chief difficulty lies in obtaining varieties which can be relied upon never to reverse mutate into virulent forms. Determination of this stability would be difficult unless one could associate changes in morphological characteristics with

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[13] Alexander Hollaender, Radiation Biology (Vol. 1, pt. 1, New York, 1954), p. 440.

changes in virulence. This can frequently be done. Once a non-virulent strain has been developed, the rate of mutation within this strain can be increased by irradiation. If, after many generations, no virulent strains are produced, one can be safe in assuming the strain will not reverse mutate for the virulent characteristic. Fortunately this 100 per cent stability in regard to a given characteristic is not required in the case of other genetically desirable types of organisms.

The decision as to whether the radiation technique will be helpful must depend upon a number of factors which differ according to the species and variety in question. In general, the larger, the slower growing, and the smaller the potential number of offspring per individual, the less suited it is to the use of the radiation technique for its improvement, since the individuals are less expendable. Radiation is accordingly most likely to be found advantageous for small, rapidly multiplying organisms, including especially those which are habitually selfing, or which have major haploid phases, and which have therefore been weeded comparatively free of spontaneous mutants.

Commercially valuable mutations in mushrooms effecting color, growth rate, and fruiting time have been produced by using nuclear radiations as a mutagen. Other commercially valuable mutations which have been produced in this manner are: cereals which ripen earlier or give higher yields; peanuts which are superior in yield, better adapted to mechanical harvesting and are resistant to a serious leaf spot disease; a variety of oats resistant to stem rust and another normally susceptible to Victoria blight with a greatly increased resistance to this disease. In addition several interesting somatic mutations, capable of vegetative propagation, have been produced in carnations and other flowers.

It is evident that microorganisms are particularly well adapted for the study of the mutagenic effects of radiation. One example is provided in the experimentation with the mold Penicillium, initiated by Hollaender in 1945 and continued by another group of investigators. In this work, the irradiation was carried out in several successful steps. Following each exposure, that mutant was selected which had the best yield of penicillin and the colony derived from it was then used for the next exposure. In this way after many generations of exposure, a strain was produced, incorporating all the mutations together, which had a yield of some four to five times as high as that of the original variety.

A similar experiment was conducted with a suitable variety of the mold Aspergillus terreus. This particular mold is the primary source of the economically important substance itaconic acid. The results of this process was a better variety of mold which gives a higher yield of itaconic acid with a lower concentration of contaminating substances.

For the future there are many possibilities. It should be possible by successive mutations to adapt bacteria, fungi, protozoa, and viruses to hew hosts, or even to make free living ones parasitic as well as increase their virulence for their hosts so as to make them useful in the control of insects, predators, noxious weeds, and other undesirable species. Tissue specificities also could be developed in parasitic microorganisms, to attack certain malignant tumors. The opportunities of establishing, in organisms not now posessing them, other beneficial forms of symbiosis with microorganisms, such as already exist for example in ruminants and termites, seem to have been exploited only to a very limited degree. There is a vast room for the further development of valuable or potentially valuable microorganisms for the production of foods, food accessories, and pharmaceuticals such as cheeses, wines, and antibiotics; and as food themselves such as yeast, plankton, and algae; and for use in varied biochemical reactions of economic importance in other ways as in the production of important industrial organic materials; and in the synthesis, by the efficient use of solar energy, of energy rich combustibles.

Mutagens other than radiation can be successfully used. Experience to date has indicated that radiation is the most satisfactory because of its convenience of application, its penetration, and the ready control of its intensity and timing.

CHAPTER V

SUMMARY AND CONCLUSIONS

The preceeding chapters are involved with the blessings which can be associated with nuclear energy. Limitations and disadvantages are also associated with this agent.

In working with radioactive sources adequate shielding must be established for two reasons:

(1) to protect the handler,

(2) to eliminate, as much as possible, the background or natural radiation.

Radiation sources cannot be handled indiscriminately because of the harmful effects it has on human tissue. Earlier it was stated that the more highly developed animals were also the most radiosensitive. It is then relatively easy to draw a conclusion as to its effects on human tissue.

In the second case, if a critical measure is needed, background radiation can completely nullify hours of careful preparation if adequate shielding is not furnished. Since this shielding is most usually made from lead, it doesn't have the convenience of portability.

Man has progressed far in the use of nuclear radiation in the field of progressive biology in a few short years. It is anticipated that new discoveries will evolve at an ever increasing rate.

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