EFFECT OF RADIATION
ON
HUMAN HEREDITY

Report of a Study Group convened by WHO
together with
Papers presented by Various Members
of the Group

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PREFACE

In 1956, two committees—one set up by the National Academy of Sciences of the United States of America and the other by the Medical Research Council of Great Britain—reported on the effects of ionizing radiation on man. Although difficult to compare in detail, these reports come to remarkably similar conclusions as to the probable effects on the descendants of populations exposed to increased amounts of such radiations. The emphasis in both these reports was, however, on trying to set some quantitative limits to the potential risks in the light of existing knowledge rather than on attempting to assess the long-term dangers.

WHO's purpose in convening the Study Group on the Effect of Radiation on Human Heredity, whose report is presented here, was essentially twofold: The first aim was to obtain the opinions also of authorities on genetics from countries other than those whose national committees have already stated their views. The second was to hear the opinions of a number of experts on an aspect relatively lightly touched upon in the national reports—namely, the lines of research which should be followed, in the light of present knowledge, to increase our understanding of the genetic effects of ionizing radiations on man.

In addition to the formal report of the Group, the papers presented by various members have been reproduced. It should be emphasized, however, that while the Group's report is intended to represent the views of all the participants, the opinions expressed in the individual papers are those of the authors and do not necessarily represent the views of the Group as a whole.

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PART I

REPORT OF STUDY GROUP

REPORT OF STUDY GROUP ON THE EFFECT OF RADIATION ON HUMAN HEREDITY

The Study Group on the Effect of Radiation on Human Heredity met, by courtesy of the Rector of the University of Copenhagen, in the Council Room of the University, from 7 to 11 August 1956. The agenda adopted was intended to permit exploration of the views of the members of the Group on the theoretical and practical difficulties in closing present gaps in knowledge. The procedure followed was for a number of members to open discussions either by short statements or by the presentation of invited papers. The opportunity was also taken to discuss a number of subjects not formally introduced.

The proceedings were opened by Dr P. Dorolle, Deputy Director-General of the World Health Organization, and the Group elected Dr A. Hollaender as Chairman.

1. Introduction

Man's most precious trust is his genetic heritage, upon which must depend the health and orderly development of future generations. The Group is of the opinion that the well-being of descendants of the present generation is threatened by developments in the use of nuclear energy and of sources of radiation. Both of these developments are inevitable and they should contribute much to man's social and cultural development. It would seem therefore that some risk must be accepted, but if the dangers are to be minimized every possible step must be taken to reduce the exposure of man and to understand the effects of exposure. Only in the light of more knowledge can decisions be taken to define more accurately the maximum amount of exposure which may be accepted by individuals and populations without risk of serious harm.

Radiation has been demonstrated to be one of the agents which produces mutation in a wide range of organisms from bacteria to mammals. The Group is agreed that additional mutation produced in man will be harmful to individuals and to their descendants. While there may be inherent and environmental mechanisms which modify the impact of these mutations over periods of many generations, the effectiveness of such mechanisms in man is not known. In essence then, all man-made radiation must be regarded as harmful to man from the genetic point of view.

In recent years, considerable quantitative knowledge has been accumulated on the basic mechanisms of genetics. There are strong grounds for believing that most genetic effects are very closely additive so that a small amount of radiation received by each of a large number of individuals can do an appreciable amount of damage to the population as a whole. There are, however, many gaps in knowledge particularly concerning these effects in man. These gaps will only be closed after a great expansion of general and *ad hoc* research in genetics and other fields of biology.

The Group has received the following resolution passed by the First International Congress of Human Genetics in Copenhagen, and it notes and agrees (while at the same time noting that WHO's work is only concerned with the peaceful use of atomic energy) that:

"The damage produced by ionizing radiation on the hereditary material is real and should be taken seriously into consideration in both the peaceful and military uses of nuclear energy as well as in all medical, commercial and industrial practices in which X-rays or other ionizing radiation is emitted. It is recommended that the investigation of the amount and type of damage and of related genetic questions, be greatly extended and intensified with a view to safe-guarding the well-being of future generations."

The Group agrees with the memorandum, entitled "Human and Medical Genetics", which was submitted in 1955 by the Government of Denmark to the World Health Organization.[1]

This Group takes note of the report of the National Academy of Sciences of the United States of America[2] and that of the Medical Research Council of Great Britain.[3] It is not intended to reproduce any of the material in these reports, but the Group notes the substantial similarity of the findings and recommendations of these reports and is in essential agreement with them.

2. Natural and Man-made Sources of Ionizing Radiation

The present sources of ionizing radiations of interest for the treatment of problems related to the genetic effects i ma include the following:

Natural sources

1. Cosmic radiation.

2. Naturally occurring amounts of radium, thorium and potassium in the earth's crust.

3. Content of natural radioactive elements in living tissues.

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[1] *Off. Rec. Wld Hlth Org.*, **68**, 147

[2] United States of America, National Academy of Sciences (1956) *The biological effects of atomic radiation*, Washington, D.C.

[3] Great Britain, Medical Research Council (1956) *The hazards to man of nuclear and allied radiations*, London

Man-made sources

4. Radioactive material and technical arrangements producing ionizing radiation (such as X-ray tubes and other particle accelerators, nuclear reactors, etc.) used in education, science, medicine, industry and commerce.

5. Sources used by the population for other purposes than those mentioned in 4 (radioactive luminous compounds on watches and other articles for common use, television sets, etc.), although such sources are much less significant than those mentioned in 4 and 6. It is important, however, that their existence be recognized.

6. Artificial radioactive elements distributed by man in nature.

Information as to the contributions to the doses received by individuals and by large population groups from the various sources listed above is summarized in Professor R. M. Sievert's paper (see page 63), from which it is obvious that as regards the average dose to the gonads the most important contributions are at present those from the natural radiation (normal level: between 2 and 5 r per individual in 30 years) and from the radiation received by patients undergoing medical X-ray examination (probable average: between 1 and 3 r per individual in 30 years). If therapeutic exposures are also considered, the "total" exposure to a population might be greater. It is, however, difficult to get sound data for estimating how much exposure is received in therapeutic exposures to persons before the age at which procreation may be expected to be ended.

It may be noted that at the present time the highest dose to the gonads caused by natural radiation in areas with a large population seems to exist in parts of Travancore, India, on ground containing monazite sand (possibly of the order of between 10 and 20 r per individual in 30 years).

3. Importance of Recording Radiation Exposure in Individuals and Populations

From a genetic point of view the total accumulated dose is the important one and for this reason the measurement of exposure to ionizing radiations is an essential preliminary to attempts to relate dosage received to effects in man. For such measurements to be useful, the information must be recorded systematically. Unless the information is available in the form of the dose received by individuals, records of exposure would be unsuitable for many purposes and therefore some system of registration is essential. The effect of recording would almost certainly be to cut down the exposures given in medical diagnosis and treatment, since it would impress radiologists and technicians with the magnitude of such exposures. In one hospital where such recording was started there has been a 30% reduction in the total exposure of the staff. Doubtless a similar system of recording in diagnostic practice would reduce the exposure to the patients. This in itself would

be a sufficient justification for introducing the procedure. It seems likely that the two national reports will already have done much to overcome the hesitation to record the dose on the part of those who would be concerned in making such records, but that a recommendation from this Group would also be helpful.

The Group is conscious that the adoption of any system of recording dosage will give rise to difficulties because it will increase the burden of work of radiologists and their staffs. Nevertheless, they feel that the importance of these procedures is such, and is so well recognized by radiologists, that both those in charge of radiological departments and other physicians who use X-rays will be co-operative.

Whatever system is adopted should take into account three desirable requirements:

1. That the individual will not, through lack of information, accumulate excessive exposure.

2. That information becomes available as to how much exposure to the gonads is received at each age in individuals and on an average per head of population.

3. That it should be possible to recognize the amount of exposure received by the parents of a given child. (Eventually, the information would be available for several generations.) This information is particularly valuable for purposes of genetic analysis.

The Group suspects that exposures in some industries and in scientific work are unnecessarily high. Exposures from these sources should be recorded in such a way that the dosage received can be related in individuals and populations to that received from other sources.

It seems unlikely that all countries would favour, or indeed would be able to introduce, the same standards of registration. Although it is expected that recommendations on mechanisms of recording will shortly be available from the International Commission on Radiological Protection, there should not be any delay in improving the standard of recording of exposures.

Whatever procedures of recording and registration are adopted will entail a large expenditure of money and effort. The need, however, is urgent. Further, the present is the appropriate time to initiate such procedures, since the introduction of atomic energy for industrial use and the extension of the use of radiation tools in biology and medicine make it possible to start with such procedures at an early stage of a period of rapid development.

4. Research

General

Additions to the understanding of the effects of radiation in man come from a very wide field of research. It is impossible to forecast what work in biology or genetics will contribute information relative to the problems. Accordingly, the Group is strongly of the opinion not only that as much experimental work as possible should be done on radiation effects on suitable organisms and such controlled observation studies as offer in man, but that there should be an intensification of all human and experimental genetic research. The Group feels that there should be the closest possible collaboration between those working in the experimental and human fields: their work is complementary. Each should be stimulating the other's research projects. This need for intensification of research in man and in other organisms raises problems of finance and of shortages of trained research workers. Both these difficulties are likely to be intensified if new areas of work, such as that on tissue cultures, chemical mutagenesis, serology, biochemical genetics and epidemiological problems of genetic disease, are to develop as rapidly as is desirable. The problem of manpower shortages, in regard to both biologists and physicians, tends to be perpetuated by lack of career opportunity for those working on genetics. There is also an insufficient number of institutions where an adequate training in genetics, particularly in human genetics, can be given.

It is possible that the results of much effort in these fields will prove disappointing. Nevertheless, research workers and those supporting their work must have the courage to face the possibilities of such disappointments and still go forward.

The developments of nuclear energy would never have been made unless enormous risks of failure had been accepted. These innovations have extremely important implications among which the possible effects on man's genetic composition are outstanding. If there is to be a climate of public opinion favourable to the development of nuclear energy, the peoples must be assured that investigations essential for their future health and welfare and that of their children will be undertaken on an adequate scale. This will require recognition by governments that very substancial financial provision must be made for genetic and other biological investigations essential to an understanding of the effects of radiation on man. Biological research in the past has suffered severely from lack of funds.

Specific

The Group does not feel that it should attempt to recommend specific research projects. Nevertheless, it seems desirable to recognize the larger

gaps in knowledge as they appear at the present time. Among the fields in which the need for further work is urgent, if the genetic hazards of the irradiation of human populations are to be understood, the following appear outstanding. It should be emphasized that the rapid developments in genetics and other sciences must determine that recommendations for lines of research should only be accepted as tentative and should be revised periodically.

(*a*) *Further study of spontaneous and artificially induced mutation.* There is need for further study of the number and kinds of mutations produced by various doses and types of irradiation applied at different stages of the life-cycle under a variety of conditions and utilizing different kinds of organisms. The relatively limited opportunities to study irradiated human beings and their offspring should be exploited to the fullest extent possible. The appreciation of radiation-produced mutations is intimately related to a similar extension of knowledge concerning mutations that appear to arise spontaneously or as the result of the action of chemicals and of physical agents other than ionizing radiation.

(*b*) *Mutational component in the somatic changes produced by radiation and other means.* The role of changes in the hereditary material of somatic cells in the genesis of leukaemia, in other forms of neoplasms, and in alterations in the life-span is at present a controversial field which needs clarification. The effects of low doses of radiation, including those from radioisotopes, require special study. An important method of attack on this problem is opened by recent developments in tissue-culture techniques.

(*c*) *Means of protection against mutagenic agents.* The pioneer studies which indicate the possibility that the production of radiation-induced mutations can be modified by various means have important implications for man and require extension in many directions.

(*d*) *Development of new and improved techniques for the identification of mutants.* Efforts directed at developing more exact methods for the recognition of mutant individuals, and the distinction between the latter and phenocopies, should be intensified. It is important to prosecute studies of the frequency of a wide range of types of mutations, including those with extremely small effects, recognizable only through special statistical or breeding techniques.

(*e*) *Manner of gene action.* The phenomena of dominance, synergism and other forms of gene interaction, the multiple effects of a single gene and the role of environmental factors in the determination of traits require a great deal of elucidation, since they are highly important in appraising the effects of radiations. They should be studied both in man and in other organisms. In this connexion, the prospects raised by the rapid advances being made on human biochemical specificities are of particular interest.

(*f*) *Selective factors in populations, with particular reference to the special conditions in man.* Very little is known concerning the detailed effects of natural selection on the frequency of specific genes, constellations of genes, or cytological alterations. Such information is basic to attempts to understand the genetic composition of present and past human communities and to predict future trends consequent upon changes in radiation levels, medical practices, and social and economic conditions. These gaps in knowledge can in part be filled by the collection of relevant demographic and experimental data.

(*g*) *Patterns of mating in human populations and their genetic implications.* A standard type of information always required in understanding the genetic composition of human populations and the effect on it of various amounts of radiation is the recording and interpretation of data on the consequences of inbreeding, assortative mating, geographical and cultural isolation and random genetic fluctuations.

(*h*) *Twin studies in man.* These are recognized as being helpful in understanding many problems of human heredity. Such studies have already been extensively used, but could be advanced by standardized registration of twins in various countries. They give useful information concerning the relative importance of hereditary and environmental influences.

(*i*) *Determination of the frequency of diseases with a significant genetic component, with particular reference to their epidemiology.* This is fundamental for investigations on the significance of mutation as a cause of disease in man. In this connexion central registration of human inbreeding, hereditary disease and variation is of the utmost importance. It is also of importance to know the number of people who, on account of hereditary lesions, have to be treated in hospitals or institutions or given social aid.

(*j*) *Study of populations of special genetic interest.* Important information is to be obtained from the study of relatively stable, primitive communities, long isolated by geography or culture. Studies of this type require for their execution teams of persons from a variety of disciplines, such as cultural anthropologists, physicians and geneticists. It should be emphasized that the understanding of the genetic structure of contemporary populations will be greatly aided through these studies, which should be maintained continuously over a considerable period of time. The opportunity for these studies diminishes with each passing year. Among special communities to be studied are those receiving unusually large amounts of radiation, those in which the degree of inbreeding has long been very high or low, and those in which special conditions of selection have prevailed. In some investigations radiation physicists would be essential members of the teams.

(*k*) *Genetic mapping of human chromosomes.* This is a highly specialized field in which encouraging advances are now being made. Among the possibilities to be exploited is the use of such data to aid in the identification of independently occurring mutant genes and in the study of chromosome rearrangements.

(*l*) *Cytochemistry and human cytology.* Direct cytological observations should be conducted both on normal individuals and on those with suspected chromosomal abnormalities. Material from the individuals themselves as well as mutant cells of tissue cultures may be used in such work. Basic information concerning the ultra-microscopic structure and chemical composition of the hereditary material, and the manner in which this is altered by irradiation and other mutagens, is essential and should include information on lower organisms as well as man. The new developments in biochemistry, the emerging immunobiochemical investigation of tissue proteins, bone-marrow and other tissues, the metabolic investigations which may elucidate both physical and mental pathology, the new developments in electronmicroscopy which advanced our knowledge of the structure of human sperm all indicate the development of new tools for the study of human genetics.

(*m*) *Development of further statistical methods.* New mathematical methods have continually to be developed to deal analytically with problems which arise as the result of researches in human and in experimental population genetics. This is particularly so in relation to observations on the genetic structure of and intensity of selection in populations with regard both to traits due to single gene and those due to multiple gene effects. Special techniques requiring electronic computers will also be required for analysing data on genetic linkage in man.

5. Some Conclusions

(*a*) The Group is of the opinion that there are too few institutions or large university departments devoted to general genetics and even fewer concerned with human genetics. It recommends the establishment of such institutions and departments and suggests that there could be no one ideal pattern. One of the benefits of such institutions would be to accustom people of different scientific disciplines having implications for genetics to work together. Physicians, general biologists, geneticists, biochemists, cytologists, serologists and statisticians are examples of the kind of workers who may be needed. When such institutions are concerned with human genetics their location should have regard to the adequacy of existing medical services, to the kind and size of human populations available for field studies and to the adequacy of background vital statistics and general demographic information on the population concerned. For many purposes

a population of about two million is optimal, particularly for intensive epidemiological investigations. Such institutions, in addition to their research functions, could eventually serve as centres of elementary and advanced training in genetics.

(*b*) Such research departments and institutions should contribute much to teaching in general and human genetics. Medical undergraduates should all receive training in genetics and the teaching should be co-ordinated with that in radiology and in the use of radioactive substances in medicine, so that the genetic hazards of diagnostic and therapeutic procedures are thoroughly understood. Medical men training as radiologists should have specific, more advanced instruction in genetics. Health physicists, radiological physicists and radiological technicians should also receive instruction in genetics as part of their technical training.

It seems essential that instruction in genetics should be given to all scientists, particularly those whose work is likely to involve the use of radiation and radioactive materials in research. The principles of human genetics could with advantage be conveyed to those training in the social sciences by means of formal instruction. Finally, the Group is of the opinion that public education in genetics should be more common and adequate than it is at present.

(*c*) In the future it would be necessary from the point of view of preventive medicine and genetic hygiene to register serious hereditary diseases and defects in various populations or countries in the same way as, for instance, epidemic diseases. For that purpose, genetic-hygiene ascertainment or registration will be an indispensable and necessary step. The recording of hereditary diseases and defects in various countries and regions is to be highly recommended.

(*d*) In many countries there are very few biologists or physicians properly trained in genetics. This situation will only be solved by producing more career opportunities in genetics, but may be alleviated by granting fellowships or subsidizing training at approved institutions in countries which can offer training facilities. It is possible, also, that advice and technical assistance could be given in connexion with research projects in countries with insufficient resources in trained manpower to carry them out.

(*e*) It might be possible for a United Nations Agency to assist on request in administration or supervision of studies of specific populations over a period of years or by strengthening a research team or by giving advice on organization.

(*f*) In the past, United Nations Agencies have done useful service in contributing to the collection and standardization of vital and health statistics. It is recommended that such agencies continue their efforts and

stimulate the efforts of others in the collection and publication of specific data such as fertility, consanguineous marriages and parental ages, which are so essential as background information in many studies in human biology.

(*g*) The Group wishes to call attention to the evidence that damage to body tissues produced by radiation after relatively small doses is, at least in part, mediated through effects on genes and chromosomes. There is also some evidence that the life-span may be reduced in mammals even by relatively small doses. *Ad hoc* investigations are urgently needed.

(*h*) The Group is particularly impressed with the genetic hazards of man-made radiation from sources used in medicine, industry, commerce and experimental science, etc. Both as an approach to control and as providing basic background information for relating quantitatively radiation exposure and effects on man, it is essential that methods be found of recording exposures to individuals and populations, however difficult this may prove.

There is reason to believe that radiation exposure can be much reduced; therefore, those in charge of sources of ionizing radiations should always ensure that there is adequate justification for exposing individuals to doses however small. On account of the danger to offspring resulting from irradiation of the gonads by X-rays, consideration should be given to determining what efficient means of shielding the gonads could be devised and brought into general use. In addition, in every exposure, the X-ray beam ought as far as practicable to be directed so that a minimum of radiation reaches the gonads.

Annex

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PART II

PAPERS PRESENTED AT STUDY GROUP

DAMAGE FROM POINT MUTATIONS IN RELATION TO RADIATION DOSE AND BIOLOGICAL CONDITIONS *

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Accumulation

A topic which the writer has been requested to discuss in this paper is that of the accumulation of point mutations following repeated irradiation. An accurately additive accumulation in the germ cells throughout life has as its necessary and sufficient conditions (*a*) that the induced mutations are stable, i.e., not subject to repair, (*b*) that there is no important amount of intercellular selection to alter the relative frequencies of the mutant and non-mutant cells within a given individual during his lifetime, and (*c*) that radiation given at one time does not by some long-term after-effect influence the mutagenicity of cells irradiated at a later period. These questions will be considered in turn.

(*a*) Changes of a point-mutational nature induced by radiation have not shown, as a class, unusual instability as compared with those arising spontaneously. Although the possibility is not excluded that there may be a relatively short period, of the order of one or a few cell cycles, before a mutation becomes fully completed and permanent (as in the work of D. Lewis [22] on *Oenothera*), this circumstance would not in ordinary cases affect the accumulation process.

(*b*) As for intercellular selection, except for the special case of drastic lethals arising in the X chromosome of a male, which have been shown in a series of experiments with *Drosophila* (by Kossikov,[19] Shapiro,[57] Serebrovskaya & Shapiro[56], to be subject to selective elimination in spermatogonia, there is no reason to expect point mutations of the usual "recessive" sort, appearing heterozygously, to influence the multiplication or survival of immature germ cells appreciably. That mature germ cells are not thus influenced was shown long ago by Muller & Settles.[41] The most pertinent evidence on this point as regards immature germ cells, in an organism related to

* This paper is a considerably modified version of that presented at the Study Group on the Effect of Radiation on Human Heredity.

man, is given by experiments carried out by Russell [53, 54] in mice to test this very question. The failure of the mutation rate to decline in groups of offspring derived from spermatozoa ejaculated at increasing intervals after spermatogonial irradiation, shows both the absence of germinal selection against the mutant cells (point *b*) and the essential permanence of the mutant genes (point *a*).

(*c*) Direct tests of the accuracy of accumulation of lethals induced in *Drosophila* spermatozoa have been made by comparing their frequency at a given total dose after one treatment concentrated into a short time with that after a divided treatment of the same intensity and after a protracted treatment delivered at a low dose rate. It was found that the frequency depended on the total dose regardless of its distribution in time. When the diverse experiments of this kind carried out by different investigators (see review by Muller[35] (p. 478), citing work of Patterson, Timoféeff-Ressovsky, Ray-Chaudhuri, Makhijani, Stern and others) are all taken into consideration together, it is found that the time-intensity relation was varied over a range of about 300 000 times without influencing the frequency of the mutations produced. Thus, a dose delivered in divided or protracted form over a period of a month was as effective as one of the same total amount given in a few minutes. Tests have also been carried out, by Kerkis,[16] by Timoféeff-Ressovsky,[64] and recently by Oster,[46] that showed an additive relation when irradiation was given successively at two widely separated stages, to the immature and mature male germ cells respectively.

The reservation must be made that mutations not of the point variety, that is, those involving gross structural changes of chromosomes, which result from a combination of two or more independently produced chromosome breaks (Muller[28, 29]), do, as expected, show an increase in frequency when the radiation is delivered in more concentrated form, provided union of the broken ends of the chromosomes can occur to an appreciable extent during the time of the longer treatment. This condition does not hold in mature spermatozoa, the type of cell used for most of the timing experiments mentioned above, for union of broken ends cannot occur during this stage (Muller[29]), but it does hold in other germ cells, in which, therefore, more lethals of the structural type result from concentrated than from very protracted or divided treatments (Herskowitz & Abrahamson[11]). In the experiments cited in the preceding paragraph in which both immature and mature male germ cells were used, this matter was not put to the test, since the intervals between irradiations were long enough to avoid interactions between the effects of different exposures.

On the other hand, in gonial cells, which allow union of broken ends during treatment, relatively few of the mutations are of the "structural" type anyway. Moreover, low doses or dose-rates, such as those ordinarily encountered in human occupational exposures, produce relatively few

structural changes as compared with point mutations even in the cells (spermatids and spermatozoa) most susceptible to their production, and produce still fewer in gonia. It must further be noted that at these low doses or dose-rates the rare structural changes which do occur must in most cases have had both or all of their constituent breaks arising as effects of the same fast particle. The frequency of these changes would therefore, in such cases, be independent of the time distribution of the irradiation. For these reasons, conditions would seldom be encountered, except in oocytes, that resulted in over-all frequencies of mutations (counting, together, both those of a point and those of a grosser nature) differing perceptibly from those expected on an additive relation to the radiation dose. And when point mutations only were considered, the relation would be accurately additive.

Linear Relation to Dose

Another expression of this additive relation, in the case of point mutations, is shown by the linear dependence of their frequency on the radiation dose. That lethals induced in *Drosophila* spermatozoa do vary in frequency in this way has been abundantly shown for moderate and low doses, at which most of them are point mutations, in a great array of investigations, beginning with those of Hanson & Heys[10] and of Oliver[44] and proceeding through many others to those of Uphoff & Stern,[67] which brought the dose down to 50 and 25 roentgens (r). In experiments involving a lesser range of dose applied to spermatozoa of *Drosophila*, visible, non-lethal mutations, which include fewer structural changes than lethals, were found by Timoféeff-Ressovsky to show a linear relation to dose, and a linear relation for them was likewise found by our group at Indiana University when appreciable structural changes were excluded by cytological examination. Russell has also found a linear relation for visible mutations resulting from the irradiation of the spermatogonia of mice with moderate doses. A linear relation for visible mutations in higher plants was found by Stadler[59] and in lower plants, for moderate doses, by Hollaender and others (see review previously cited[35]).

It is true that in occasional experiments with very low doses results different from those expected on a strictly linear relation have been obtained. For instance, too few induced lethals seemed to be obtained by Caspari & Stern [4] and too many induced visibles by Bonnier & Lüning.[2] However, these experiments were carried on at dose levels so low that small sources of error had a relatively great effect. These sources of error include, in the case of visible mutations, differences in the degree of adverse selection against the mutants as between the control and the treated series, caused, for instance, by differences in the degree of crowding. In the case of both lethals and visibles, the numbers of mutations obtained at these doses are

so low as to have a relatively large statistical variation. Moreover, the proportion of those obtained which were induced by the radiation is subject to a far greater error still, since it is represented by the difference between the frequency found at the low dose and that found in the control material. Inasmuch as at doses of 25 and 50 r the spontaneous (control) frequency may be a good deal higher than the induced frequency, the error of this difference may be relatively enormous. This is especially true because the spontaneous frequency itself is subject to much more variation than that of random sampling. One source of such variation lies in the origination of mutations in clusters of common origin, caused by mutations in early germ cells. Another lies in the great differences between the spontaneous mutation rates existing in different lines, which may be as great as one order of magnitude and give evidence of being caused by genes (Muller[26]), now called "mutator genes". Finally, both the spontaneous and the induced mutation rates vary considerably according to the history of the germ cells used (for example, Muller;[30] Lüning[23]). Very special techniques are necessary for minimizing these various sources of error.

In view of these difficulties it is not surprising that experiments to test the linear relation have not yet been pushed below 25 r. At Indiana University, however, over the course of several years genetic and other techniques have been worked out which should now make it possible for significant results to be obtained at doses as low as 10 or even 5 r. Work on the necessary scale would require the co-operation of a group working for some two years and examining several hundred thousand cultures—a project that we estimate might cost some $ 18 000. We are not especially desirous to carry out the study ourselves, since even if the necessary financial support were provided the work would inevitably entail much digression from our other activities. But we should be glad to co-operate by furnishing the stocks and techniques and aiding in the supervision of the work if it were to be carried out elsewhere; and if no other suitable place could be found we would not exclude the possibility of our conducting the investigation.

The fact that the relation is linear at 50 r, and even when the irradiation of the sperm cells is protracted for several weeks, makes it very probable that it remains so all the way down to zero. For, in some of this work, it can be shown that hours must have elapsed between the traversing of a sperm cell by one ionization track and its traversal by another. If, however, the linear relation can be pushed down to doses as low as 5 r (or if at this dose the frequency can merely be shown to be more nearly proportional to the dose itself than to its 1.5 or 0.5 power), then we should be able to conclude with a very high degree of assurance that the relation was indeed linear all the way down to zero. This is because the ionizations are not produced separately, but occur in the course of the tracks of the fast ionizing particles (the released electrons). Thus the ionizations come in spurts and

a cell either gets a spurt or it does not. With very low doses, such as 5 r or less, an individual spermatozoon would hardly ever be traversed by more than one track, that is, it would not receive more than one spurt. Hence lowering the dose would not have the effect of lessening the number of ionizations in cells that received a spurt, but only of lessening the number of cells that received any spurt at all. For these very low doses, then, the mutation frequency would be proportional only to the number of cells "hit", which is necessarily proportional to the dose. Therefore we could justifiably extrapolate the results from 5 r linearly all the way down to zero. We need only make the one proviso here that the mutations produced in a cell by ionizing radiation result from ionizations or activations arising in that cell itself and not from those in the medium; and there is evidence from other work (see Muller[35]) that this is true of mutations produced by ionizing radiation in *Drosophila.*

Influence of Local Concentration of Activations

Even if we assumed the linear relation to hold all the way down to zero for X-rays and gamma-rays, this still would not mean that a given mutation necessarily results from just one ionization or excitation. For many of the ionizations and excitations are grouped together in small clusters in the course of the tracks of the fast particles, and it is possible that a cluster rather than a single quantum change is usually required to cause a gene mutation or chromosome break. At first sight it might be thought that this view is contradicted by the lack of influence of intensity changes on the dose—mutation rate relation, inasmuch as this result indicates that a given number of nearby ionizations when crowded together in time are no more mutagenic than when scattered in their time distribution. However, this inference is inapplicable to the question at issue, because the crowding attained in this way is much less than that within the minute clusters formed in the course of the track of a fast particle. That a cluster of such density is in fact more effective mutagenically than the same number of scattered activations is indicated by recent work (for example, Ives et al.;[14] Mickey;[24] Muller[33]) which seems to show neutrons to be more effective than X-rays in producing both point mutations and chromosome breaks. Other evidence to the same effect lies in the lower mutagenic effectiveness apparently shown by betatron radiation with an energy of about 15 Mev., as compared with ordinary X-rays, inasmuch as the radiation of higher energy is thought to result in a somewhat lesser amount of clustering than do ordinary X-rays (Herskowitz, Muller & Laughlin[12]). One may interpret the seemingly greater effectiveness of more densely crowded activations in terms of the Watson-Crick model of chromosome structure, by supposing that a hit on both complementary strands at nearly corresponding

points is more likely to result in a permanent alteration in the chromosome than a hit on just one of the strands. However, the dosimetric criteria used in the works cited are still open to doubt (see Zimmer [68]).

Complications at High Doses

The breakage of chromosomes by radiation complicates in more than one way, at high doses, the relation between the radiation dose and the observed frequency of visible or lethal mutations. For one thing, the ensuing chromosome abnormalities often kill the affected cells or their descendant-cells by causing chromosome bridges at a subsequent mitosis, and, short of such an effect, can lower the multiplication rate of the descendant-cells or even kill them by means of the resulting aneuploidy (the abnormal proportions existing between different chromosome-parts). This circumstance would not in itself affect the observed frequency of point mutations were it not for the fact that germ cells in different stages of the reproductive and mitotic cycles differ from one another in their susceptibility to having their chromosomes broken, and differ in a parallel manner in their susceptibility to having point mutations induced within them. At higher doses there is necessarily more killing off of the more susceptible cells, relatively to the less susceptible ones, by means of chromosome changes, than at lower doses (as well as a greater reduction in the multiplication rate of those not actually killed). Now, since the cells of the groups more injured in this way are also the ones that have had more point mutations produced in them, it follows that at high doses there is more selective elimination (or reduction in relative numbers) of the germ cells containing point mutations as compared with the unmutated ones than there is at low doses. Hence, at higher and higher doses the frequency of point mutations observed among the offspring will fall further and further short (in a relative sense) of the frequency with which the point mutations had actually been produced, and the graph of the observed results will bend down ever further from the straight line extrapolated from the data obtained at low and moderate doses.

It is evident that the more heterogeneous the susceptibilities of the group of irradiated germ cells from which the given offspring are derived, the more pronounced will the falling off from linearity be. A very marked illustration of this effect, involving only a one-and-a-half-fold increase in observed lethal mutation frequency with a four-fold increase in dose (from 1000 to 4000 r), was obtained (Muller et al. [40]) by taking offspring from copulations of *Drosophila* males that had occurred 7 to 10 days after their irradiation as newly hatched imagos. The reason the effect was here so marked was because, as Lüning's work already referred to had shown, the germ cells released during this period were at the time of irradiation

in a number of different stages, having widely different susceptibilities. Although the irradiation of a completely homogeneous group of germ cells would, theoretically, fail to give rise to any such effect, this has so far, in *Drosophila*, remained an ideal situation that has probably not been obtained in practice.

Even gonial cells are of differing mutagenic susceptibilities, depending, for one thing, upon whether or not they happen to be in mitosis at the time of irradiation. As Oster [45] has shown, gonial cells containing the condensed chromosomes of mitotic stages (produced in this case by colchicine or acenaphthene treatment) are, like other cells with condensed chromosomes, more susceptible to radiation mutagenesis. This fits in with Russell's finding that the mutation frequencies observed on examination of mice derived from irradiated spermatogonia, although linear for the dose range 300 r to 600 r, fell markedly below the expectation for linearity when a dose of 1000 r was used.

In organisms such as *Drosophila* and, probably, moulds, in which mutations of visible of lethal expression can arise in connexion with gross structural changes or chromosomes, either as position effects or as deficiencies, the complication exists that the frequency of these structural changes rises more rapidly than the dose (approximately as its 3/2 power [28,29]). The observed mutants, unless analysed for gross structural changes, will represent a mixture of these and point mutations (the latter in turn consisting of gene mutations and minute structural changes, both of which vary linearly with the dose). Thus at lower doses, where the great majority of the mutations are in the point category, the frequency will be linearly related to the dose, but at high doses, where the gross structural changes become numerically important, it might be expected that the over-all frequency of lethal and of visible mutations would gradually rise, to approach the 3/2 power relation. Such a rise in frequency is seen in the results for visible mutations observed by Stapleton, Hollaender & Martin [60]) after irradiation of spores of the mould *Aspergillus*; but the offspring obtained after irradiation of mature *Drosophila* males have in most experiments seemed to show a linear relation for lethal and for visible mutations even at high doses. The explanation of this result, which at first sight seems paradoxical, is doubtless to be sought in the fact that in the experiments with *Drosophila* the germ cells used were heterogeneous enough when irradiated to result in a tendency of the frequency to fall off from linearity, in consequence of selective elimination of the products of the more susceptible germ cells, and that this tendency largely compensated for the rise above linearity that would otherwise have been produced by the ever greater relative numbers of structural-change mutants arising at the higher doses.

Because of these complications results with high doses are apt to be erratic and difficult of analysis. Thus observations with moderate doses

are better suited for arriving at an understanding of the fundamental frequency-dose relationship.[a]

Influence of Cell Type on Induced Mutation Rate

It has long been known (see, for example, Stadler;[59] Muller[27]) that cells of different types or stages differ considerably in their susceptibility to mutagenesis by ionizing radiation. Althourgh gross structural changes of chromosomes show the most variation in frequency with cell type, point mutations (including what are probably changes within a gene as well as minute deficiencies and rearrangements of one to a few genes) probably have a frequency range of at least four-fold when a given dose is applied to different types of germ cells. This is to be concluded both from results on lethals arising at moderate doses (at which relatively few of the changes are in gross chromosome structure) and from visible mutations found by cytological observation to be free of discernible changes in the chromosomes.

Putting together the results of earlier and later studies (see review previously cited[35] and also recent papers by Bonnier & Lüning,[3] Telfer & Abrahamson,[62] Abrahamson & Telfer,[1] and Oster[47]), whe find that the early germ cells and gonia have the lowest frequency of induced point mutations yet the highest ratio of point mutations to changes of any kind that can be demonstrated to be structural (i.e., in these cells the structural changes fall to a minimum which is relatively much lower still). At these stages, the mutation frequency and distribution of types is much the same in male and female. In the later male germ cells, the over-all mutation frequency, including that of recessive lethals, rises to a sharp maximum during the period of spermatid formation and transformation (although we must omit the preceding meiotic stages from consideration here as not being well enough known in this respects). Lüning has given reasons for inferring that much or all of the exceptionally high frequency of recessive lethals induced in the spermatid period involves those connected with gross and minute structural changes of chromosomes rather than true gene mutations. The over-all mutation frequency, including that of recessive lethals, then falls sharply from the spermatid period to a second minimum in the immature spermatozoa (a minimum not nearly as low, however, as the preceding one in the gonia), only to rise again within the next few days until the time of ejaculation. After insemination, within the reproductive tract of the female, the male germ cells attain, and maintain at a relatively constant level, their highest known frequency of recessive lethals as well as of demonstrable structural changes, except for that found in the spermatids.

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a) Since the foregoing was written, C.W. Edington has reported finding, in *Drosophila*, the expected rise above linearity at higher doses (see *Genetics*, 1956, 41, 814). — H.J.M., 30 April 1957.

In rodents, the fact has long been known that ionizing radiation has a far more damaging effect on the genetic material when applied to mature or nearly mature male germ cells than when applied to immature ones (gonia), as judged by the killing of the resulting embryos. It remained for Snell [58] to provide evidence that these effects, and the inherited "semi-sterility" which he found also to be induced in mice, were caused by gross structural changes of chromosomes, a class of effects with which we are not primarily concerned in this paper. Later, however, evidence was obtained by Hertwig [18] that at these same stages there is also a relatively high frequency of production of point mutations by ionizing radiation, just as was known to be true in *Drosophila*. Fortunately, in man, the period during which the germ cells of the male remain in the gonial stage is over a hundred times longer than that of the spermatid and spermatozoon stages, so that the high susceptibility of the latter stages presents a relatively minor practical problem. Thus it is the less mutable gonia of mammals, studied mainly by Russell, which are of greater interest in assessing the genetic damage produced by radiation in human populations. As noted earlier (see page 31), however, gonia themselves do not constitute one homogeneous class so far as susceptibility to mutagenesis is concerned, but may differ considerably, according to their developmental and mitotic stage, and perhaps also their physiological condition.

As for the female germ cells, the point mutation frequency in the late oocytes of *Drosophila*, during the last three or four days before ovulation, attains a level almost as high as that in the nearly mature unejaculated spermatozoa, when high doses of radiation are used (Muller, Valencia & Valencia [43]). However, in the previously mentioned work of Herskowitz & Abrahamson it was found that lethals induced at this stage show dependence on a higher power of the dose than 1, and on the timing of the dose, as well as other peculiarities, all indicating that a high proportion of them consists of small structural changes involving two independently produced chromosome breaks. These mutations (like many of those induced in spermatids and spermatozoa), although not strictly point mutations, must usually be classed with them operationally, since the making of the distinction is commonly impracticable or even impossible.

In mammals the germ cells of females may, according to one view, remain for a long time in a stage corresponding to the late oocytes of *Drosophila*. It will therefore be important to determine to what extent mammalian female germ cells follow similar principles to those of *Drosophila* late oocytes in regard to induced mutations. If they remain long in such a stage, we should have to admit a notable departure from linearity for female germ cells. Whatever the answer may be, however, it is to be expected that for low doses, such as those received in most occupational and diagnostic exposures, the frequency would be linearly proportional to dose even in late oocytes (because any given mutagenically sensitive region

is so seldom traversed by more than one track), and that the frequency for a given low dose would not be lower in them than in gonia.

That somatic cells, like germ cells, can have point mutations induced in them by ionizing radiation was first shown by Patterson,[48] using *Drosophila* embryos and larvae. Calculations which the writer made on the basis of Patterson's early results, confirmed by studies by Timoféeff-Ressovsky [43] and, more recently, by Lefevre,[30] show that for given genes the frequency of point mutations is similar to that obtained for gonia, though perhaps somewhat higher. This point is of importance in considerations of those effects of radiation on the exposed individual himself, such as leukaemia and other malignancies, which might have their basis in point mutations of his somatic cells.[a]

With the development by Puck and his co-workers of methods for culturing and subculturing human somatic cells like micro-organisms, for finding and breeding lines of mutant cells,[51] and for determining the effects of different doses of ionizing radiation,[50] the way has now been paved for carrying forward to man the exact study of the induction of point mutations and other genetic changes in somatic cells. From this study, some evidence has already been adduced (Puck & Marcus [50]) that the killing effect of the radiation on the cells is, as was to have been expected, caused by chromosome structural change rather than point mutation. It is probable on a number of grounds that this genetic killing of individual cells and genetic impairment of others, caused by gross chromosome changes, lies at the root of much of the damaging effect of radiation on the body of the exposed individual, such as epilation, leucocytopenia, destruction of the intestina, tlining and other manifestations of radiation sickness, production of cataracts, retardation and distortion of growth, reduction of regenerative capacity, and—probably the most important effect—reduction of the life-span (see discussions by Muller,[32 38] Quastler,[52] and Sacher [55]).

Estimation of Total Damage from Point Mutations

The prime questions regarding the damage done to posterity by a given amount of radiation are: what will the total amount of that damage be, and how will it be distributed? In the previous sections we have discussed how the frequency of lethal or visible mutations varies with dose and with types of cell, but we have not considered the absolute frequency of such mutations for any given dose, still less the total frequency of mutations of all kinds. It is this total frequency that counts. For, as shown long ago by Haldane [9] and developed later by Muller,[31] in a population at mutational equilibrium

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a) For a recent treatment of radiation-induced leukaemia from this viewpoint, involving a calculation of its frequency per roentgen, see Lewis, E.B. (1957) *Science*, 125 (in press). — H.J.M., 30 April 1957.

(i.e., a population in which about as many mutant genes are dying out in each generation through death, or failure to reproduce, of the individuals containing them as are arising anew through mutation) the average reduction in fitness of an individual lies between the total frequency of all detrimental mutations, counting equally those with large and those with small effects, and twice that frequency. If all the mutant genes were strictly recessive, the lower figure (the mutation rate, μ, itself) would apply, whereas if they were all dominant enough to be eliminated as heterozygotes the figure would be twice this (2μ). As Muller [31] pointed out, there is good reason for assuming the higher figure, 2μ, to be nearly correct both in *Drosophila* and in man. This same figure for reduction of fitness would on the whole express the proportion of individuals in the population who would have to suffer "genetic death" (selective elimination by death before maturity or failure to reproduce) to maintain the genetic equilibrium. Some reduction of the figure for the elimination rate (probably by not more than a factor of 2) might, however, have to be made to allow for some synergistic operation by detrimental genes: a mode of action giving individuals with multiple defects a lower survival rate than the product of the survival rates of those with the separate defects.

In estimating this total mutation rate for practical purposes only point mutations need usually be considered, since the great majority both of spontaneous mutations and of those that would be likely to be produced by radiation in a human population are of this nature. The first approach towards determining the total mutation rate in any organism was made independently and simultaneously in 1934-35 by Kerkis (working in collaboration with the writer) and by Timoféeff-Ressovsky, using descendants of irradiated *Drosophila* males. [16,65] Special techniques were used for the detection of mutations which have neither a visible nor a fully lethal effect, but only reduce the expectation of survival to maturity: the so-called "detrimental" mutations. Both studies showed that these detrimental mutations arose some three to four times as frequently as the fully lethal mutations. Essentially similar results have recently been reported by Käfer,[15] working under the guidance of Hadorn, and Falk,[7] working under the guidance of Bonnier.

It is admitted by all these investigators, however, that with their techniques there was little chance of detecting mutations that reduced survival up to maturity by less than some 5-10%. Moreover, there must be many mutations, undetectable by these techniques, the detrimental effect of which occurs mainly after maturity is reached or which affect reproductive capacity rather than individual survival. Thus the estimate that in *Drosophila* there are some 5 times as many harmful mutations altogether as the number of lethals, and some 30 times the number of sex-linked lethals, is a bare minimum, possibly only half the true value. It now becomes of great importance to extend the range of detected

mutations to those with still less effect, and with other types of effect, so as to throw light on the extent to which the present estimate should be raised. As in the case of the proposed investigation of low dosage, we have for some years been developing techniques for carrying out such a study in *Drosophila*, but again the work would necessarily be on so large a scale that team-work and considerable expenditure (comparable in magnitude with that for the low-dosage project) would be required.

In absolute numbers the above estimate becomes for a dose of, say, 100 r applied to the spermatozoa of young *Drosophila* males a day or two before their mating, and applied to late oocytes, about one induced mutation in every 12 germ cells or one in every 6 offspring. Thus a continuation of this exposure, applied to both sexes through many successive generations, would reduce the average fitness of the individual in the equilibrium population by about a sixth (some 17%) and would cause about one individual in 6 to meet "genetic death" in consequence of the irradiation. It can further be estimated (see below) that the total effect of spontaneous mutations in *Drosophila* is about half as great as this; that is, the given amount of radiation, applied at the stages specified, would constitute about twice the "doubling dose". But it should be borne in mind that these present estimates are in both cases minimal ones.

Manner of Distribution and Expression of the Total Damage

How does this mutational damage become distributed and expressed among the descendants? The amount of damage done by any given mutant gene in a heterozygous descendant may be represented as the amount of detrimental effect it would exert when homozygous multiplied by its amount of dominance (the ratio of its effect when heterozygous to that when homozygous). Now the dominance of lethals in *Drosophila* has been found both by Stern and his co-workers (see Stern et al.[61]) and by the present author and Campbell (see Morton, Crow & Muller [25]) to average about 0.04 to 0.05, so that even these mutant genes with extreme effects would individually reduce viability in the heterozygote by only some 5%. The merely detrimental genes are suspected on theoretical grounds (Muller [31]) to have somewhat more dominance than the lethals, and there has recently been some direct evidence for this (Falk [7]); but, even when considerable allowance is made for this possibility, the effect exerted in a heterozygote by a detrimental is expected, on the average, to be less, absolutely, than that exerted by a lethal. Thus, taking individual mutant genes of all degrees, they should average well below 5% in individually lowering the fitness of the heterozygote. Since at the same time the visible effects of these genes in the heterozygote, *taken individually*, usually escape notice, it follows that the effects of mutations induced by radiation in any

one generation at a frequency comparable with that considered above would not ordinarily be observed among the next or any subsequent generation. Nevertheless, the total loss of fitness in the next generation, being about one in 6 (the minimum frequency of offspring with newly induced mutations) times, say, 1% (to take a bare minimum for their average expression in heterozygotes) would in a population of 1 000 000 entail the "genetic death" of at least 1700 individuals of that generation. Moreover, a comparable amount of damage would continue to be exerted for scores of generations.

The number of generations through which a mutant gene persists before causing genetic death is on the average approximately the reciprocal of the amount of damage it does to the heterozygote, so that the average *Drosophila* lethal in an autosome might be expected to persist for some 22 generations. However, the average persistence of a group of mutant genes is the *harmonic*, not the arithmetic, mean of the persistence of the individual mutant genes, and this value for the *Drosophila* lethals investigated turns out to be about 50 generations, though with a high error (see Morton, Crow & Muller [25]). The persistence of detrimentals must be even greater. This is the so-called "accumulation figure", which represents not only the average persistence of the mutant genes arising in a given generation, but also the average amount of overlapping, within the individuals of any given generation, of the mutant genes that arose in different generations, provided that the same mutation rate has existed in successive generations for a long period and mutational equilibrium has therefore been established. Hence if the 100 r exposure postulated above were to be applied to *Drosophila* for many generations it is to be expected that each generation would be damaged by an amount at least 50 times greater than that calculated above for the first generation of offspring (in fact, by an amount equal to 2μ or, in this case, 17%). Moreover, instead of one individual in six carrying a mutant gene induced by the radiation, each individual would contain at least $50 \times 1/6$, or at least eight of them, on the average. Thus, although the effects of the mutant genes would seldom be individually noticed, their collective effect would in the great majority of individuals be quite appreciable. It would of course tend to give a different pattern of impairment from one individual to another.

The Induced in Relation to the Spontaneous Mutational Damage

The damage caused by the induced mutations is of course intermingled with that caused by spontaneous mutations. Although the amount of the radiation-induced mutational damage is largely independent of that caused by the spontaneous mutations, it is helpful, in grasping its meaning, to compare it with that of the naturally existing mutational impairment, since a species is in a sense adjusted to the latter and since, in man, we have

a rough pragmatic familiarity with it. For this purpose it is desirable to be able to express spontaneous mutations in the same terms as those used above for induced mutations—namely, in terms of total mutation rate and loss of fitness. This is easily done, once estimates of these total values have been made for the *induced* mutations occurring at some given dose, provided only that the frequency of some particular group of mutations, e.g., sex-linked lethals, or visibles of a given collection or category (but preferably not those confined to just one allele-series), has been determined under comparable circumstances both in unirradiated and in irradiated material. For there is good reason to believe that, for point mutations, the following relation will approximately hold: total spontaneous mutations/spontaneous mutations of a given category = total induced mutations/induced mutations of the same category. Thus, if figures are obtainable for the last three terms, an estimate for the first one (the spontaneous total) can be calculated. The particular category best determined and most used for this purpose in *Drosophila* work has been that of sex-linked lethals.

Any *one* particular allele-series (or "locus") cannot be relied upon by itself for the above purpose because the frequencies of mutation of different series may not bear the same relation to one another for spontaneous as for radiation-induced or otherwise induced mutations (see, for example, Giles [8]). However, there is no reason to suspect that any broad phenotypic category or section of chromatin, or a whole group of allele-series chosen for their technical convenience, will show any consistent preference as between spontaneous and radiation-induced mutability. Experimental evidence that there is no such differential susceptibility in *Drosophila* was obtained in the observation, by Timoféeff-Ressovsky,[66] the writer (see Patterson & Muller [49]), and others, of the similar ratio of sex-linked lethals to sex-linked visibles in both unirradiated and irradiated material (especially when allowance is made for the relatively higher frequency of deficiencies and other structural changes after irradiation).

As noted earlier, the ratio of "total" mutations to sex-linked lethals in *Drosophila* when radiation is used has been estimated to be at least 30, and we may therefore, in accordance with the above formula, multiply the spontaneous sex-linked lethal frequency by 30 to obtain the spontaneous total. The problem arises, however, of what observed value of the spontaneous sex-linked lethal frequency to choose. For this value has been found to vary by at least one order of magnitude from one experiment to another according to the stocks used (Muller,[26] confirmed by later workers), and by more than half an order of magnitude according to the developmental history of the germ cells (Muller,[30] and unpublished data), not to speak of the variations caused by temperature and other environmental differences within the natural range. However, the upshot of a large number of studies of the spontaneous sex-linked lethal frequency in *Drosophila*, by different investigators, has shown that the great majority of individuals

bred at 25°C under reasonably favourable conditions, in such a manner that the germ cells used to produce the offspring do not give undue representation to those with extreme developmental histories, have a sex-linked lethal frequency averaging about 0.1% to 0.2%. This is true in both sexes, but the female value appears to vary less with germ-cell history and commonly to approximate 0.17%, whereas the male value, which is higher (0.2%) for the sperm released very early, is a good deal lower (e.g., 0.06%) for those released in what might be called the prime of life. Taking 0.14% as a reasonable average and multiplying it by 30, our minimum figure for the total spontaneous mutation rate per gamete is 4.2% and that for the zygote is 8.4%, a figure which also represents the average reduction in fitness or risk of genetic death as a result of spontaneous mutations. It was on the basis of this estimate that an irradiation of 100 r given to *Drosophila* in the manner specified earlier (see page 36) was there stated to constitute about twice the doubling dose, inasmuch as it had been calculated to give an induced rate of 17% per zygote.

From the above it will be seen that, in *Drosophila* at least, there is much more uncertainty about the amount of spontaneous mutational damage, because of the high variability of the spontaneous mutation rate, than about the damage caused by any given amount of radiation applied to a known stage or group of stages. Because of this uncertainty, determinations of the spontaneous mutation rate of any particular category of mutants in *Drosophila*, such as a given group of "visibles", should always, in order to have significance in relation to other work, be accompanied by a yardstick indicating the general mutability characteristic of the material studied. At present the most convenient such yardstick is to be found in the sex-linked lethal rate, which must be ascertained under precisely the same conditions. Only when such a yardstick is provided can we, for example, use data on the frequency of spontaneous mutations of given types to estimate the ratio they bear to the total mutation frequency, or to the frequency of some other particular category, inasmuch as these other quantities themselves are properly expressed in relation to a corresponding yardstick.

It is true that the radiation-induced rate also varies to some extent according to the stocks used (see below), the environmental conditions, and the germ-cell stages involved. These differences, however, are not usually likely to throw out our reckoning nearly so much as in the case of spontaneous mutations, since we have more knowledge of how they may be allowed for. But they must be taken into account.

Species Differences and the Problem of Extrapolation

In view of the evidence already referred to of the variation in the radiation-induced frequency of point mutations in *Drosophila* according to the type of cell irradiated, and the abundant evidence that has been

obtained in recent years of the influence of conditions associated with the irradiation, such as oxygen concentration, enzyme-inhibitors, etc., on the frequency (see author's review [35]), it would be strange if genetic differences failed to affect the result. Indeed, Dubovsky [6] reported that some stocks of *D. melanogaster* from widely separated localities differed by a factor of about two in the frequency of lethals produced by irradiation of the male. It is true that such differences can be produced in the same stock by slight differences in the timing of the germ cells used, a fact not then realized, and that stocks may also differ genetically in their natural timing, but genetic differences of many kinds would be expected to be capable of influencing the result. In the light of these considerations, however, it is rather noteworthy that, contrariwise, even the specific difference between *D. simulans* and *D. melanogaster* was found by Kossikov [18] not to be associated with a significant difference between the induced frequencies of lethals in flies of these two kinds. This similarity may indicate that the induced frequency, like the spontaneous one (see below), even though readily altered, tends to be maintained at a certain level by some active selective processes operating on features that, perhaps as a by-product, tend to maintain susceptibility to these mutagenic factors at the level found.

However that may be, it is not to be expected that widely different species, such as those of different phyla, would have similar induced or spontaneous mutation frequencies, either total or of any giver over-all phenotypic class and/or chromosomal type (such as sterility mutations or sex-linked lethals), nor that they would have a similar ratio of total mutation rate to mutation rate in such a category. One reason for this disparity is that the amount and distribution of the genetic material must differ enormously as between such organisms; another is that the processes whereby the genes reach expression must be so different that a superficial resemblance in effect would provide little or no indication of a homologous genetic basis. Thus even if the frequency of production of, for example, sex-linked lethals were known in a mammal, one certainly would not be justified in multiplying this figure by the *Drosophila* factor of 30, to estimate the total frequency of induced mutations in the mammal.

The case is, however, different when we use as our index of relative mutation rates in two widely different species a category consisting of the average frequency of origination, in each species, of members of a single allele (or pseudo-allele) series, often called the "specific-lotus rate", provided that this average has been determined through observations of a number of different series ("loci") in each species and that most of the values found for the different series of the same species show (as they have done) a tendency to be clustered within about one order of magnitude. The reasonable agreement between the results for some 12 different allele-series involving visible point mutations (including those that are at the same time lethal) after irradiation of the spermatozoa of *Drosophila* (Muller, [34] and

unpublished data) and also for some 7 series after irradiation of the spermatogonia of mice (Russell;[53, 54] Kimball [17]), justifies us in speaking of an average or modal induced mutability for such an allele-series in each species. We may then infer that differences in the detectability of the mutations of the different series, in the complexity of the genetic regions concerned, and in their actual mutability, are usually insufficient to cause inordinate discrepancies between the values for the different series.

In *Drosophila* the ratio between the "total" and the average single allele-series rate is at least 10 000 (for example, Muller [36]) and is probably a good deal higher. This value has been obtained by multiplying the ratio of "all" detrimentals and lethals to sex-linked lethals by the ratio of the latter to the average single allele-series frequency. (These two constituent ratios have of course been obtained in different experiments, under different conditions.) Are we now justified in assuming that a mammal would have at least as high a ratio as a fly of the "total" to the average single allele-series rate, and may we therefore multiply the latter rate, as determined in Russell's irradiation experiments, by 10 000, to obtain a minimum value for the total induced mutation rate in mice?

The justification for this procedure lies almost entirely in general considerations. The main consideration is that a mammal, by no matter what criterion, stands at least as high in the scale of biological organization as a fly, and probably a good deal higher as judged by its complexity of gross and histological structure, physiology, and behaviour. It would therefore be surprising if the genetic basis of the mammal were not at least as complicated and, accordingly, compounded of as many parts (such as nucleotides) as that of the fly. This would imply also that it had at least as many, and probably more, different ways of mutating, and that any one allele-series, on the average, represented no larger, but probably a smaller, fraction of all the mutational potentialities in the case of the mammal than in the case of the fly. The several times greater DNA content of the mammalian than of the *Drosophila* chromosome-set tends to support this inference.

It is to be noted that this method of obtaining a minimum estimate of the total induced rate in the mouse avoids any assumptions regarding the means of defining the limits of a gene or locus, and the number of such entities. It is true that in the past the argument has usually been stated in terms of genes or loci (but see Muller [36, 37, 39]), but this has, for the present writer at least, been only a short-cut mode of expression. For, what was meant by the "specific locus" frequency was really the frequency with which mutations arose that were on operational grounds to be classed as probably being members of the same allele-series, without assumptions being made as to what proportion of mutations actually occurring in the chromosome region in question would fall into the given allele category. Moreover, although 10 000 was sometimes stated to be a minimum value

of the number of genes or loci, as estimated by several very different methods, the justification for using it also as the ratio of total mutations to mutations in one average allele-series ("specific locus") was that, empirically, the experiments on detrimental mutations, lethals, and allele-series mutations had shown this ratio to hold, irrespective of the number of genes or the way in which they were defined. It is quite possible, for instance, that some of the same chromosome regions that gave rise by mutation to members of a given visible allele-series also gave rise to lethals and/or detrimentals (which may or may not have been included in the count of the allele-series frequency, according to whether or not they also produced the visible effect that served as the criterion), but this was irrelevant to the determination of the ratio since all the lethals and detrimentals of sufficient detectability to be recorded as such were included in the measurement of the frequency of these classes and therefore in the "total" rate. Thus the only relevant questions concerning the validity of the extrapolation process for obtaining a minimum estimate are whether or not a sufficiently representative sample of allele-series has been obtained, and whether we are willing to admit the probability of the proposition that the average allele-series, as operationally defined, would contribute at least as small a fraction of the total mutation rate in a mammal as in a fly.

If we grant these points and apply our factor of 10 000 to Russell's observed allele-series rate of 25×10^{-8} mutations per r in the spermatogonia of mice, we find as our minimum estimate of the total induced frequency in this material 25×10^{-4}, which may also be expressed by saying that there is at least one mutation per germ cell for every 400 r. As for the human induced mutation rate, we can at present only say that this is what it would be if it were like that of mice, that there are no data from man as yet that are inconsistent with this, and that this rate is about one order of magnitude higher than the induced rate in *Drosophila.*

On the other hand, we do have for man, as well as for the mouse, some data that allow us to estimate the spontaneous mutation frequency for allele-series. As this matter has recently been discussed elsewhere (Muller [39]), the writer will not attempt an appraisal of the validity of this evidence here, beyond pointing out, first, that the determination for man has the advantage of being based on large-scale data that give, as it were, a cross-section of results from different genetic lines and from different ages and conditions of reproduction, and, secondly, that the results of the different allele-series agree reasonably well with each other and, what is more surprising, that their consensus agrees well with the average based on mice.

Here again, then, is evidence of the operation of selective processes that tend to stabilize the mutation rate, as was noted earlier (see page 40) in connexion with the radiation-induced rate. Even more striking evidence of this, in the case of spontaneous mutation, is the unexpected similarity

between both these human and mouse values for the spontaneous allele-series rate and that (in the neighbourhood of 0.5×10^{-5}) deduced to be characteristic of *Drosophila*. It is true that thus far there has only been one published experiment (Muller, Valencia & Valencia [42]) in which a considerable group of spontaneous allele-series rates in *Drosophila* has been directly determined and in which, at the same time, a yardstick (sex-linked lethals) was used so that the rates obtained could be converted (as proved necessary) into more typical ones. However, approximately the same figure had been reached earlier by taking the typical spontaneous sex-linked lethal rate and dividing it by the ratio found to hold between the induced sex-linked lethal rate and the induced allele-series rate. Moreover, confirmation of the order of magnitude of this value (although probably involving some reduction of the value itself) is now being obtained in another series of direct observations, checked by lethals, conducted by Schalet in our laboratory at Indiana University. In any case, such a correspondence between such different species tends to impart confidence in the estimated orders of magnitude.

When, now, the factor of 10 000 is applied to the estimated value for an allele-series in man, taking for the latter the rather conservative figure of 10^{-5}, we find that the minimum estimate of the "total" spontaneous mutation rate turns out to be 0.1 per gamete or 0.2 per individual, a value higher than has commonly been suspected to apply to our own species.

Light from Another Source

Extrapolation of the type discussed above is not the only means of arriving at estimates of the spontaneous mutation rate in man on the basis of existing data. As explained by Morton, Crow & Muller [25] in a parallel paper (see also Crow [6] and Muller [39]), several different studies of the mortality found among the offspring of consanguineous as compared with non-consanguineous matings in man agree reasonably well in giving evidence from which it can be deduced that the average human gamete carries a mutational load accumulated from past generations which, if it became homozygous, would be twice as great as needed to kill the individual bearing it at some time between a late foetal and an early adult stage. Much of this load is probably scattered among diverse mutant genes any one of which would, if homozygous, entail a relatively small risk of death. There must in addition be a considerable load of detrimental genes in the gamete that tend to cause death before or after the period studied, or that interfere with reproduction rather than with survival. Moreover, in a population living under more primitive conditions than those studied, more genes would find such expression than did so in the given populations. Finally, the individual himself carries twice as many such genes as the gamete. All in all, then, the load carried, mainly heterozygously, by the

gamete is probably (if expressed in terms of the damage it would do homozygously) as much as about 8 "lethal equivalents".

Now this rather directly measured load does not in itself tell us anything of the mutation rate per generation. However, if there are means of obtaining a reasonable estimate, by extrapolation or otherwise, of the relative amount of expression which this load actually attains in the average individual (a matter dependent upon the degree of dominance of the mutant genes and on the frequency with which occasional homozygosity occurs), we should then have a value for the average reduction in fitness. As noted previously, this would be almost equal to μ (the total spontaneous mutation rate) if the eliminations in the given population are brought about mainly through the homozygous effects and almost 2μ if the dominance is enough for elimination usually to be caused by the heterozygous effects. Now although the data from man are insufficient to allow us to set a value for the average dominance of mutant genes, there are considerations (pointed out in some of the papers cited above) that allow us to set some fairly reasonable limits to such a value. Moreover, the value found for *Drosophila* lethals lies well between these limits. It is also possible to arrive at reasonable limits for the frequency of homozygosity caused by inbreeding. If then we extrapolate by taking the value for dominance found in *Drosophila*, and at the same time use in our reckoning the human inbreeding factor, we reach a value for reduction in fitness of approximately 0.1 per gamete or 0.2 per individual. This in turn gives us, as the value for the"total" spontaneous mutation rate, $\mu = 0.1$ per gamete, as was estimated by the other method, explained in the foregoing section.

It must be pointed out that the present method involves data and methods of calculation both of which are entirely separate, as well as different in character, from those used in the other mode of attack. Although extrapolation is employed at one point in the present attack—namely, for estimating the degree of dominance—this item did not enter at all into the earlier calculation. Moreover, there seems little doubt, in the light of observations concerned with man himself (see, for example, Levit [21]), that the dominance factor in man would at least be within the same order of magnitude as that assumed here on the basis of extrapolation. If this is true, then the estimate for mutation rate arrived at here is likewise of the right order of magnitude, at least as a minimum value. A further circumstance to be taken into consideration in evaluation of the present result is that it was not realized until the calculations were carried through that they would give a value even distantly in agreement with what had been obtained by the other method, and that no attempt was made to manipulate them to obtain a satisfactory fit to expectation. For these reasons, it would seem that the present result, although itself involving extrapolation, lends material support, from an independent direction, to that arrived at previously.

Although the present mode of attack is concerned only with spontaneous mutations, the estimate of the total spontaneous rate, as well as of the total load, thereby arrived at affords an important independent possibility for gauging the total mutational damage which would be produced in a human population by radiation. Before this could be accomplished, however, there would have to be some means of determining, for some limited genetic category capable of being used as an index, the relation between the spontaneous rate and the rate induced by a given dose of radiation. Possibly somatic or tissue-culture mutations, if there were good reason to assume them to be of the point type, would be useful for providing such an index. At any rate, if it were once furnished, it would then be relatively easy to combine this information with that on the total load, derived from the results of inbreeding, so as to obtain a realistic view of the all-round and long-term meaning of a given dose of radiation.

Of course we are far from the final or exact answers concerning the total frequency of either induced or spontaneous mutations, or concerning the persistence factor, for any lower organism; and we are much further yet from these answers for man. But the ways are opening up, and there seems good reason to believe that our present estimates for man, although involving extrapolation, may with assurance be regarded as minimal ones, and of the right order of magnitude. Before this point could be arrived at it was necessary to carry out a vast amount of work in the genetics of lower organisms, and also to collect very considerable data from man, and to consider these in connexion with one another. An increasing attack along both lines will be necessary if we are to attain the knowledge we need for the adequate protection and the fostering of our most precious trust, our genetic heritage.

REFERENCES

1. Abrahamson, S. & Telfer, J. D. (1956) *Genetics*, **41**, 677
2. Bonnier, G. & Lüning, K. G. (1949) *Hereditas, (Lund)*, **35**, 163
3. Bonnier, G. & Lüning, K. G. (1953) *Hereditas (Lund)*, **39**, 193
4. Caspari, E. & Stern, C. (1948) *Genetics*, **33**, 75
5. Crow, J. F. (1956) *Eugen. Quart.*, **3**, 201
6. Dubovsky, N. V. (1935) *C.R. Acad. Sci. U.R.S.S.* (n.s.), **4**, 95
7. Falk, R. (1955) *Hereditas (Lund)*, **41**, 259
8. Giles, N. H., jr (1952) *Cold Spr. Harb. Symp. quant. Biol.*, **16**, 283
9. Haldane, J. B. S. (1937) *Amer. Nat.*, **71**, 337
10. Hanson, F. B. & Heys, F. (1929) *Amer. Nat.*, **63**, 511
11. Herskowitz, I. H. & Abrahamson, S. (1956) *Genetics*, **41**, 646
12. Herskowitz, I. H., Muller, H. J. & Laughlin, J. S. (1956) *Genetics*, **41**, 646
13. Hertwig, P. (1941) *Erbänderungen bei Mäusen nach Röntgenbestrahlung.* In: *Proceedings of the Seventh International Congress of Genetics, Edinburgh, 1939* (*J. Genet.*, Suppl.), p. 145

14. Ives, P. T., Levine. R. P. & Yost, H. T., jr (1954) *Proc. nat. Acad. Sci. (Wash.)*, **40**, 165
15. Käfer, E. (1952) *Z. indukt. Abstamm.- u. VererbLehre*, **84**, 508
16. Kerkis, J. J. (1938) *Izv. Akad. Nauk SSSR (Otd. mat.-est., Ser. biol.)*, pp. 67, 75
17. Kimball, A. W. (1956) *Amer. Nat.*, **90**, 369
18. Kossikov, K. V. (1935) *Bull. Inst. Genet. Acad. Sci. U.R.S.S.*, **10**, 189
19. Kossikov, K. V. (1936) *C. R. Acad. Sci. U.R.S.S.* (n.s.), **2 (11)**, 115
20. Lefevre, G. (1950) *Amer. Nat.*, **84**, 341
21. Levit, S. G. (1936) *J. Genet.*, **33**, 411
22. Lewis, D. (1951) *Heredity*, **5**, 399
23. Lüning, K. (1952) *Studies on X-ray induced mutations in various stages of spermatogenesis in* Drosophila melanogaster, Stockholm (Thesis)
24. Mickey, G. H. (1954) *Amer. Nat.*, **88**, 241
25. Morton, N. E., Crow, J. F. & Muller, H. J. (1956) *Proc. nat. Acad. Sci. (Wash.)*, **42**, 855
26. Muller, H. J. (1928) *Genetics*, **13**, 279
27. Muller, H. J. (1930) *Amer. Nat.*, **64**, 220
28. Muller, H. J. (1938) *Collect. Net (Woods Hole)*, **13**, 181, 198
29. Muller, H. J. (1940) *J. Genet.*, **40**, 1
30. Muller, H. J. (1946) *Yearb. Amer. philos. Soc., 1945*, p. 150
31. Muller, H. J. (1950) *Amer. J. hum. Genet.*, **2**, 111
32. Muller, H. J. (1950) *Amer. Sci.*, **38**, 399
33. Muller, H. J. (1954) *Genetics*, **39**, 985
34. Muller, H. J. (1954) *The nature of the genetic effects produced by radiation.* In: Hollaender, A., ed., *Radiation biology*, New York, vol. 1, p. 351
35. Muller, H. J. (1954) *The manner of production of mutations by radiation.* In: Hollaender, A., ed., *Radiation biology*, New York, vol. 1, p. 475
36. Muller, H. J. (1955) *J. Hered.*, **46**, 199
37. Muller, H. J. (1956) *How radiation changes the genetic constitution.* In: United Nations, *Peaceful uses of atomic energy*, New York, vol. 11, p. 387
38. Muller, H. J. (1956) *J. Amer. Soc. Safety Engrs*, **1**, 42
39. Muller, H. J. (1957) *Further studies bearing on the load of mutations in man.* In: *Proceedings of the First International Congress of Human Genetics, Copenhagen, 1956 (Acta. genet. (Basel)*, **6**, 157)
40. Muller, H. J. et al. (1954) *Genetics*, **39**, 741
41. Muller, H. J. & Settles, F. (1927) *Anat. Rec.*, **31**, 347
42. Muller, H. J., Valencia, J. I. & Valencia, R. M. (1950) *Genetics*, **35**, 125
43. Muller, H. J., Valencia, R. M. & Valencia, J. I. (1950) *Genetics*, **35**, 126
44. Oliver, C. P. (1930) *Science*, **71**, 44
45. Oster, I. I. (1954) *Excerpta med. (Amst.), Sect. XIV*, **8**, 406
46. Oster, I. I. (1955) *Genetics*, **40**, 692
47. Oster, I. I. (1957) *Modification of X-ray mutagenesis in* Drosophila: *II. Relative sensitivity of spermatids and mature spermatozoa.* In: *Proceedings of the Fifth International Symposium on Radiobiology, Stockholm, 1956* (In press)
48. Patterson, J. T. (1928) *Science*, **68**, 41
49. Patterson, J. T. & Muller, H. J. (1930) *Genetics*, **15**, 495
50. Puck, T. T. & Marcus, P. I. (1956) *J. exp. Med.*, **103**, 653
51. Puck, T. T., Marcus, P. I. & Cieciura, S. J. (1956) *J. exp. Med.*, **103**, 273
52. Quastler, H. (1956) *Modes of acute radiation death.* In: United Nations, *Peaceful uses of atomic energy*, New York, vol. 11, p. 121
53. Russell, W. L. (1952) *Cold Spr. Harb. Symp. quant. Biol.*, **16**, 327
54. Russell, W. L. (1956) *Genetic effects of radiation in mice and their bearing on the estimation of human hazards.* In: United Nations, *Peaceful uses of atomic energy*, New York, vol. 11, p. 382

55. Sacher, G. A. (1956) *Radiology*, **67**, 250
56. Serebrovskaya, R. I. & Shapiro, N. I. (1935) *C.R. Acad. Sci. U.R.S.S.* (n.s.), **2** (**11**), 421
57. Shapiro, N. I. (1936) *C.R. Acad. Sci. U.R.S.S.* (n.s.), **2** (**11**), 119
58. Snell, G. D. (1935) *Genetics*, **20**, 545
59. Stadler, L. J. (1928) *Anat. Rec.*, **41**, 97
60. Stapleton, G. E., Hollaender, A. & Martin, F. L. (1952) *J. cell. comp. Physiol.*, **39**, Suppl. 1, p. 87
61. Stern, C. et al. (1952) *Genetics*, **37**, 413
62. Telfer, J. D. & Abrahamson, S. (1954) *Drosophila Inform. Serv.*, **28**, 161
63. Timoféeff-Ressovsky, N. W. (1929) *Amer. Nat.*, **63**, 118
64. Timoféeff-Ressosvky, N. W. (1934) *Strahlentherapie*, **49**, 463
65. Timoféeff-Ressovsky, N. W. (1934) *Strahlentherapie*, **51**, 658
66. Timoféeff-Ressovsky, N. W. (1937) *Experimentelle Mutationsforschung in der Vererbungslehre. Beeinflussung der Erbanlagen durch Strahlung und andere Faktoren*, Dresden & Leipzig (*Wiss. ForschBer.*, **42**)
67. Uphoff, D. E. & Stern, C. (1949) *Science*, **109**, 609
68 Zimmer, K. G. (1956) *Acta radiol. (Stockh.)*, **46**, 595

TYPES OF MUTATION PRODUCED AT KNOWN GENE LOCI AND POSSIBILITY OF HITHERTO UNRECOGNIZED MUTATIONS BEING INDUCED

Irradiation of Animal Populations: Results and Work Needed

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It is commonly accepted as a working hypothesis that ionizing radiations do not induce new types of mutation, but only raise the mutation rates of existing alleles. The basis of this assumption is partly theoretical and partly experimental. The theoretical argument rests on the fact that all living matter is continuously exposed to natural background radiation, and always has been so exposed; therefore, it is argued, any mutation which could be induced by ionizing radiation must already have been induced by natural background radiation at some time in the past; therefore no new type of mutation could be induced by man-made radiation. The experimental data, which are now extensive, do not disprove this; but in so far as the hypothesis is essentially negative, and fails to specify the extent of either spontaneous or induced mutation, it is by its very nature not amenable to experimental test. Thus, if in some experiment radiation exposure induces mutations of a type previously unknown, this can always be explained as nothing more than a manifestation of the limited nature of prior knowledge of spontaneous mutation; conversely, if exposure fails to induce mutations of a type previously known, that can always be explained as a manifestation of the finite nature of the experimental set-up. The hypothesis that ionizing radiations do not induce new types of mutation is therefore, like so many others in biology, unprovable and undisprovable. As such, it can only be of heuristic value; the extent of its value depends on our assessment of the extent to which it may be true, and the extent to which we are willing to use it as a guide in planning future action.

In point of fact, geneticists are willing to place so much faith in its validity that this hypothesis forms the basis of all present-day estimates of the genetic hazard of ionizing radiation to man. It is therefore worth while to ask if circumstances can be visualized in which it might break down. The supposition underlying it is that man-made radiations do not differ in any essential respect from natural background radiations. In

respect of dose-rate they extend far beyond the natural range, but we have no clear evidence of dose-rate thresholds for the induction of genetic effects. So far as present knowledge goes, it seems that linear energy transfer is the biologically most important characteristic of a radiation; and in this respect natural background radiation covers the whole known range, from the sparse ionization of naturally occurring gamma-rays to the dense ionization produced by alpha particles and heavy cosmic nuclei. Thus there does not at present appear to be any obvious theoretical reason for expecting man-made radiations to induce alleles that were previously unknown. On the other hand, there is no theoretical basis for the converse supposition, namely, that ionizing radiation can induce all known alleles; in fact, there is a certain amount of experimental evidence that such radiation tends to induce especially the more extreme alleles at a locus.

It is worth noting that though this hypothesis has a theoretical basis which is probably valid for mutagenesis by ionizing radiations, the analogous hypothesis for chemical mutagenesis has none. There is no ground for postulating the natural occurrence in biological material of all chemical mutagens which might be synthesized in the laboratory. Furthermore, some chemical mutagens might be expected to have a relatively mild action, and induce subtle genetic changes, compared with the generally destructive action of ionizing radiation. Recent experimental work in this field—notably that of Fahmy & Fahmy [5]—supports an interpretation of this type. Furthermore, if (as seems probable) ionizing radiation is responsible for only about one-tenth of human spontaneous mutations, leaving nine-tenths to be accounted for, we should be unwise to ignore the possibility that chemical substances may be much more important than ionizing radiation as a cause of human mutation.

Thus far in this paper the gene has been considered only as the unit of mutation, its constancy between mutational events being implicit. But a gene is also a unit of action, its presence being recognizable only by its effect on the phenotype of an individual. Furthermore, the final effect of a gene, unlike the gene itself, may be extremely variable, depending upon the other allele at the same locus, the alleles at other loci and the mass of non-genetic factors, grouped together under the term "environment". In no two individuals are the total genotype and the total environment identical, and therefore in no two individuals can the same allele be expected *a priori* to produce identical end effects. Variability of gene expression may be very great where the end effect is putatively remote from the primary gene product, as with many morphological mutants; conversely, it may be relatively slight where the effect observed is believed to be close to the primary gene product, as with the blood-group antigens. In so far as the practices of civilization have wrought great changes in the macro-and micro-environment of man, we must suppose that they have changed and are changing the expression of many human alleles.

Many systems can be invented for the classification of human genes, and which particular system is used will depend on the interests of the user. The population geneticist is interested primarily in the biological value of a genotype. He will therefore classify alleles according to their average effect on the fitness of their carriers, that is to say, on the number of zygotes that will be contributed to the next generation by a zygote of the present generation. It will be a twofold classification, according as the allele is in the homozygous or heterozygous state. Mutant alleles are probably almost always disadvantageous to some extent when homozygous, but their action in heterozygotes may vary from severe detriment through neutrality to advantage. This fact divides them into two broad classes: those which are unconditionally disadvantageous, and those which are disadvantageous in some individuals but advantageous in others. The distinction is fundamental, for it determines the nature of the forces which will maintain the allele in the population and the frequency at which it will be maintained. An unconditionally detrimental allele will be maintained at a low frequency under the opposed action of mutation to the allele and natural selection against it. On the other hand, an allele which is advantageous in some individuals and disadvantageous in others will be maintained at a high frequency, depending on the degree of advantage or disadvantage in the various individuals. Mutation will play only a minor role, or even none at all, in determining the structure of the population in respect of alleles of this type. It is therefore of importance to any assessment of the genetical hazards of radiations to man to know whether alleles of this type are of common occurrence. Unfortunately, it is a problem of exceptional inherent difficulty, because we may expect that the more easily recognized genes will largely be among those with notably detrimental effects; and, conversely, that the conditionally advantageous genes will be mainly among those with minor effects and may, for just this reason, be difficult to recognize.

At this point I find it necessary to voice some misgivings which I have felt for a long time about one aspect of what might be called "genetical public relations". Soon after Muller's demonstration that X-rays have a mutagenic action, it was realized that they present a genetic hazard to man. At that time genes were thought of as consisting mainly, if not entirely, of common, advantageous, wild-type alleles and rare, deleterious, mutant alleles. They were unconditionally good or bad. Mutation was viewed as a necessary evil; it was something which happened, without which the species would lack the heritable variation on which future evolution depends, but it introduced into the population a load of mutant alleles which had to be eliminated by processes of natural selection. Each mutational event implied the occurrence of another mutant allele to be eliminated sooner or later through the "genetic death" of some individual if equilibrium were to be maintained.

I do not think anyone seriously doubts that this is a reasonably accurate representation of the state of affairs in respect of grossly deleterious autosomal dominant or sex-linked genes such as retinoblastoma or haemophilia. On the other hand, I think many geneticists would now doubt whether this concept is valid for more than a relatively small proportion of all human genes. Clear-cut, unconditionally deleterious oligogenes may be relative rareties. They may represent only one tail of a distribution; numerically, they may come far behind the polygenes, each with an effect so small as to be virtually undetectable by the methods of classical genetics, yet together of major importance because they regulate the quantitatively variable characteristics of each species through which evolution must largely operate. Now the outstanding feature of almost any quantitative character is that it has a central optimum; the extremes in either direction appear to be at a disadvantage, in respect of biological fitness, compared with some intermediate phenotype. The theoretical interpretation is that heterozygotes for genes affecting a quantitative character have a greater biological fitness than the corresponding homozygotes; and this implies that the mutation rate may be relatively unimportant in determining the gene frequency.

The above argument has been based mainly on theoretical considerations; but there is now a great mass of observational and experimental evidence that heterozygosity is the rule rather than the exception in wild populations. If anyone doubts it, he should re-read the writings of Dobzhansky and his co-workers on wild *Drosophila* populations, of Bruce Wallace on irradiated *Drosophila* populations and of Dunn on mouse populations; or he should try inbreeding any species that is normally cross-bred.

In the face of all this it is disconcerting to find that geneticists, when writing for the public, still often base their argument on an assertion that all mutation (or very nearly all) is harmful. I make this statement in the full knowledge that I myself use exactly the same argument when, as happens all too often nowadays, I have to give a talk on radiation hazards to an intelligent but genetically uninstructed audience. Perhaps its attraction is that it is a relatively easy argument to put over; or perhaps it is used because one can draw quantitative inferences about the genetic load due to some unconditionally deleterious human alleles, whereas at present it is almost impossible to speak quantitatively about human polygenic characters. But, whatever the reason, it is extremely important that geneticists should not blind themselves to the fact that unconditionally deleterious oligogenes may constitute only a small fraction of the human genome.

The necessity for using an argument such as this stems essentially from one fact: we know something about mutation in man and experimental animals, but we know very little about the effect on a population in which mutation is induced. We know enough to be reasonably certain that the

current theory of Mendelian populations is over-simplified and unable to accomodate some essential features of real populations; but we have not yet got a satisfactory theory to put in its place. For the present there can be only one corollary; we must have more research on the genetic structure of populations, in the hope that the nature of the facts will become clearer and will stimulate the development of a more complete theory. This theory would have to cover the origin and loss of variation in populations: its origin by spontaneous or artificially enhanced mutation and by environmental action, and its loss by natural or artificial selection.

There have been many genetic studies of wild populations. Although in most of them the object was to study the effects of natural selection, it is only rarely that direct evidence has been obtained that the effect observed really was due to this cause. For example, the spread of melanic forms of various species of moths in industrial areas has been observed for over a century; and it has been assumed throughout that the spread was due to a selective advantage of the melanic form, following an environmental change from the relatively clean agricultural to the sooty industrial economy; but it was only last year that Kettlewell [6] was able to confirm the validity of this assumption, by direct observation of the numbers of moths of the various phenotypes taken by bird predators. It has also been a characteristic of studies of wild populations that, with few exceptions, the material studied has been polymorphic. This must have been due largely to subjective selection by the investigator, since a polymorphic population holds an obvious interest which a monomorphic population lacks. Nevertheless, where an apparently monomorphic population has been sufficiently closely observed, it has often proved to be polymorphic, even though the polymorphism may have been cryptic. Obvious examples are the populations of various *Drosophila* species studied by Dobzhansky and his school (see Wallace [11]), and the mouse populations studied by Dunn. [4] Dunn's work is of especial interest, because it shows that mechanisms whereby coadapted blocks of genes could come into existence are not peculiar to *Drosophila*. The mechanism in the mouse differs from that in *Drosophila*, but the effects are the same: suppression of crossing over and selective advantage of the heterozygous genotype in which it has been suppressed, even at the cost of a high proportion of inviable homozygotes. His findings gain significance in the light of the recent demonstration by my colleague Dr Mary Lyon, using an induced translocation, that the region of suppressed crossing over is at least five times as long as the short segment marked in Dunn's experiments.

The study of mutation and artificial selection in the laboratory and of natural selection in wild populations are three approaches to a much more difficult study—namely, that of populations with mutation rates that have been enhanced by ionizing radiations or other mutagens. Nor must it be forgotten that ionizing radiations have other genetic effects besides

mutagenesis; they increase crossing over, a fact which was known before their mutagenic action was discovered and which may be of great importance in the study of polygenic systems.

So far, few have attempted to work with irradiated animal populations. History dictated that one of the first studies in this field should be of a human population; but the genetical work of the Atomic Bomb Casualty Commission was almost foredoomed to failure, in the sense that it was very unlikely that statistically significant observations could have been made, even on the basis of the most extreme assumption, namely, that all human "spontaneous" mutation is really induced by background radiation and that the doubling dose for man is consequently as low as 3 or 4 r. In the event the results were, with one possible exception, negative; but all who are concerned with planning human radiation genetic studies in the future will owe a debt to Neel and his colleagues for doing the pioneer work in this field and exposing some of the problems (Neel et al. [6]). The only other genetic studies of irradiated human populations of which I am aware are those of Crow [3] and of Macht & Lawrence; [7] in each case the irradiated population consisted of radiologists. Here also the results were, in the main, negative; and the work suffered from the further limitation that it was impossible to estimate, even roughly, the radiation dose received.

There remain the experimental studies of irradiated animal populations. Of these there have been exceptionally few; and in almost all the experimental material has been *Drosophila melanogaster*. There are two reasons for this: first, a population to be maintained under known irradiation conditions must almost of necessity be kept in the laboratory; secondly, to guard against the possible effects of genetic drift, the effective breeding population should be at least of several hundred individuals. These requirements of laboratory culture and population size can be reconciled only by limiting the size of the individual animal. Subject to this limitation, *D. melanogaster* is the obvious choice, being exceptionally well known genetically. We hope to develop techniques at Harwell for maintaining mouse populations in the laboratory, but I am doubtful whether it would be feasible to keep free-living populations of larger animals in an irradiated space. A possible solution might be to find an isolated wild colony and irradiate its habitat; this procedure would have the inherent defect, however, that one could not be sure of obtaining a truly comparable control population; and in this work controls are a *sine qua non*.

If anything were needed to show how wide is the gap between observational fact and existing population genetic theory, the few published studies of irradiated *Drosophila* populations would do it. The various writers have given up any attempt to interpret their observations in terms of gene frequencies, contenting themselves with observing what happens in their populations and attempting to interpret their observations in terms

appropriate to the polygenic systems studied. Various combinations of selection type and mutational status have been used, and various types of foundation population. Wallace [10] has observed the effects of natural selection on biological fitness in populations originally derived from an inbred strain, but now heterogeneous, which were exposed to various levels of acute and chronic irradiation. Buzzati-Traverso [1] likewise observed the effects of natural selection in irradiated populations; but here the foundation populations were inbred and the effects observed were egg-production and the incidence of the *non-spineless* phenotype due to modification of the genetic milieu in a homozygous *spineless* population. Clayton & Robertson [2] likewise used inbred foundation populations; they observed the variance of the number of abdominal bristles and the response to artificial selection for this character. Scossiroli [9] selected for sternopleural hairs in an irradiated population which was genetically heterogeneous, but which had previously been selected by Mather without irradiation and had reached a plateau.

It is too early to attempt to draw general conclusions from these experiments, but some things are clear. Buzzati-Traverso's work shows that irradiation of an inbred population can release genetic variability in a character such as egg-production, which is one of the components of biological fitness, and can thereby enable natural selection to increase fitness. Scossiroli's work shows that irradiation can release genetic variability in a heterogeneous population which has reached a selection limit, and can thereby enable the limit to be surpassed. Wallace's work shows that populations can live successfully under conditions of irradiation in which a large proportion of their chromosomes carry gene combinations which are lethal when homozygous, but that some of these combinations may be advantageous when heterozygous. The work of Clayton & Robertson shows that the amount of genetic variability arising spontaneously through new mutation in each generation is only a minute fraction, perhaps a thousandth, of that normally present in a *Drosophila* population; and that a part only of the additional genetic variability released by irradiation may be available for selection.

Just what the full implications are for human genetics it is impossible at present to assess; but two conclusions seem inescapable:

1. It is essential to extend work of this type and to cover other species, including mammals, with a much lower reproductive potential than *Drosophila*; the results might be very different in species where the female produced only ten young instead of hundreds, and selection differentials were consequently lower.

2. We have no mandate from experimental fact to extend to the whole human genome the theoretical treatment of the genetic hazard of radiations that we now apply with a fair measure of confidence to grossly deleterious

gene mutations. It follows that for the present we must limit quantitative assessment to this part of the hazard alone; and this implies that the first task of human genetics must be to identify as completely as possible that part of the social load which is due to genes in this class.

REFERENCES

1. Buzzati-Traverso, A. (1954) *On the role of mutation rate in evolution.* In: *Proceedings of the Ninth International Congress of Genetics (Caryologia (Torino)*, Suppl.), p. 450
2. Clayton, G. & Robertson, A. (1955) *Amer. Nat.*, **89**, 151
3. Crow, J. F. (1955) *Amer. J. Roentgenol.*, **73**, 467
4. Dunn, L. C. (1953) *Acta genet. (Basel)*, **4**, 139
5. Fahmy, O. G. & Fahmy, M. J. (1956) *J. Genet.*, **54**, 146
6. Kettlewell, H. B. D. (1955) *Heredity*, **9**, 323
7. Macht, S. H. & Lawrence, P. S. (1955) *Amer. J. Roentgenol.*, **73**, 442
8. Neel, J. V. et al. (1953) *Science*, **118**, 537
9. Scossiroli, R. E. (1954) *I.U.B.S. Publ., Series B*, **15**, 42
10. Wallace, B. (1952) *Evolution*, **6**, 333
11. Wallace, B. (1954) *I.U.B.S. Publ., Series B*, **15**, 67

SOME OF THE PROBLEMS ACCOMPANYING AN INCREASE OF MUTATION RATES IN MENDELIAN POPULATIONS

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Problems arising from the exposure of man to irradiation are extremely numerous. They bear on many aspects of his health and his children's health. To the extent that the original exposure—medical or industrial—aims at improving man's welfare, he benefits; to the extent, however, that the exposure does him bodily harm or induces gene mutations that will harm his offspring, he suffers.

The mutagenic effects of radiation pose problems of immediate concern to the geneticist. These problems are of three major types: the development of a theory of population genetics adequate for the formulation of predictions; the design of experiments capable of testing the theory and of supplying empirical values for various parameters; and the extrapolation of theory and experimental results to human populations.

The postulated role of mutations in Mendelian populations depends largely upon the basic concept one entertains regarding the genetic structure of populations. In the main, there are two contrasting but not mutually exclusive concepts: the first is based upon the superiority of homozygous individuals; the second, upon the superiority of heterozygotes.

The first concept postulates that individuals of the highest possible fitness are completely homozygous. Natural selection acting within a constant environment would favour these individuals and would tend to establish a population composed entirely of homozygous individuals. In such a population the individuals of each generation should, ideally, be identical, and the individuals of one generation should be identical with those of the next. Mutations in a population such as this operate to frustrate the aims of natural selection. By definition, the new mutations are deleterious and, consequently, their constant formation prevents the population from reaching the level of fitness theoretically possible. Furthermore, under equilibrium conditions, the deleterious effect of mutations on the population is a function of mutation rates and is independent of the harm done to any one individual by any one mutation. Theoretical treatments of this problem have been given by Haldane,[2] by

Crow,[1] and, in great detail, by Muller.[6] Although no one actually believes that environmental conditions are constant or that the ideal population described above actually exists, the model is nevertheless reasonable if one assumes that near-equilibrium conditions exist at any moment and that genetic changes within populations occur slowly (see, for instance, Haldane [4]).

The second concept assumes that even under constant environmental conditions the individual with the highest fitness is genetically heterozygous rather than homozygous. Furthermore, there need not be one ideal genotype, but many. An ideal population of this sort would consist of individuals as phenotypically uniform as possible consistent with the demands of natural selection, but these individuals would be genetically diverse. Similarly, individuals of one generation would not be genetically identical, even under ideal conditions, with those of the next. The selective coefficient of any gene in this type of population would be a function of the genetic situation prevailing within that population. Since the population consists of individuals of diverse genotypes, selection would be constantly shuffling gene frequencies and selective values, simply because of the uncertainties associated with the formation of chance gene combinations. The details of this model have not been developed in a way comparable with the first; one can say, however, that gene frequencies under this model are primarily a function of selection and only secondarily a function of mutation rates.

These two concepts are not mutually exclusive. It may develop that one or the other is substantially correct. It may be that for some loci one is correct while for others the second applies. It is quite probable that different species differ in their genetic structure. Finally, at different times and in different places the genetic structure of a population may shift from one model to the other.

A few remarks may be made regarding the logic underlying these two concepts of population structure. Genes and chromosomes are the means by which information is passed from one generation to the next. In some cases they are the only means; in others this hereditary information is supplemented by the "spoken" word, which allows individuals of one generation to communicate with those of the next. The first concept, that based on the superiority of homozygous individuals, stresses the accuracy of the transmitted information. In the absence of mutations and of environmental change, every individual of a generation would be supplied with precisely that information which has proved valuable in the past. There is no wastage through the formation of ill-adapted individuals. Furthermore, it is a moral system in that, under ideal conditions, every individual is his neighbour's equal. The second concept entails wastage; certain individuals must obtain hereditary information that is not perfectly accurate. In so far as this wastage can be equated with suffering (and it

certainly can be considered in this way for human beings), the second concept is morally deficient.

What arguments, then, can be mustered to support the second concept and to justify giving it serious consideration? First, to the extent that genes are semi-dominant, their frequencies are changed much more rapidly by the action of selection on heterozygous individuals than by that on rare homozygotes. Secondly, a gene that is beneficial through some semi-dominant effect need not be beneficial when homozygous; the nature of these homozygous individuals is unimportant to the population at the time selection favours the heterozygotes. Thirdly, the replacement of superior aa' individuals by equally good $a''a''$ individuals requires that the allele a'' also be advantageous when heterozygous. Fourthly, there are physiological reasons for doubting in some instances whether a single allele in homozygous individuals can actually duplicate the action of two contrasting alleles in heterozygotes.

One difficulty confronting the second concept is more apparent than real. It arises from the geneticist's inability to distinguish which of two alleles is a favourable dominant and which is a deleterious recessive (see Crow,[1] footnote to p. 285). A geneticist can detect gene effects by substitution only. Genetic changes within a population are determined by the sequence in which mutations occur. By completely ignoring the sequence of genetic change and by regarding the favourable dominant as "normal", one is forced to the absurd conclusion that the origin of each favourable dominant (or semi-dominant) lowers the fitness of the population and that the population regains its normal fitness only if the new dominant attains fixation in the population.

Finally, in reference to the first concept, the writer has mental reservations that stem from the assumed independence of the effect of the gene mutation on the population and the effect of the gene on individuals of the population. In other words, no matter how slight the deviation from the "normal" allele, the effect of a given class of mutant alleles is said to be proportional to mutation rate alone. In fact, Muller [5] mentions the possibility that small harmful mutations may be even worse for the population than fully lethal ones. The writer does not question the calculations that demonstrate this fact; he questions the assumptions upon which the calculations are based and which result in a curve with an abrupt break, regardless of how infinitesimal the effect of the mutation might be.

The problems mentioned so far lie in the realm of theoretical speculation. They are problems one meets when attempting to visualize techniques employed by Mendelian populations in meeting the demands of existence, techniques compatible with the known facts of genetics. The second large class of problems arises in connexion with the design of experiments aimed at testing the validity of theoretical models. Regardless of one's concept of the genetic structure of a population, obtaining experimental data to

verify the concept or to furnish evidence regarding certain parameters is an overwhelming chore.

Information required for the manipulation of equations under the model that stresses homozygosity includes estimations of numbers of loci, total mutation rates, distributions of mutations in terms of their effects on various components of fitness (viability and fertility in particular), the distribution of deleterious mutations among individuals of a population, and dominance-recessive relationships. Information along these lines is being gathered at the Oak Ridge National Laboratory, Tenn., USA, and at the University of Indiana, under the direction of Dr Russell and of Professor Muller, respectively, as well as at a number of other laboratories. The writer feels sure that all geneticists will appreciate the tremendous effort required to obtain this information.

In our laboratory we have taken what appears superficially to be a somewhat simpler approach: the simultaneous analysis of the genetic content of experimental populations of *Drosophila melanogaster* in terms of genes affecting fitness and measures of fitness itself. The latter measure will be required for the final verification of one's concept of population structure regardless of which of the two one entertains. The chief difficulties in this approach lie in the estimation of fitness and in determining the amount of selection required to maintain this fitness. These difficulties are compounded by the necessity to limit one's studies to components of fitness and to carry out the analyses outside the population, outside even an experimental population. In studies of components of fitness one generally assumes that these components are to some degree correlated with one another and with their sum. Robertson [7] has pointed out, though, that in a population at genetic equilibrium the components of fitness must be negatively correlated. This indicates that a technique for measuring total fitness must eventually be found if the role of mutations in populations is to be evaluated experimentally.

Difficulties associated with the determination of selection pressures within populations do not seem insurmountable at the moment. Specific genetic changes within populations offer one source of information—for instance, the increase in frequency of one particular mutation, the establishment of equilibrium frequencies, or the loss of mutations following the cessation of irradiation. Estimations of population size shed light on the extent of inter-progeny selection; that is, one can judge whether a population exists because a few parents leave many offspring or because many parents leave a few each. Furthermore, larval mortality rates can be altered substantially without changing the adult population size to any appreciable extent; manipulations of this sort will offer an approach to the study of intra-progeny selection.

The final group of problems deals with the extrapolation of theory and experimental findings to man. The first problem that comes to mind is

the shift in emphasis demanded by the importance of man's intellect. In experimental material, "fitness" is equated with the ability to live and to reproduce; the emphasis in eugenic studies on the differential fertility existing in relation to IQ and racial origins shows that the experimental concept of fitness is not completely acceptable for human populations. Furthermore, although a long life and a full life is highly desirable, length *per se* is not all-important. Although the pertinent facts lie outside the realm of genetics, the writer suspects that the change from a 60-hour to a 40-hour working week has added more pleasurable, livable years to the average working man's life than he has lost by way of industrial and automobile accidents. These and similar problems are concerned with values; although there may be a consensus of opinion regarding these matters, there are bound to be sharp disagreements between societies and persons and even sharp changes in the views held by the same individual at different times.

Additional problems arise, too, because man is a social animal. The two concepts of populations described earlier dealt with ideal individuals with the highest possible fitness; these concepts are applicable to populations in which, with the exception of mating, there is no interaction between individuals in determining the fitness of the population. Such concepts are inadequate for dealing with populations of social organisms in which the fitness of the population is a function not only of the fitness of the individual members but also of the interaction between individuals. It would seem that before one can approach the problem of the ideal genetic architecture of populations of social organisms, including man, one would have to solve the simpler problem of the ideal constellation of phenotypes. The writer does not recall having seen such an analysis for human populations.

The next problems to be discussed concern what may be described as experimental human ecology. The central problem concerns the extent by which the visible human population, or, better, the reproducing human population, differs from the initial population of fertilized eggs from which it came. How strong are the selective forces operating within human populations? Mortality figures are available for the post-natal and late pre-natal periods. Figures are undoubtedly available, too, for the proportion of individuals who remain childless throughout life. Good data concerning the mortality of individuals in the early post-fertilization periods are not available at the moment. Lacking, too, are indications of the extent to which this mortality and sterility (effective, if not actual, sterility) are selective; random elimination, of course, is ineffective in bringing about genetic changes within populations. Haldane [3] has developed a method for estimating the intensity of selection that utilizes phenotypic measurements only; this method may prove valuable in the analysis of human populations. Other data that would shed light on the

selective potentialities of human populations are those dealing with the rapidity with which resistance to certain diseases has spread within memory of man and the effectiveness of this newly acquired resistance; this information would need to include the price in terms of mortality that the affected populations paid while selection operated. Along these same lines, it would be of particular interest to determine the factors responsible for limiting the number of children per couple in many human societies. When the average number of offspring per pair falls irrevocably below two for any species, that species can no longer replace itself numerically from one generation to the next, and extinction is inevitable. In some human communities the present average is but slightly above two. Since this average is determined by a combination of sociological and biological factors, some effort should be expended to determine the actual biological limit for the number of offspring human couples can have.

If selection should turn out to be more effective in man than we have suspected, we must nevertheless be wary of those who claim that radiation will do no harm to the human species. The rate at which mutant genes enter the gene pool of a population must equal the rate at which they leave. Mutant genes leave the gene pool by the effective elimination of individuals either through death, sterility, failure to reproduce, or a tendency to reproduce at a reduced rate. Effective elimination of individuals means, for human beings, that one individual is placed at a disadvantage relative to another; in many instances the "elimination" is accompanied by mental or physical suffering. Therefore, regardless of the ability or inability of "natural" selection within human populations to forestall extinction or to maintain the "fitness" of the population as a whole, we are still forced to the conclusion that every exposure of individuals to irradiation must be justifiable in terms of the beneficial effects that exposure confers either to the exposed individual or to the population as a whole. In the light of the known effects of radiation, it is essential that accidental or unnecessarily high exposures be avoided.

REFERENCES

1. Crow, J. F. (1952) *Dominance and overdominance.* In: Gowen, J. S., ed., *Heterosis*, Ames, Iowa, p. 282
2. Haldane, J. B. S. (1937) *Amer. Nat.*, **71**, 337
3. Haldane, J. B. S. (1954) *The measurement of natural selection.* In: *Proceedings of the Ninth International Congress of Genetics (Caryologia (Torino)*, Suppl.), p. 480
4. Haldane, J. B. S. (1954) *The statics of evolution.* In: Huxley, J., Hardy, A. C. & Ford, E. B., ed., *Evolution as a process*, London, p. 109
5. Muller, H. J. (1948) *Bull. N.Y. Acad. Med.*, **24**, 447
6. Muller, H. J. (1950) *Amer. J. hum. Genet.*, **2**, 111
7. Robertson, A. (1956) *Cold Spr. Harb. Symp. quant. Biol.*, **20**, 225

EXPOSURE OF MAN TO IONIZING RADIATIONS, WITH SPECIAL REFERENCE TO POSSIBLE GENETIC HAZARDS

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The purpose of this review is to show which generally occurring sources of ionizing radiation may at present be relevant and which irrelevant in discussing the effects of ionizing radiations on man.

We have to consider the direct effects on human tissues, as well as the indirect effects due to mutations of somatic cells or of germ cells. Mutations of the former cells do harm to the individual himself. Mutations of the germ cells may involve risks for the offspring as early as the next generation, consequently being of interest to the individual himself, or may—in the case of irradiation of a large number of inhabitants—constitute a long-term problem in the entire population.

The present sources of ionizing radiations which are of interest in connexion with the effects just mentioned include the following:

Natural sources of radiation

(*a*) Sources of cosmic radiation.

(*b*) The natural radioactive elements, particularly radium, thorium and potassium, in the earth's crust.

(*c*) The natural content of radioactive elements in man.

Man-made sources of radiation

(*a*) Radioactive material and technical arrangements producing radiation (X-ray tubes, other particle accelerators and nuclear reactors) used under such circumstances that the user is generally aware of the presence of the radiation (e.g., used in education, science, medicine and industry).

(*b*) Sources of radiation used for purposes in which, as a rule, only the specialist is aware of the presence of ionizing radiation (e.g., radioactive luminous compounds on watches and other articles for common use, television sets, etc.).

(*c*) Artificial radioactive elements distributed by man in nature.

Maximum Permissible Levels of Ionizing Radiation for Individuals and Large Populations

Before describing the different sources of radiation which contribute to a larger or smaller extent in producing the present level of ionizing radiation in man, a brief account of the maximum permissible doses recommended may be given.

The International Commission on Radiological Protection (ICRP) at its session in Geneva, in April 1956, decided to make the following additions to their earlier recommendations:

"A *controlled* area is one in which the occupational exposure of personnel to radiation or radioactive material is under the supervision of a radiation safety officer.

"For such personnel the maximum permissible levels of exposure are those specified for occupational exposure. In the case of prolonged exposure to radiation from external sources the maximum permissible levels for occupational exposure are represented by weekly doses of 600 mrem in the skin and 300 mrem in the blood-forming organs, the gonads and the lenses of the eyes."

* * *

"For any person in any place outside of controlled areas the maximum permissible levels of exposure are 10 % of the occupational exposure levels."

* * *

"When genetic aspects of the effects of radiation are considered, the dose received by the whole population is of importance. Scientific data derived from human as distinct from experimental animal populations are so scanty that no precise permissible dose for a population can, at present, be set. The available information is being assessed by the Commission and other groups including geneticists. Until general agreement is reached, it is prudent to limit the dose of radiation received by gametes from all sources additional to the natural background to an amount of the order of the natural background in presently inhabited regions of the earth."

* * *

"The recommended maximum permissible weekly doses and the modified values for special circumstances, permit a desirable degree of flexibility for their application. In practice it has been found that in order not to exceed these maximum limits and also to comply with the general recommendations of the Commission 'that exposure to radiation be kept at the lowest practicable level in all cases' a considerable factor of safety must be allowed in the design of protective devices and operating procedures. Therefore, under present conditions, it is expected that the average yearly occupational dose actually received by an occupationally exposed person would be about 5 rems and the accumulated dose in the employment period up to 30 years of age would be about 50 rems. Accordingly, the Commission recommends continuation of the present conservative practice as regards doses actually received by occupationally exposed personnel, to keep the accumulated dose as low as practicable especially up to age 30."

In the report of the Medical Research Council of Great Britain (MRC) [3] "The Hazards to Man of Nuclear and Allied Radiations", issued in June 1956, the following conclusions are drawn:

"2. *Dose levels to the individual*

(*a*) In conditions involving persistent exposure to ionizing radiations, the present standard, recommended by the International Commission on Radiological Protection,

that the dose received shall not exceed 0.3 r weekly, averaged over any period of 13 consecutive weeks, should, for the present, continue to be accepted.

(*b*) During his whole lifetime, an individual should not be allowed to accumulate more than 200 r of *whole-body* radiation, in addition to that received from the natural background, and this allowance should be spread over tens of years, but every endeavour should be made to keep the level of exposure as low as possible.

(*c*) An individual should not be allowed to accumulate more than 50 r of radiation *to the gonads*, in addition to that received from the natural background, from conception to the age of 30 years; and this allowance should not apply to more than one-fiftieth of the total population of this country.

"3. *Dose level to the population*

Those responsible for authorising the development and use of sources of ionizing radiation should be advised that the upper limit, which future knowledge may set to the total dose of extra radiation which may be received by the population as a whole, is not likely to be more than twice the dose which is already received from the natural background; the recommended figure may indeed be appreciably lower than this." [3] (p. 80)

In the report of the US National Academy of Sciences (NAS) [11] "The Biological Effects of Atomic Radiation", the following recommendations are made:

"C) That for the present it be accepted as a uniform national standard that X-ray installations (medical and nonmedical), power installations, disposal of radioactive wastes, experimental installations, testing of weapons, and all other humanly controllable sources of radiations be so restricted that members of our general population shall not receive from such sources an average of more than 10 roentgens, in addition to background, of ionizing radiation as a total accumulated dose to the reproductive cells from conception to age 30."

* * *

"E) That individual persons not receive more than a total accumulated dose to the reproductive cells of 50 roentgens up to age 30 years (by which age, on the average, over half of the children will have been born), and not more than 50 roentgens additional up to age 40 (by which time about nine tenths of their children will have been born)." [11] (p. 29)

Evidently it is generally agreed that at present it is desirable to limit the doses received by the gonads of individuals to less than 5 roentgens (r) per year and 50 r before 30 years of age, and that the average dose to the gonads of the population as a whole should be kept very low: of the order of the natural background (ICRP); twice this level (MRC); and 10 r before 30 years of age (NAS). The difference in these figures is not very important as their order of magnitude will in practice be about the same.

According to our present knowledge it seems likely that a dose of 30-80 r will (according to MRC) double the natural mutation rate in man, which is probably only to a minor fraction (perhaps about 10%) caused by ionizing radiations. The rest of the natural mutations will, to an unknown extent, be due to chemicals and to the thermal movements of the molecules. It seems to be highly desirable for the mutations induced by chemicals in particular to be investigated, in order to elucidate the relative role of radiation-induced mutations.

The recommendations made by the organizations quoted above are as regards the whole population based mainly on the natural level of ionizing radiation. It is not the place here to discuss whether this is a correct starting-point, nor whether or not the maximum permissible dose levels recommended are reasonable. They have been fixed after careful consideration based on our present, unfortunately very incomplete, knowledge of the biological effects of small radiation doses, but have been agreed to by specialists in biology, genetics, haematology, physics, and radiology with long experience in radiation protection both on the research and on the practical side.

With respect to the risks of injurious effects on man, one matter may, however, be stressed. There must always be a reasonable ratio between what can be gained by the use of ionizing radiation and the risks of its injurious effects. The use in medicine of ionizing radiation for examination and treatment of patients therefore occupies an exceptional position, which has not always been taken into account in recent years, in the discussion of the problems of the general irradiation of mankind. There must certainly be a sound balance between the benefits of a good health service and the risks to the patients with respect to onset of malignant disease or genetic damage of which, however, we at present do not know very much.

Natural Sources of Ionizing Radiations

Cosmic radiation

The cosmic radiations produce the doses shown in Table I.

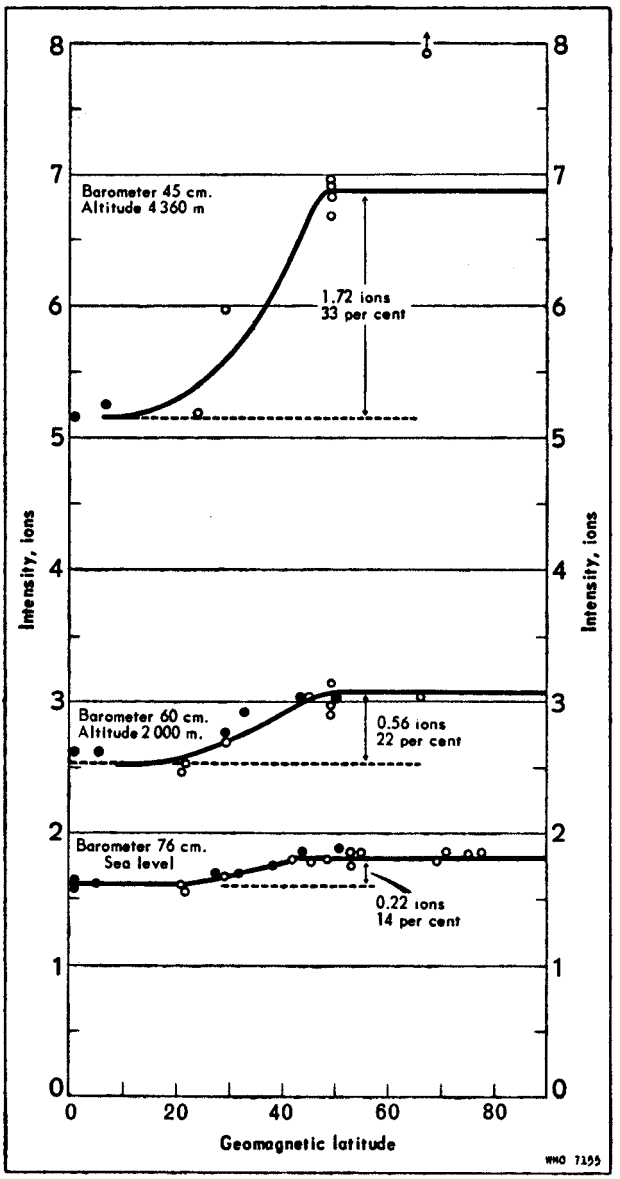
The values for 0-4000 m have been calculated from the work of Compton and co-workers (Fig. 1), taking into consideration that some reduction due to absorption may be justified indoors, and the values for 6000-18 000 m from Millikan and co-workers (Fig. 2). The values are

TABLE I. ROUGHLY ESTIMATED DOSES IN SOFT TISSUE FROM COSMIC RADIATION, EXPRESSED IN RAD PER THIRTY YEARS

Geomagnetic latitude	*Dose, in rad, at an altitude above sea level, in metres, of*						*Hours per week to accumulate a dose of 50 rem during 10 years at 18 000 m*
	0*	2 000*	4 000*	6 000**	12 000**	18 000**	
0°	0.7	1.1	2.0	8	35	40 (400) †	63
40°	0.8	1.3	2.5	12	70	110 (1100)†	25
60°	0.8	1.4	2.7	14	85	150 (1500)†	17

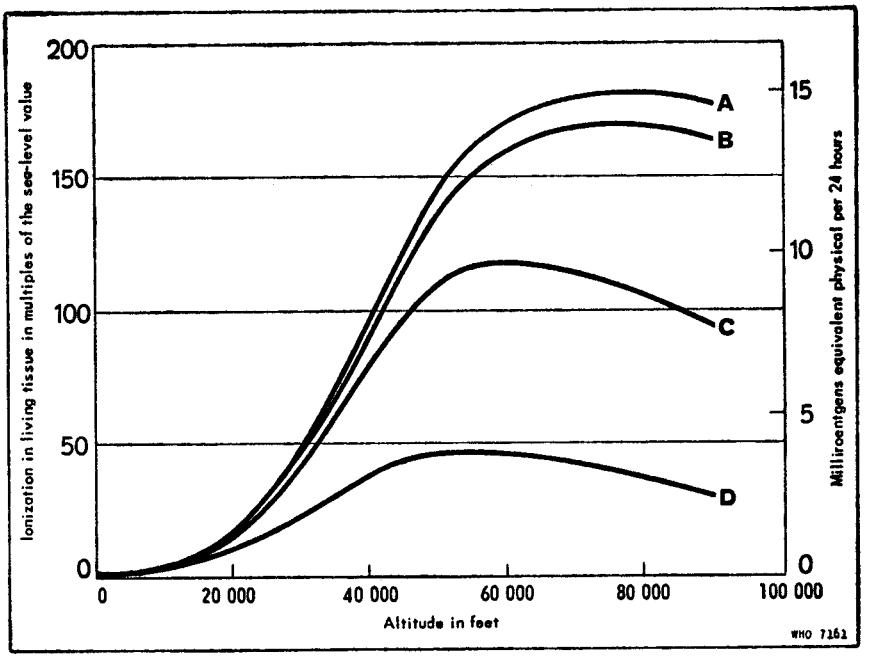
* Calculated from the measurements of A. H. Compton and co-workers (see Halliday[4]).
** Calculated from the measurements of R. A. Millikan and co-workers (see Schaefer[7]).
† The figures in brackets are estimated values in rem, assuming an RBE of 10.

FIG. 1. COSMIC RADIATION AT VARIOUS ALTITUDES, ACCORDING TO COMPTON AND CO-WORKERS



fairly approximate, as there are many factors which are difficult to allow for at the higher altitudes, especially with regard to the unknown relative biological effectiveness of heavy nuclei rays.

FIG. 2. COSMIC RADIATION AT HIGH ALTITUDES, ACCORDING TO MILLIKAN AND CO-WORKERS



A. *Geomagnetic latitude* : 60° N
B. „ „ : 51° N
C. „ „ : 38° N
D. „ „ : 3° N

Variations with time. Major variations in cosmic radiation occur during short periods only, and the few occasions associated with an appreciable increase are so rare and of such short duration that the doses caused by cosmic radiation for a certain altitude and geomagnetic latitude may, at the earth's surface, be regarded from the practical point of view as constant. A record of the variation in the cosmic radiation at six places in Sweden on 23 February 1956 is shown in Fig. 3. This is one of the occasions on which an extraordinary increase was observed. The dose due to this temporary increase was, at sea level, less than 0.03 millirem.

As to the long-term variations, it seems highly unlikely that any major variations in cosmic radiation have taken place during the past 2000 years.

Variations with site. The maximum variation between different places on the earth's surface, excluding mountains more than 4000 m high, is about 2 r per 30 years.

Doses to individuals. The doses to individuals may be of importance at very high altitudes. The present development of communication by air

FIG. 3(A). VARIATION IN COSMIC RADIATION AT ALTITUDE OF 50-500 METRES ABOVE SEA LEVEL RECORDED IN FEBRUARY 1956 AT THE PLACES I-VI SHOWN IN FIG. 3(B)

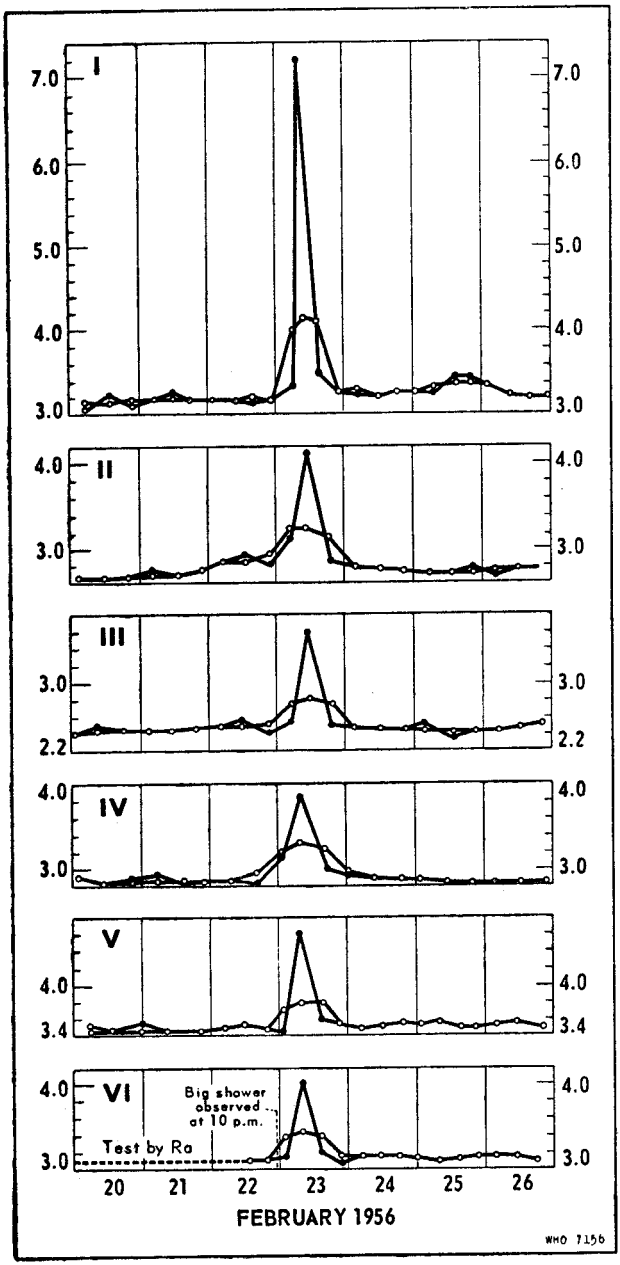
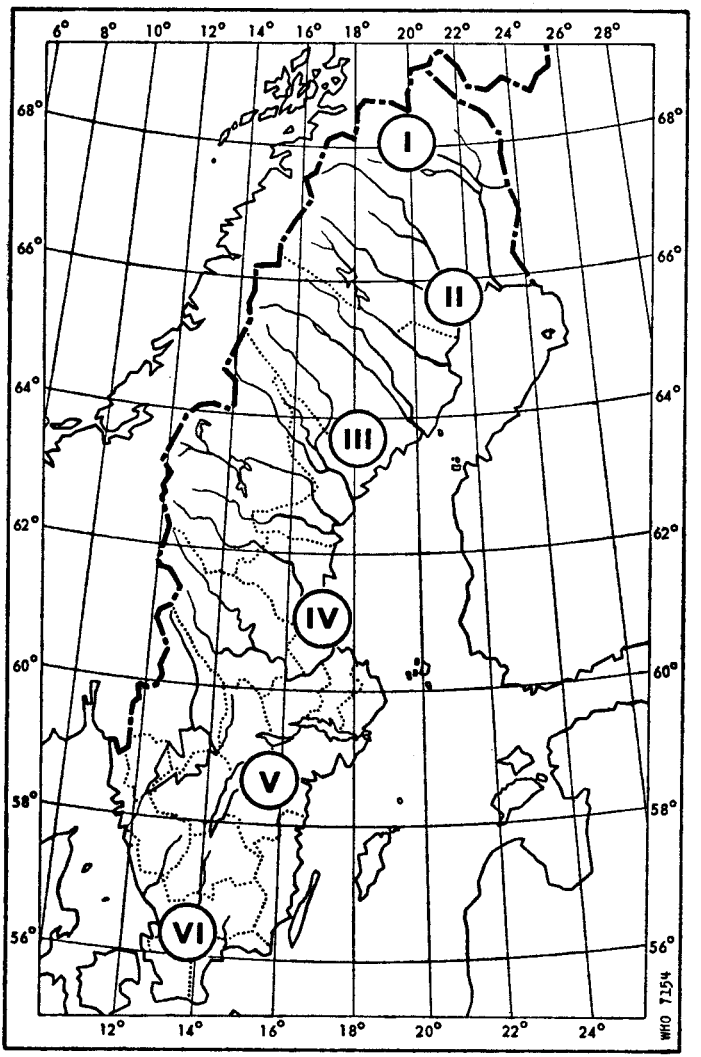


FIG. 3(B.) SITE OF SWEDISH BACKGROUND-RADIATION RECORDING-STATIONS



makes it necessary to take into account the fact that, at very high altitudes, the maximum permissible dose of 50 rem may, especially at high geomagnetic latitudes, be exceeded if on the average some 10 hours per week are spent at this altitude over a period of 10 years, which might well be possible in the future for personnel in aircraft. The increase in cosmic radiation on 23 February 1956 at altitudes of 20 000 corresponds, perhaps, to a dose of less than some tenths of 1 rem obtained during a few hours, and is therefore probably of limited biological significance.

Doses to large populations. The contribution to the irradiation of large population groups (> 100 000) varies between 0.7 and 2.7 r or approximately between 1 and 3 rem per 30 years.

The fact that an appreciable part of the radiation can be screened off by reasonable quantities of material may be of certain value for judging the risk for stratosphere and interstellar traffic. Investigations of the biological effects of cosmic radiation at very high altitudes are, however, desirable, because of the lack of knowledge as to the relative biological efficiency (RBE) values for heavy nuclei radiations.

Natural external γ-radiation

The external γ-radiation in nature varies with the radium, thorium, and potassium content of the ground and of the building material in houses. The γ-dose in free air produced above level ground can be calculated according to the simple formulae given below: [5]

	Dose, in r, per 30 years
Radium	$0.57 \times 10^{12} \times$ s (Ra)
Uranium	$0.20 \times 10^{6} \times$ s (U)
Thorium	$0.094 \times 10^{6} \times$ s (Th)
Potassium	$41 \times$ s (K-39)

in which s (Ra), s (U), s (Th) and s (K-39), are the contents of radium, uranium, thorium, and potassium in g of element per g of ground substance.

To obtain an estimate of the dose to the gonads in rad,[a] the doses in free air have to be multiplied by a factor of 0.5 for women and 0.7 for men,[5] or on the average 0.6, to account for the absorption in the shielding part of the body. The same factor may be approximately applicable to most of the other organs. For the skeleton, the factor might be considered, on the average, to be about 0.8.

The doses due to natural γ-radiation over ground containing various minerals are given in Table II, and the doses in dwellings in Sweden [5] are seen from Table III and Fig. 4. These are in good agreement with the few observations which have been made in other countries.

The γ-radiation from the ground is absorbed by snow, as seen from Fig. 5. A snow cover of 40 cm depth and of medium volume and weight absorbs about 50% of the γ-radiation from the ground.

A factor of considerable importance is the relation between the time spent indoors and out of doors. Here, it is assumed that on an average in large population groups one quarter of the life is spent out of doors.

As an additional contribution to the irradiation of man from natural radioactive elements in the earth's crust, the radon and thoron of the air

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a) 1 rad corresponds to a dose of about 1.07 r in soft tissue.

TABLE II. CALCULATED GONAD DOSES ABOVE VARIOUS MINERALS

Mineral	*Ionization (ionpairs/cm³-sec) due to content of*			*Gonad dose (excluding cosmic radiation) in r per 30 years*
	Ra	*Th*	*K*	
Igneous rocks				
Average	1.6	2.5	2.4	1.9
Granites:			3.2	
North America, Greenland	2.0	1.7		2.0
Finland	5.9	5.9		4.1
Alps	5.5	6.9		4.3
Basalts:			1.2	
North America, Greenland	1.2	2.1		1.2
England, Germany, France and Hungary	1.6	1.9		1.2
Sedimentary rocks				
Sandstone	0.4	1.0	0.9	0.8
Limestone	1.3	0.2	0.3	0.6
Alum shales in Sweden	75	0.3	3.2	21.0
Ore containing*				
1% U	—	—	—	1000
1% Th	—	—	—	500
0.01-0.001% Th**	—	—	—	0.5-5.0

* The uranium and thorium are in most cases very unevenly distributed and therefore the figures given here may be of limited practical value. According to a personal communication from Professor Z. M. Bacq, University of Liège, the background radiation in Katanga, Belgian Congo, will reach 1001-50 times the normal background.

** Travancore sand, containing monazite, according to a personal communication from J. Eklund, Geological Survey of Sweden.

may play an important role in special cases. In general, the content of these elements in the air is too small to contribute to the dose received by the human body by more than a few per cent. In some places and during some periods, however, this content can be fairly high—for instance, in rooms where water of high radon concentration is used or the ventilation is insufficient,[5] in cellars where radon and thoron come up from the earth, and in large cities during calm weather.[1] Such cases seem only occasionally to have been investigated, and it would probably be worth while to make more systematic studies in this field. At present, these sources of natural radiation are too little known to be treated in this survey and will therefore be disregarded, although it is possible that they are of significance for the irradiation of the pulmonary system of a comparatively large number of individuals living in certain areas.

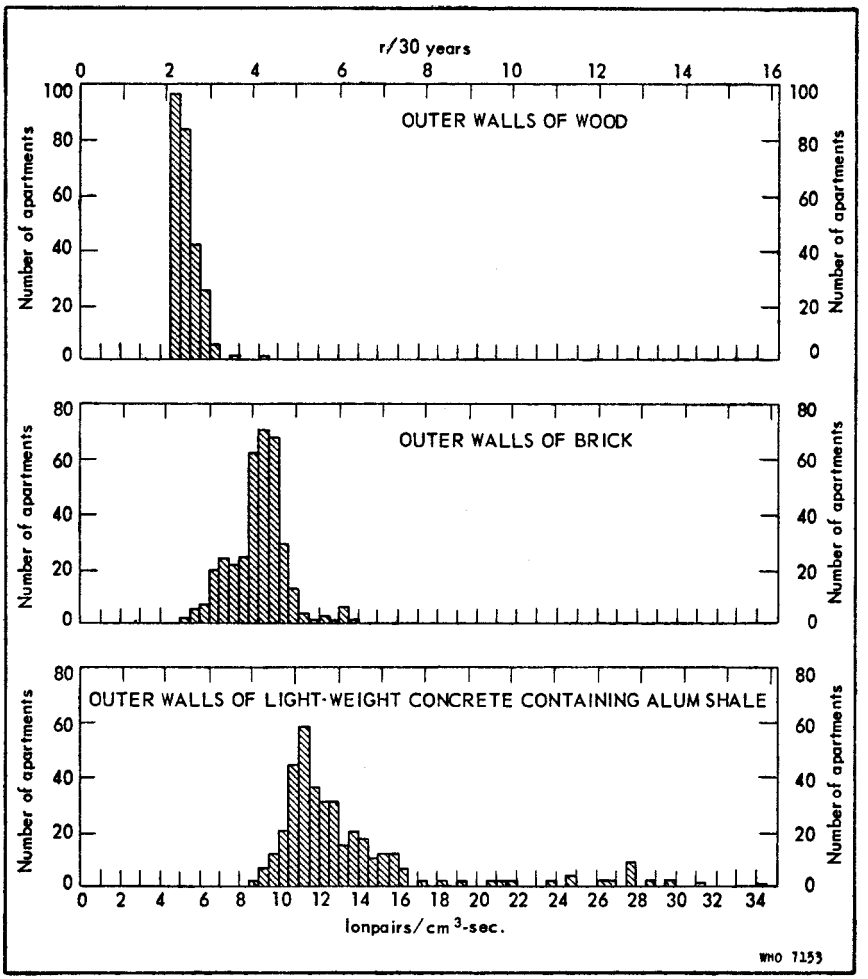
Variation with time. The average annual dose to human beings due to natural sources has probably remained roughly the same throughout the present geological period. A slight decrease in the radiation occurred when man learned to use wood for building houses, and stopped living in earthern huts or in rocks where the amount of radon in the air was sometimes probably quite high. The level subsequently increased again with the use

TABLE III. SUMMARY OF RESULTS OF GAMMA-RADIATION MEASUREMENTS IN SWEDISH DWELLINGS*

Building material in outer walls	*Mean gonad dose in r per 30 years*		
	middle of room	*highest value recorded*	*lowest value recorded*
Wood	1.0	1.1	0.95
Brick	2.0	2.2	1.9
Light-weight concrete containing alum shale	3.2	3.8	3.0

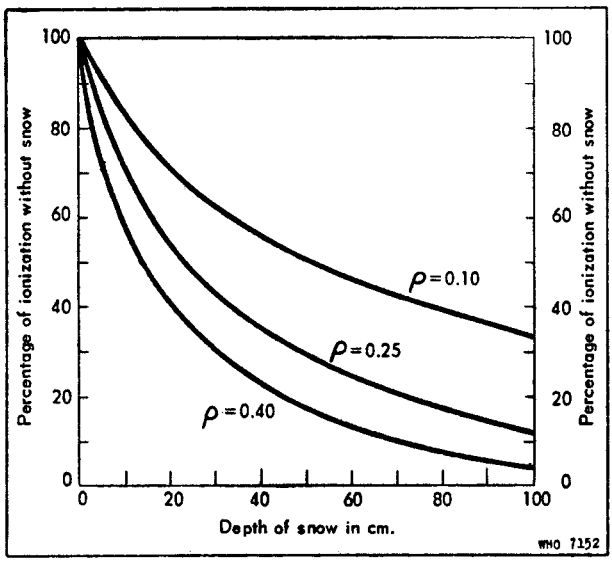
* Calculated from Hultqvist's figures; [§] cosmic radiation excluded.

FIG. 4. DISTRIBUTION OF AVERAGE RADIATION IN SWEDISH DWELLINGS OF THREE TYPES



of bricks and concrete as building materials, and when people moved to cities, where the material surrounding them more frequently contains minerals.

FIG. 5. DECREASE IN GAMMA-RADIATION WITH DEPTH OF SNOW COVER AT THREE DIFFERENT SNOW DENSITIES

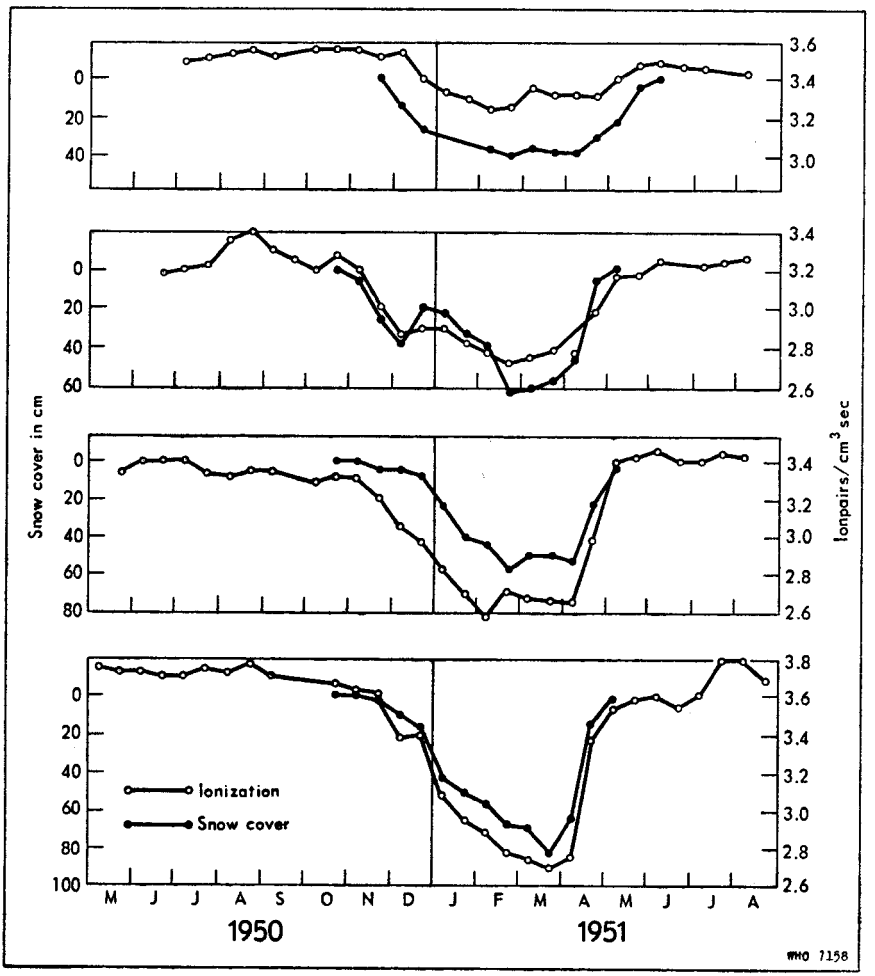


A circumstance which may have brought about a reduction in the environmental γ-radiation for some populations is that, during the ice periods, certain areas were covered with ice and snow for a greater part of the year than they are today. As already shown, snow absorbs γ-radiation from the ground, and thus appreciably reduces the irradiation out of doors, and produces a seasonal variation (see Fig. 6) in the irradiation of large population groups, especially those living in rural districts.

Variation with site. As a rule, the difference in the level of natural γ-radiation in different parts of the world is probably not very large. Even over areas containing rich uranium or thorium ores, the γ-doses to the inhabitants only in rare cases exceed a few times the normal level. This is because the ores are generally very unevenly distributed in both rocks and sands, and are often covered or surrounded by material of normal radioactivity. The inhabitants moving over the area in question might thus, on an average, be exposed to doses which are much lower than could be conceived. This experience based on observations in Sweden needs further verification, but will probably be found to apply to most population groups throughout the world.

The doses of γ-radiation to persons living in places more or less permanently covered with deep ice or snow, and to those spending most of their time at sea, are generally very small. Here, the amount of γ-radiation

FIG. 6. SEASONAL VARIATION IN GAMMA-RADIATION RECORDED AT FOUR PLACES IN SWEDEN (I-IV IN FIG. 3(B)) AND CORRESPONDING SNOW COVER PLOTTED AS NEGATIVE VALUES



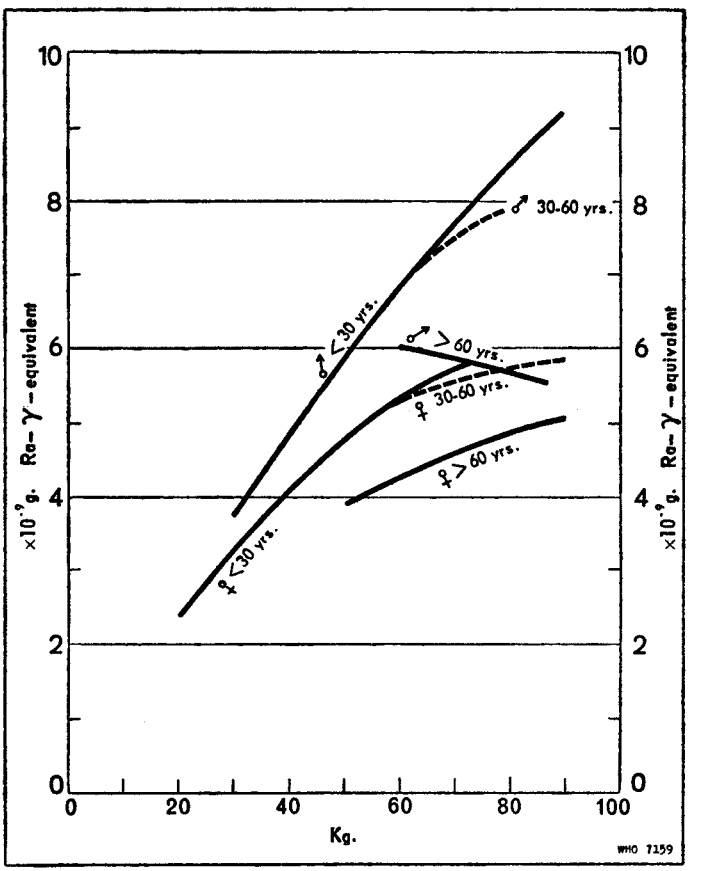
from the earth is often so minute that it can be entirely disregarded in comparison with the radiation from other natural sources. Recent investigations of the radiation level on wooden and iron vessels of different sizes have shown that the γ-radiation at a distance of only a few metres from a granite wharf is entirely negligible.

The results of investigations in Great Britain, Sweden and the USA have shown that the average values for the irradiation of large population groups due to natural sources in these three countries are as follows:

Great Britain.	2-3 rem per 30 years
Sweden	2-5 rem per 30 years
USA	4.3 rem per 30 years

In view of the statements made above with regard to the average doses received by individuals, it would be of interest to carry out long-term measurements by means of personal monitoring, in order to arrive at reliable data on the doses actually received.

FIG. 7. VARIATIONS IN GAMMA-RADIATION* FROM MALE AND FEMALE HUMAN SUBJECTS OF DIFFERENT AGES AND BODY-WEIGHTS



* *More than 95% of the radiation is due to the potassium-40 content.*

Natural content of radioactive elements in man

In areas where the radium content of drinking-water and food is not exceptionally high, the potassium content of human tissues is the main source of internal irradiation (see Fig. 7). The doses, in rad, due to the amount of potassium-40 (0.012% in natural potassium) in some human organs are shown in Table IV. With respect to some tissues, particularly bone, the data of different authors vary considerably.

TABLE IV. POTASSIUM CONTENT IN ADULT HUMAN SUBJECTS ACCORDING TO SHOHL[5] (A) AND FORBES & LEWIS[3] (B AND C) AND DOSE DUE TO K-40

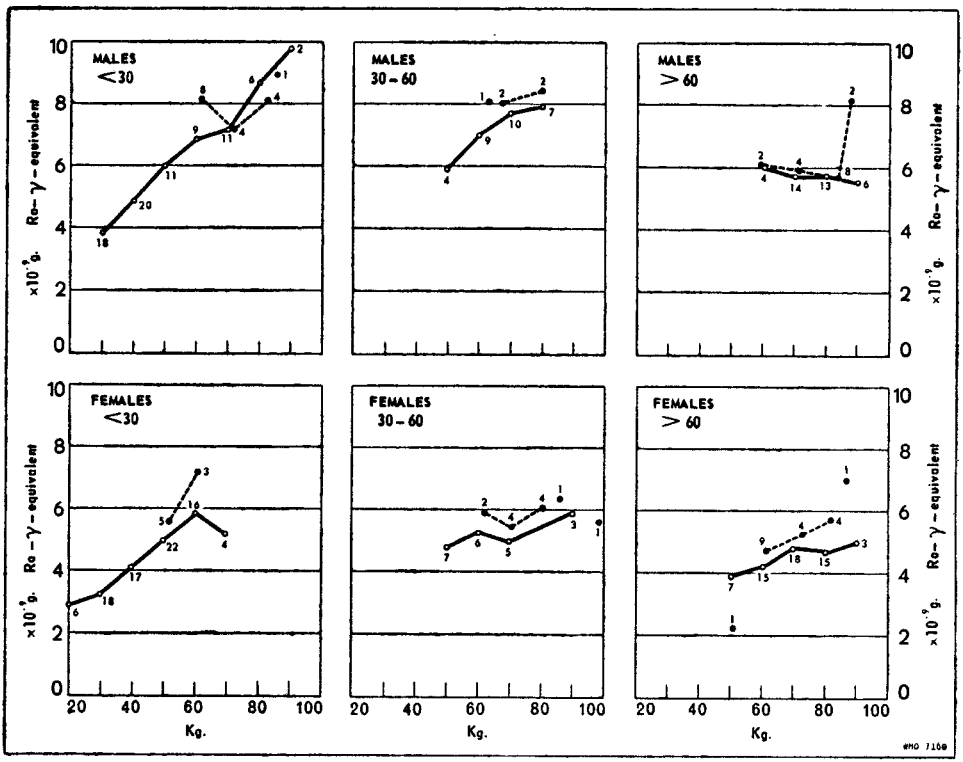
Organ	Weight in % of whole body			% K-39			Dose in organ in r per 30 years (mean of B and C)
	A	B	C	A	B	C	
Skin	7.3	6.4	6.5	0.09	0.15	0.16	0.30
Skeleton	17.5	17.5	14.7	0.055	0.10	0.11	0.20
Tibia	—	1.4	—	—	—	0.05	—
Muscle	43.0	39.5	39.6	0.42	0.33	0.30	0.62
Nerve	—	3.0	2.1	—	0.28	0.29	0.56
Liver	2.7	2.3	2.3	0.17	0.27	0.22	0.49
Heart	0.5	0.5	0.6	0.13	0.22	0.19	0.40
Lungs	1.5	3.3	2.2	0.15	0.24	0.26	0.50
Kidneys	0.5	0.5	0.4	0.17	0.16	0.22	0.38
GI. tract	—	1.8	1.5	—	0.13	0.13	0.26
Adipose	—	11.3	21.4	—	0.08	0.06	0.14
Remainder	—	11.3	6.4	—	0.18	0.17	0.34
Weight loss on dissection	—	2.6	2.2	—	—	—	
Total body	70 kg	53.8 kg	73.5 kg	0.205	0.212	0.190	0.40

The content of carbon-14 and radon contributes about 5% and 10%, respectively, of the average potassium radiation.

According to measurements made by Hursh & Gates [6] and recently by Sievert & Hultqvist [9] (Fig. 8), the radium content of the skeleton is probably less than 0.3×10^{-9} g, in areas with a radium content in the water of less than 0.2 μμg per litre. According to Spiers,[10] the mean dose to the osteocytes is about 6 rem per 30 years for 0.5×10^{-9} g of total radium body burden. The amount of radium is, however, very unevenly distributed in the skeleton, and the dose significant for the production of osteosarcoma therefore seems to be extremely difficult to assess.

The *variation with time and site* in the natural internal irradiation is mainly a question of the variations in the radium content of water and food and of the radon in the air. Referring to what has already been said, it may be stated that there are not at present sufficient data available to give

FIG. 8. COMPARISON OF WHOLE-BODY GAMMA-RADIATION OF MALE AND FEMALE PERSONS IN VARIOUS AGE-GROUPS LIVING IN CITIES WITH DIFFERENT CONTENTS OF RADIUM IN THE WATER-SUPPLY



——— *0,2 μμg of Ra per litre of water* --------- *1-2 μμg of Ra per litre of water*

any reliable figures for different areas in the world. This also applies to the problem of the natural radioactive elements taken up in the pulmonary system. With respect to these matters, reference can be made to a recent publication by Hultqvist,[5] in which an extensive bibliography is given.

The common limits of the doses ta large populations groups (> 100 000) and to individuals from natural radiations are shown in Table V.

Man-made Sources of Ionizing Radiations

Radioactive material and technical arrangements producing ionizing radiation used under such circumstances that the user is generally aware of the presence of the radiation

Here, *occupational exposure* and *exposure of patients* undergoing treatment or investigation in radiology are the two matters to be considered.

TABLE V. ESTIMATED VALUES FOR IRRADIATION OF THE GONADS OF THE POPULATION FROM NATURAL SOURCES EXPRESSED IN REM PER THIRTY YEARS

	For large population groups			*For individuals*	
	minimum	*maximum*	*average*	*minimum*	*maximum*
Cosmic radiation	0.7 (including screening in dwellings)	3? (at about 4000 m above sea level)	1?	0.5? (for some miners)	5? (50?)* (* 3% of 30 years spent at 18 000 m above sea level)
Natural radiation ¼ of 30 years out of doors	< 0.1 (above water, snow and ice)	1 (above igneous rocks)	0.5	0	15 (20?)
¾ of 30 years indoors	0.9 (in wooden houses)	3 (in some types of brick and concrete houses)	2		
Radon in air	0.03 (out of doors and in wooden houses with good ventilation) (3×10^{-13}c/1)	0.8 (in cellars and in stone houses with poor ventilation) (50×10^{-13}c/1)	0.2	<0.01 (<10^{-13}c/1)	2.0 (10^{-11}c/1)
K-40 in body (+ 0.03 for C-14)	0.5	0.5	0.5	0.5	0.5
Approximate sum for gonads	2	6 (8?)	4	1	20 (> 50?)

The doses received by those carrying out work with ionizing radiation in education, science, medicine, and industry are in most cases small, as the personnel can generally be adequately protected. Furthermore, in all work where patients are not involved, there is no reason to permit irradiation which can in any way cause ill-effects. Here, the maximum permissible levels for individuals and large population groups are exceeded in rare cases only.

In radiology, especially some procedures in γ-ray therapy and in examinations using X-rays, circumstances do not always permit entirely satisfactory protection of doctors and personnel. Here, the individual dose will sometimes be close to the maximum permissible levels, and may even exceed them occasionally.

The occupational doses contribute to the radiation per capita of whole populations an amount which in Great Britain [3] has been estimated at about 2.5 r per year as an average for about 14 000 research, medical, and industrial workers, and at about 0.4 r per year for about 7 000 people engaged in atomic energy work. Altogether, the average gonad dose per capita due to occupational exposure is estimated at 0.0016 r per year; in other words, if 10 years is assumed to be the average period of work before reproduction, the relevant average gonad dose for the whole population may be less than 0.02 r before 30 years of age. An estimate of the corresponding figure for Sweden has given a considerably lower figure.

The occupational dose is apparently throughout the world attributed mainly to medical radiology, but the figures are presumably very uncertain. It seems, however, that occupational irradiation does not at present contribute any appreciable amount to the gonad dose of whole populations.

The doses received by patients undergoing treatment and examination by means of ionizing radiations, on the other hand, are of decisive importance, since they constitute by far the largest exposure of the population to man-made sources of radiation. In France, Germany, Great Britain, Sweden, and the USA, investigations have been carried out in order to ascertain the doses to patients during various types of radiological procedure. Numerous publications are available, but up to now estimations of the present average dose to the whole population due to the irradiation of patients have been made only in Great Britain, Sweden, and the USA. The results show that the average gonad dose per capita due to irradiated patients seems to be of the order of 1-3 r in 30 years. The reliability of these estimates has been much discussed, and it seems advisable to await further investigations, based on radiation measurements and some type of sampling method, before accepting any definite figures. It is nevertheless highly probable that the order of magnitude of the figures quoted is correct, since the estimations were made independently in three different countries.

Sources of radiation used for purposes in which, as a rule, only the specialist is aware of the presence of ionizing radiation

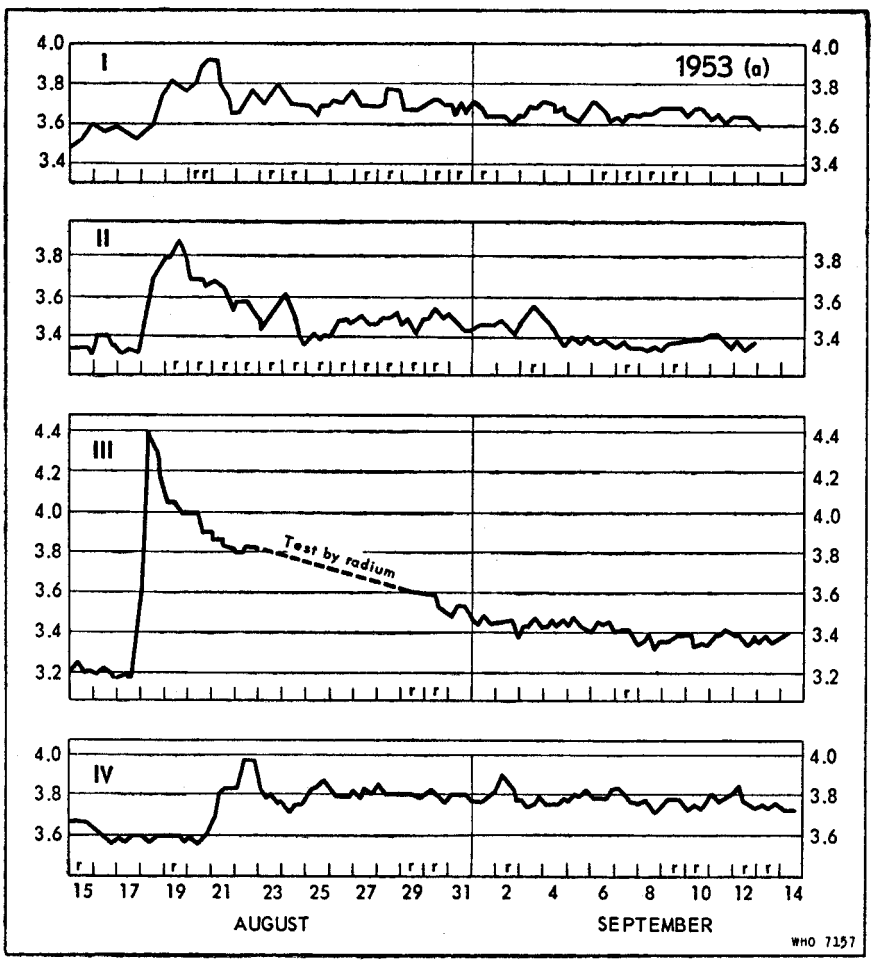
At present, we are faced in this field with only a few matters of minor significance. The average gonad dose from luminous compounds in watches is found in Great Britain [8] to contribute 0.01 r per year to the average gonad dose, and the radiation from television sets contributes a still more insignificant dose.

In the future development of atomic energy it seems highly probable, however, that the use of radioisotopes for various purposes will change the situation, and that the resultant distribution in the community of a large number of small radiation sources, each completely harmless individually, but collectively raising the level of irradiation of the population, will give rise to a new problem.

Artificial radioactive elements distributed in nature

The World Health Organization and its Study Group on the Effects of Radiation on Human Heredity are concerned with the peaceful uses of

FIG. 9. GAMMA-RADIATION OBSERVED AT THE PLACES I-IV IN FIG. 3(B) FIVE DAYS AFTER A DISTANT HYDROGEN-BOMB TEST



atomic energy and the results of, for instance, the disposal of radioactive wastes from such uses. However, it is essential to take into consideration here the evidence available from atomic-weapon tests, since the distribution of artificial radioactive elements in nature is, at present, mainly due to fall-out from these tests. The dose due to external γ-radiation from fall-out may at the time of writing (December 1956) be disregarded in comparison with the internal dose.

Leaving aside the fall-out in the vicinity of the test area and the effects of radiation during the first few days after the explosion, two different effects may be of interest. One is caused by mixed fission products with a medium half-life (a few days to less than one year), the other by the fission products with a long half-life, particularly Sr-90 (28 years) and Cs-137 (33 years).

Fission products of medium half-life are very unevenly distributed over the world after an atomic explosion. Here, meteorological circumstances play a most important role, since a jet stream, a cold or warm front causing turbulence in the atmosphere, and rain or snowfall can lead to a concentration of the radioactive material in some areas even at a great distance (several thousand kilometres) from the explosion.

A typical example of such an effect is given in Fig. 9, which shows the γ-radiation recorded during about one month in the four northernmost places indicated in Fig. 3(B). The increase in the γ-radiation occurred about five days after an atomic-bomb test. It is obvious from these observations that a comparatively narrow set of stations is required to give an adequate picture of the distribution.

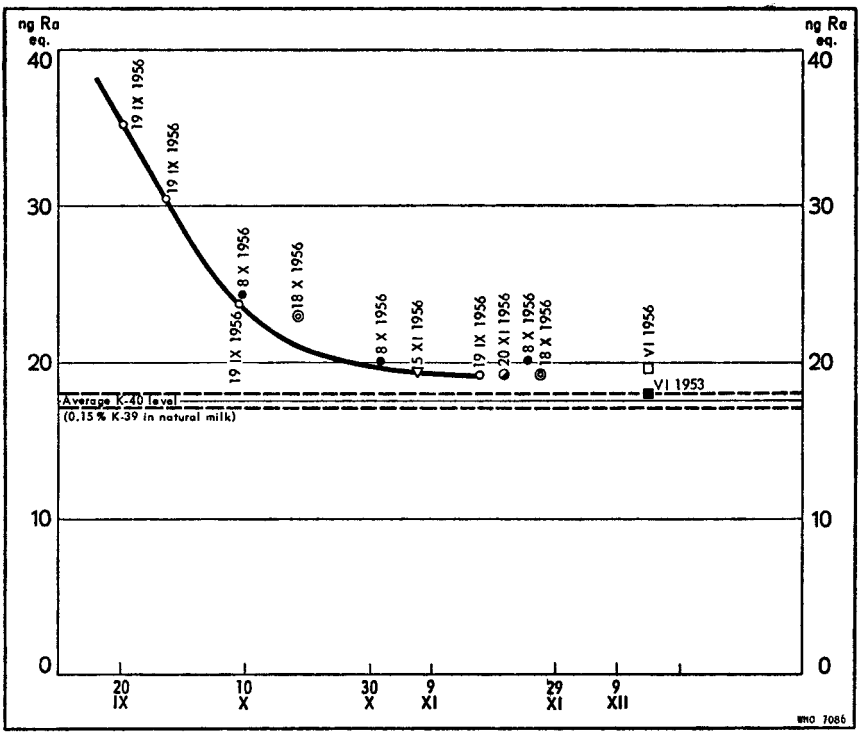
It has been shown by recent measurements of the γ-radiation from large samples of foodstuffs in Sweden that most of our food today (milk, beef, cereals and vegetables) contains artificial radioactive elements, in many cases greatly exceeding the K-40 radiation level of animals and plants. As an example, a decay curve obtained from powdered milk is shown in Fig. 10.

After some bomb tests I-131 is easily detectable in the thyroid of growing cattle. The content of this element in Swedish cattle during September - October 1956 is shown in Fig. 11. The maximum dose per week was here 0.04 rad, or about 20 times the dose due to the average natural radiation, which can be considered to be about 0.002 rad per week. It is to be noted that the effects demonstrated in Fig. 10 and 11 are due mainly to atomic-bomb tests carried out in August and September 1956, but that even before that time the foodstuffs were contaminated to an easily detectable extent, partly owing to medium half-life elements.

It is difficult to estimate today what doses have been received by populations in different parts of the world from mixed fission products. In comparison with the doses from the fall-out of Sr-90 and Cs-137, the mixed fission products may in many cases give smaller doses calculated over a long period. It must, however, be borne in mind that many

more biological effects may be dependent on the intensity of the radiation than we at present know. Our knowledge of the effects of small doses over long periods is very scanty and we cannot as yet be sure that the time-intensity factor can be disregarded, even with respect to genetic effects.

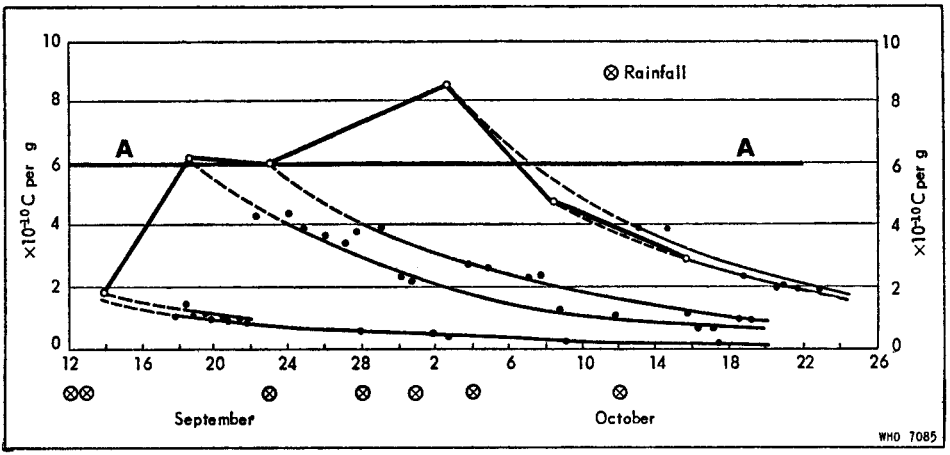
FIG. 10. DECAY CURVE FOR GAMMA-RADIATION FROM MIXED FISSION PRODUCTS IN POWDERED MILK



The sample of powdered milk was taken in September 1956 and the radiation was measured by means of a pressure ion-chamber.

The fall-out of Sr-90 and Cs-137 has been carefully studied during the past years. These elements are probably comparatively evenly distributed over the whole world (with the possible exception of the polar regions). At present, large amounts of them remain in the upper atmosphere, but they will gradually fall, and it is estimated that the present abundance of Sr-90 and Cs-137 on the earth's surface will eventually be increased by a factor of 3-5, even if the firing of atom bombs is stopped. The incorporation of Sr-90 into the skeleton may, in places where the calcium content of the soil is small, be regarded as important.

FIG. 11. GAMMA-RADIATION FROM IODINE-131 IN THYROIDS FROM GRAZING CATTLE



1000-g samples (50 thyroids) were taken and the radiation was measured by means of a pressure ion-chamber. The dotted lines indicate the extrapolation of the decay curves observed back to the last day on which the majority of the cattle presumably grazed. The line A-A corresponds to the maximum permissible level for large human populations.

It does not yet seem possible to estimate the doses to human tissue due to fall-out, nor their distribution in time, which are necessary data for judging the possible biological significance. Experience during the past year is, however, likely to raise doubts as to the lack of biological importance of the tests of nuclear weapons, at any rate if they are continued on the present scale.

It is extremely difficult to predict what will in the future be the most important sources of radiation caused by artificial radioactive elements distributed in nature. There is reason to believe that the problems of disposal of radioactive wastes will be satisfactorily solved, and that precautions in the handling and use of radioactive material will be adequate, but accidents and unforseen events may gradually spread radioactive substances of medium and long half-life beyond control. These radioactive materials will follow unknown paths, and may be harmful to mankind in ways that will become known to us only after long experience.

REFERENCES

1. Anderson, W., Mayneord, W. V. & Turner, R. C. (1954) *Nature (Lond.)*, **174**, 424
2. Forbes, G. B. & Lewis, A. M. (1956) *J. clin. Invest.*, **35**, 596
3. Great Britain, Medical Research Council (1956) *The hazards to man of nuclear and allied radiations*, London
4. Halliday, D. (1950) *Introductory nuclear physics*, New York, p. 461
5. Hultqvist, B. (1956) *K. svenska VetenskAkad. Handl.*, Ser. 4, **6**, No. 3

6. Hursh, J. B. & Gates, A. (1950) *Nucleonics*, 7, 46
7. Schaefer, H. J. (1950) *Aviation Med.*, October, p. 383
8. Shohl, A. T. (1939) *Mineral metabolism*, New York, p. 19
9. Sievert, R. M. & Hultqvist, B. (1956) *Brit. J. Radiol.*, Suppl. 7, p. 1
10. Spiers, F. W. (1956) *Appendix J: The dose of radiation received in human tissues from natural sources.* In: Great Britain, Medical Research Council, *The hazards to man of nuclear and allied radiations*, London, p. 107
11. United States of America, National Academy of Sciences (1956) *The biological effects of atomic radiation: summary reports*, Washington, D.C.