MASTER
Project Report

Contract AT(11-1)3380

U. S. Department of Energy
Division of Biology and Medicine
Washington 25, D. C.

Project Title: The Oncogenic Action of Beta,
Proton, Alpha and Electron Radiation
on the Rat Skin

Institution: Department of Environmental Medicine
The New York University College of
Medicine
550 First Avenue
New York, N.Y. 10016

Principal Investigator: Fredric J. Burns, Ph.D.

Period Covered by
Proposal: February 1, 1980 through
January 31, 1981

Fredric J. Burns
Fredric J. Burns, Ph.D.
Professor

DISCLAIMER

This book was prepared as an account of work sponsored by an agency of the United States Government. Neither the United States Government nor any agency thereof, nor any of their employees, makes any warranty, express or implied, or assumes any legal liability or responsibility for the accuracy, completeness, or usefulness of any information, apparatus, product, or process disclosed, or represents that its use would not infringe privately owned rights. Reference herein to any specific commercial product, process, or service by trade name, trademark, manufacturer, or otherwise, does not necessarily constitute or imply its endorsement, recommendation, or favoring by the United States Government or any agency thereof. The views and opinions of authors expressed herein do not necessarily state or reflect those of the United States Government or any agency thereof.

DISTRIBUTION OF THIS DOCUMENT IS UNLIMITED

leg

DISCLAIMER

This report was prepared as an account of work sponsored by an agency of the United States Government. Neither the United States Government nor any agency Thereof, nor any of their employees, makes any warranty, express or implied, or assumes any legal liability or responsibility for the accuracy, completeness, or usefulness of any information, apparatus, product, or process disclosed, or represents that its use would not infringe privately owned rights. Reference herein to any specific commercial product, process, or service by trade name, trademark, manufacturer, or otherwise does not necessarily constitute or imply its endorsement, recommendation, or favoring by the United States Government or any agency thereof. The views and opinions of authors expressed herein do not necessarily state or reflect those of the United States Government or any agency thereof.

DISCLAIMER

Portions of this document may be illegible in electronic image products. Images are produced from the best available original document.

Technical Scope Summary

Rat skin is being utilized as an empirical model for testing dose and time related aspects of the oncogenic action of ionizing radiation, ultraviolet light and polycyclic aromatic hydrocarbons. Molecular lesions in the skin DNA, including, strand breaks and thymine dimers, are being measured and compared to tumor induction. The induction and repair kinetics of molecular lesions are being compared to split dose repair. Modifiers and radiosensitizers are being utilized to test specific aspects of a chromosome breakage theory of radiation oncogenesis.

Progress Report

DOE Contract AT(11-1)3380

Summary of Progress

Extensive data on tumor induction in rat skin by different types, doses and dose rates of ionizing radiation has been explained in terms of a new "track" model of radiation oncogenesis that includes biological repair of DNA double strand breaks. One important implication of the theory explaining dose rate effects and low dose effects is under empirical investigation to determine its validity. Molecular lesions, such as, strand breaks and thymine dimers, in the epidermal DNA will be measured and related to the radiation dose. The ability of the cells to repair or remove the molecular damage to the DNA will be compared to the ability to repair oncogenic damage as observed in split dose experiments. Skin will be exposed to argon-40 so that a determination can be made whether breaks induced in epidermal DNA by high linear energy transfer (LET) radiation are repaired differently than breaks induced in the same tissue by low LET radiation.

Multiple daily or weekly and chronic exposures to electrons are being tested in light of the predictions of the track model. Sensitizers that interact with DNA in specific ways or modify split dose recovery for endpoints other than tumor induction

will be utilized in an attempt to determine whether molecular lesions in DNA are good indicators of radiation dose relevant to oncogenesis. The repair of DNA strand breaks will be measured in the epidermis of rats and related to age and proliferation rate of the basal cells.

Scientific Background

Rat skin has proved to be a remarkably sensitive and reproducible model for studying the mechanism of radiation carcinogenesis and for formulating ideas that could lead to improved estimates of risk in radiation protection calculations. Studies of the shape of the dose-response curve (1), the effect of spatial distribution of dose (2, 3, 4), the effect of the time pattern (5, 6), and the importance of the density of energy deposition (7, 8) on the induction of skin tumors has led to important insights into radiation carcinogenesis. Whether tumor incidence is proportional to the amount of tissue irradiated was studied by irradiating rat skin in grid and sieve patterns of various pore sizes with electrons, protons and low energy X-rays. A consistent finding was the localized dose patterns were less oncogenic than uniform patterns for low linear energy transfer (LET) radiation, electrons (2) and X-rays (4), but not for high LET protons (9). It was estimated from geometrical considerations that the interaction distance between irradiated and unirradiated tissue was approximately 170 microns and mice are less sensitive than rats (10).

It was found that tumor yield was not dependent on the growth phase of hair follicles at the time of irradiation, although the growing follicle consisted of about ten times as

many cells as the resting follicle (14). When radiation penetrated sufficiently to reach the entire growing phase follicle, the number of tumors produced was not significantly greater than that observed in resting-phase skin. It was shown that radiation must penetrate to a depth of about 0.3 mm into the skin in order to induce tumors (3). The follicular stem cells may be the primary targets for oncogenesis, however, selective irradiation at 0.3 mm, the position of the stem cells, did not produce tumors. Secondary factors, such as gross tissue injury or epidermal depopulation may be required for full expression of the potential to induce cancer (11).

Skin is capable of repairing much of the radiation induced damage that leads to the formation of tumors (5, 6). Recovery of oncogenic damage from electron radiation occurred within 24 hours as demonstrated with split dose irradiation (5, 6). Our recent experiments indicate that the halftime of recovery or repair is between 2 and 3 hours (12, 13). A comparable degree of recovery was found for the carcinogenic effect of protons despite the fact that protons are more oncogenic than electrons (RBE about 2.0-2.4) (8).

The temporal aspects of tumor induction in rat skin are well documented (3). Tumors begin to appear after a latent period of about 20 weeks, and they continue to appear at a reasonably constant rate for up to 90 weeks which is a

substantial portion of lifetime. The guarantee time or minimum latent period does not depend significantly on dose except possibly at very high doses (14).

Recent Progress

A number of experiments were in progress during the past year. Each will be described individually in the following sections. In some instances, the work is sufficiently advanced that it has been put into manuscript form for submission to a journal. Such manuscripts are listed in the bibliography (15, 16, 17).

The Induction and Repair of DNA Strand Breaks

This work has been written up and submitted to Radiation Research (16). The following is a brief description of the major findings.

The dependence of DNA strand breaks (alkaline labile sites) on radiation dose and the rate of repair of such breaks was determined by means of an alkaline unwinding technique. The epidermal DNA was labeled with ^3H prior to irradiation by giving intraperitoneal injections of ^3H -thymidine. After irradiation the epidermal cells were removed and incubated in alkaline solution (pH = 11.4). After the incubation in alkali, S_1 nuclease was added to digest the single stranded portion of the DNA. The amount of undigested double stranded DNA was determined by using a liquid scintillation counter to count the ^3H in the

precipitated DNA. Plots of % double stranded DNA versus time in alkali, referred to as unwinding curves, were utilized to estimate the number of strand breaks (alkaline labile sites) per unit DNA. The strand breaks were found to increase in proportion to radiation dose within the 300 rad to 2400 rad dose range employed in the experiment. When the skin was given 1200 rads, the strand breaks were found to be removed with a halftime of about 17 min. This halftime implies a rate of removal much too rapid for single strand breaks to be the major repairable component of carcinogenic injury, since the latter was found in separate experiments to be removed with a halftime between 120 and 140 min.

A Chromosome Break Model of Radiation Carcinogenesis

A theory has been elaborated that attempts to explain radiation carcinogenesis in terms of interactions between broken chromosomes. So far the dose-response curves for electrons and argon ions, the effect of fractionation, and dose rate effects are remarkably consistent with the results of the model. The target was assumed to be DNA and the primary event produced by the radiation was assumed to be a break in one of the two strands. Such breaks are repaired with no biological consequence, unless a second break occurs in the homologous strand in close enough proximity to generate a double strand break. The theory does not explain the effect of differences in linear energy transfer (LET) and implies that recovery is accomplished by

the repair of single strand breaks. Evidence to be cited later indicates that single strand breaks are repaired too rapidly for them to be significantly involved in split dose recovery of carcinogenesis.

The model being proposed here is based on the concept that a critical event to the formation of a cancer cell is an interaction, probably a fusion, between two DNA double strand breaks. The model was developed only to explain dose-effect and recovery for radiation carcinogenesis in rat skin, although it could have more general validity if other effects of radiation are produced by the same mechanism. The fusion of DNA double strand breaks envisioned here is assumed to proceed quickly when the broken ends are in close geometrical and temporal proximity. Furthermore, the breaks are assumed to be repairable so that an interaction between two may be averted by repair of the first one.

At extremely low LET values where many individual tracks are necessary to produce a given dose, the geometrical distribution of breaks within the nucleus is essentially random. Consequently, the two members of any interacting pair of breaks most likely were produced by ionization events in different tracks. As the LET increases the number of tracks necessary to produce a given dose declines proportionally until at very high LET values, e.g., 100 kev/ μ or higher, hundreds of rads can be delivered by only

one or 2 tracks per nucleus. In the latter circumstance, the breaks, rather than having a random distribution, follow a geometrical alignment along the particle track. Consequently at high LET the members of an interacting pair of breaks were almost certainly produced within the same track.

If the LET is so high that intertrack interactions can be neglected, the proportion of intratrack breaks being close enough to interact is proportional to the number of breaks per unit track length which in turn is proportional to LET. Thus, the coefficient of the linear term in the quadratic function is proportional to LET as in Rossi and Kellerer's model. Moreover, since the breaks governing the linear term are produced and interact almost simultaneously, repair of the breaks can be of little or no consequence to the linear term. On the other hand, repair could be quite important for the dose-squared term because interacting breaks produced by different tracks more than likely would be separated in time, and the first break could be repaired before the second one is produced.

The essence of the idea presented here is that cancer induced by radiation is related to the formation of chromosomal rearrangements derived from the fusion of the broken ends of two separate DNA double strand breaks. The same mechanism of induction is assumed to apply whether the radiation is high LET or low LET. The explanation of the LET dependence is based on

the breaks being produced either in the same track leading to a linear term or in different tracks leading to a dose-squared term. At high LET the tracks are so widely spaced that the interaction of breaks from different tracks is unlikely. Conversely at low LET the individual ionizations are so widely spaced along a given track and correspondingly the number of tracks is so much greater for a given dose that the interaction of breaks from different tracks is greatly increased in comparison to intratrack interactions. It was assumed for simplicity that each type of radiation could be specified by a single average value for LET and that the relevant value was the simple average LET. When the radiation contains a mixture of LET values, what is important is the total track length associated with a given interval of LET values. Thus, the most appropriate LET value to be utilized in these equations is the track-average LET where track length is used as a weighing factor in calculating the average LET.

It is recognized that the model is overly simplified in that it neglects the cytotoxic effect of the radiation and the likelihood that a variety of factors may modify the expression of neoplasia. In this regard, the tumor yield at 80 weeks is an arbitrary endpoint, although our data are consistent with the concept that the function describing the dose and time dependence of tumor yield is separable in a mathematical sense, meaning that it can

be represented as a product of a function of dose only and a function of time only. Mathematical separability implies that the same dose-response function is applicable for all time points. It is implicit in the model that anything acting to modify the expression of neoplastic potential acts equally at all doses and for all types of radiation so that the tumor yield is a direct reflection of the early effect of the radiation on the cells. The effects of severe cell lethality are probably important as an explanation of the reduction in the yield of tumors at higher doses. However, it is assumed that cell lethality is probably important as an explanation of the reduction in the yield of tumors at higher doses. However, it is assumed that cell lethality at lower doses is not sufficient to significantly affect the cells destined to undergo neoplastic alteration.

That neoplasia can be initiated by chromosome damage has been suggested many times (18). What is unique here is the suggestion that the radiobiology of neoplasia can be explained in terms of a molecular lesion involving the fusion of two separate DNA double strand breaks that have been produced in close physical proximity by action of the radiation. It is entirely possible and consistent with the data that other endpoints, such as, chromosomal aberrations and cell lethality could be produced by the same molecular lesion.

One of the important ways to test the hypothesis proposed

here is to show that the primary molecular lesions, the double strand breaks in the DNA are repaired at the same rate of split dose recovery. Unfortunately, it is difficult to measure double strand breaks in vivo at the doses where tumors are produced. Such measurements at higher doses in other cell systems suggest double-strand break repair to be much slower than single break repair in the rat epidermis indicate that the halftime of such repair to be about 17 min. (16), which is too short to be consistent with split dose repair for carcinogenesis which has a halftime of about 3.5 hrs. Such a discrepancy argues against single strand breaks being the primary molecular lesion.

It seems likely that double strand break fusions of the type envisioned here are extremely common in irradiated cells. As many as 20% of all cells irradiated with 500 rad may exhibit chromosomal aberrations of the type involving such fusions. Probably a high proportion of such aberrations are lethal, while others may be completely benign and cells containing them may remain viable for many cell generations. Among the latter the proportion undergoing neoplastic transformation could be extremely small. Even at 80 weeks after irradiation argon ions produced only about 3 epithelial tumors in an area of skin that contained approximately 3×10^7 epithelial cells at the time of irradiation.

Tumor Yield for Multiple Small Doses of Electrons (20)

We have shown in previous studies that when polycyclic aromatic hydrocarbons are applied to the back of rats weekly,

the tumor yield increases steeply, as the 5th or 6th power of time. On the other hand, the time curve for tumors induced by irradiation is only one or 2, but the radiation is usually applied as a single dose. We are interested in determining whether radiation given weekly for indefinite periods of time increased the time exponent such that it becomes more similar to that seen in chemical carcinogenesis.

Male CD rats are shaved 1-2 days prior to each irradiation. Groups of 19 rats are given weekly doses of 0.8 MeV electrons at a dose rate of 300 rads/min. The weekly doses used are: 0 rads, 25 rads, 50 rads, 100 rads and 200 rads. In this protocol, the duration of the experiment and hence, the total dose will depend on the tumor yields. In order to facilitate irradiation the rats are taped onto a metal plate with an exposure window cut out and placed into a holding frame. The irradiated area of the dorsal skin is marked with tattoos so that the same area is irradiated at each exposure.

At this time, the animals have been given 52 weekly irradiations for total doses of 1300 rads, 2600 rads, 5200 rads and 10,400 rads. There has been no ulcerative damage to the dorsal skin that would indicate a lethal effect. No tumors have appeared as yet in the two low dose groups, one tumor appeared at 40 weeks in the 100 rad/wk exposure group and 5 tumors have appeared in the high dose group starting at 36 weeks. At this

time the preliminary evidence indicates that when electron radiation is protracted in this way the rat skin is able to tolerate much higher doses. We are as yet unable to determine if the protraction of doses will affect the power function of the time curve for tumor yield.

BUdR Sensitization of Rat Skin to Electron Tumorigenesis

The incorporation of 5-bromodeoxyuridine (BUdR) into the DNA of cells has been shown to sensitize those cells to the effects of ionizing radiation and ultraviolet light. The sensitization has been measured by a number of biological endpoints including: increased cellular lethality, DNA strand breaks, chromosomal aberrations and in vitro neoplastic transformation. The majority of these experiments were performed in in vitro cell culture systems (21). We have shown that sufficient quantities of 5-bromo-2'-deoxyuridine can be incorporated into the DNA of rat epidermal cells to sensitize those cells to electron radiation as measured by increased DNA strand breakage. The purpose of these experiments was to utilize these findings to determine whether in vivo treatment with BUdR will sensitize the rat epidermis to the tumorigenic effect of electron radiation.

Male CD rats, 21 days of age, were shaved prior to treatment in order to determine the hair growth phase. Only animals in the resting phase were chosen for these experiments. This

limited the BUdR incorporation to the basal epidermal cell and not to the hair follicle cells, and allowed us to target the combined effects of the BUdR and electron radiation to a single cell type. The animals were given intraperitoneal injections of either 5-bromo-2'-deoxyuridine (Sigma Chemical) dissolved in sterile saline or sterile saline alone, starting in the morning, four days prior to irradiation. Animals were given 5 injections of 0.5 ml volume spaced one hour apart for 3 days plus one injection on the fourth day. At this point, the injections had to be stopped because of BUdR toxicity in the high dose group.

The high BUdR dose group received a total of 16 0.5 ml injection of a 2 g/100 ml solution of BUdR for a total of 160 mg BUdR (10 mg/injection). The low BUdR dose group got 16 injections of 0.5 ml of a 1 g/100 ml solution or a total of 80 mg BUdR (5 mg/injection). The experiment was initiated with 15 animals per group.

Both acute and chronic toxicity to the BUdR was noted in the rats, particularly in the high dose group. At 55 days 53% of the animals receiving 160 mg BUdR and 93% of the animals receiving 80 mg BUdR had survived. Such systemic toxicity could possibly be due to bromine toxicity. Dental problems such as broken and chipped teeth were also noted in both BUdR treatment groups.

Irradiated groups given either 80 mg or 160 mg of BUdR showed an increased oncogenic response to 900 rads of electrons compared

to irradiation alone. The tumor response in the high BUdR dose and the saline treatment groups was essentially the same. But that the tumor response in these two groups is somewhat less than animals that are given 80 mg of BUdR or are untreated. At 1800 rads and 2100 rads the BUdR treatment appears to have inhibited the tumor response. This can be explained possibly by the fact that both 1800 rads and 2100 rads are near to the peak tumor yield. At doses higher than these the tumor yield begins to decline due to increased cell lethality. The combination of BUdR with either 1800 or 2100 rads might well cause enough cell lethality to decrease the tumor yield.

The Dose Response for Tumor Induction at Low Doses

The linear plus quadratic dose response model ($Y = AD + BD^2$) has been used to extrapolate risk assessment to lower doses of ionizing radiation (22). Presently there is little data for evaluating the A and B parameters or the effects of dose rate and recovery on the basic equation.

We have designed an experiment attempting to determine the coefficient of the linear term (A) in the dose response equation by assuming that if sufficient time is permitted between the doses

of radiation, multiple doses ought to produce the summation of their individual effects. This experiment is based on the findings of our previous experiment that the recovery halftime of radiation oncogenicity is about 2.5 hrs., therefore, 24 hrs. between doses should be sufficient to allow recovery to occur.

Male CD rats 28 days of age were treated with an oncogenic dose of 0.8 MeV electrons, subdivided into a various number of daily fractions. The following treatment protocol was used, 1500 rads as a single follicle dose, 3-500 rad doses, 5-300 rad doses and 10-150 rad doses. Two sham irradiation groups will be used as controls; a single exposure and 10 day exposure. Each group has 38 rats.

The oncogenic response to 1500 rads of electrons is significantly reduced when given in either 3, 5 or 10 exposures in comparison to single exposure. The results so far indicate that the exponent on the dose-response function is at least 2; the most probable value being about 2.6.

Influence of Retinyl Acetate on Radiation Induced Rat Skin Tumor Induction

Treatment with retinoids has been shown to inhibit tumor induction in a number of different animal systems including skin (23), mammary gland (24), bladder (25) and forestomach (26). In mouse skin retinoid treatment is not only an effective inhibitor of chemically induced tumorigenesis, but also will induce

the regression of benign (27) and malignant lesions (28). Retinoid administration has also been shown to inhibit preneoplastic changes in rat skin. While the mechanism of retinoid inhibition of carcinogenesis is largely unknown, it is felt that an influence on tissue differentiation may be involved.

The purpose of these studies is to determine whether dietary treatment with retinyl acetate will inhibit electron induced rat skin tumorigenesis and also to determine whether differences in retinyl acetate availability relative to time of irradiation will affect the inhibition.

Male CD rats 18 days of age were used. Animals were shaved upon arrival. When not on a retinoid supplemented diet, animals were given water and Purina Lab Chow; during the periods of retinoid treatment, all animals were given water and Purina Lab Chow supplemented with 250 ppm (mg/kg diet) retinyl acetate.

The animals treated with dietary retinyl acetate starting 2 weeks prior to irradiation and continuing through the life span show both a marked reduction in the number of tumors and a delay in tumor onset. If retinyl acetate treatment is begun one week after irradiation, there is a slight inhibition in the tumor yield but no delay of tumor onset. If retinyl acetate treatment is maintained for just the period of 2 weeks prior to irradiation to one week after irradiation, there is no inhibition of the tumorigenic response. This experiment will be continued and all

tumors will ultimately be histologically confirmed.

Epidermal Cell Survival in Rats and Mice Compared

We have found the rat skin to be an excellent system for the examination of radiation induced tumorigenesis. Mouse skin, on the other hand, is a traditional animal system for examining chemical carcinogenesis but has been found to be somewhat refractory to the oncogenic effect to ionizing radiation. The purpose of these experiments was to measure the acute lethality of epidermal cells of several strains of mice and rats to both ionizing and non-ionizing radiations, and chemical carcinogens, such as, 7,12 dimethylbenzanthracene.

The species and strain comparison was made with C57Bl and Balb-C mice and CD-1 rats. Male animals at 28 days of age were used in this experiment. All animals were shaved 4-5 days prior to irradiation in order to determine hair growth phase, only resting phase animals were used. The animals were then given an intraperitoneal injection of 2 μ Ci/g body weight of ^3H -thymidine (40-60 ci/mole; Amersham) in order to label the DNA. Two hours after injection the animals were given varying doses (0, 400, 800, 1200, 1600 rads) of 0.8 MeV electrons at a dose rate of 600 rads/min. Twenty-four hours after irradiation a skin biopsy was taken from the irradiated area and embedded epidermis side down on a microscope slide which was precoated with zip wax. The embedded biopsy is then placed in an ice-cold solution

of 5% trypsin in Ringers buffer for overnight. The dermis is then teased off leaving the epidermis to be developed radioautographically and stained for histological examination.

Each slide was examined microscopically for cells which had incorporated ^3H -thymidine, indicated by the presence of black grains over the cell. Cells in the replicative phase of the cell cycle at the time of ^3H -Tdr treatment will incorporate the radioactive label. If the irradiation is performed 2 hours after the thymidine injection, it is assumed that the majority of the labelled cells will still be in the replicative phase at the time of irradiation. Within the next 24 hours, the labelled cells will complete DNA replication, enter G_2 and then undergo mitosis. Using this technique any cell which incorporates ^3H -thymidine and then undergoes mitosis will appear as an adjacent pair of labelled cells, or a labelled doublet. For each animal approximately 5000 basal epidermal cells were counted, and the percentage of these found as labelled doublets and singlets was quantitated. For each animal group the percentage of labelled cells as doublets decreased with increasing radiation dose. It is assumed that the prevention of normal cell division by radiation treatment is a lethal event. The slopes of the survival curves for the two mouse strains and the rats are essentially the same, but the shapes in the low-dose regions differed considerably. The results were consistent with there being a subpopulation of

cells in the two mouse strains which is highly sensitive to the lethal effects of the radiation. The fact that the mouse skin is more sensitive to lethal effect of radiation could explain its decreased sensitivity to the tumorigenic effect of the radiation.

Epidermal Cell Survival as a Function of Age in Rats

We have reported on the results of experiments in which we have found that the oncogenic effect of radiation is age dependent (17). The oncogenicity of X-rays was shown to decrease as a function of age between one and 99 days. On the other hand, we have shown that the ability of male CD rats to heal X-ray induced skin ulcerations and to repair electron-induced DNA strand breaks decreases as a function of age (this report). In order to further examine these somewhat paradoxical findings of a decrease in both sensitivity and repair capability, we have performed an experiment to compare the acute or lethal response of young (28 day) and old (420 day) rats to electron radiation.

The labelled doublets technique previously described was used in this experiment. All rats were male CD strain at either 28 days or 420 days of age. The average lifespan for these rats is approximately 720 days. The animals were irradiated with 0.8 MeV electrons at a dose rate of 667 rads/min. at the doses indicated.

The cell survival curve for 28 day old animals exhibited a

shoulder between 0 and 800 rads. The 420 day old animals exhibited a semilog response through the dose range of 0-2400 rads; i.e., the shoulder was lacking. Since a shoulder on a survival curve such as this is indicative of the ability of the cells to accumulate radiation damage, such an ability is apparently lost in the older animals. It was found that at all doses, except 400 rads, the labelling index in the 420 day old rats was nearly half that of the 28 day old rats. This indicates that fewer basal epidermal cells were in the replicative phase of the cell cycle at the time of irradiation.

DNA Strand Break Repair as a Function of Age

We have previously shown that the ability of rat epidermal cells to repair electron induced DNA strand breaks decreases as a function of the age of the animal. We found that the half-time of repair increases from 21 min. in 28 day old rats to 69 min. in 200 day old rats to 109 min. in 400 day old rats. We are utilizing this result to continue our studies to determine whether this age-related impairment could be influenced totally or partly by exposure to deleterious environmental agents such as ultraviolet light.

Rats at 28 days of age are being exposed to single ($0, 8 \times 10^5$ ergs/mm² and 13.6×10^5 ergs/mm²) and weekly (0.4×10^5 ergs/mm²) doses of solar spectrum (275-375 nm) ultraviolet light. At various ages the dorsal skin will be assayed for the repair

of DNA strand breaks induced by a standard dose of 1200 rads of 0.8 MeV electrons. Early effects of UV exposure will be examined by measuring DNA repair 2-30 days after UV exposure while late effects will be examined 100-400 days after UV exposure.

References

1. Albert, R. E. W. Newman and B. Altshuler. The Dose Response Relationships of Beta-Ray Induced Skin Tumors in the Rat. Radiation. Res. 15:410, (1961).
2. Albert, R. E., F. J. Burns and R. D. Heimbach. Skin Damage and Tumor Formation from Grid and Sieve Patterns of Electron and Beta Radiation in the Rat. Radiat. Res. 30:525-540, (1967).
3. Albert, R. E., F. J. Burns and R. D. Heimbach. The Effect of Penetration Depth of Electron Radiation on Skin Tumor Formation in the Rat. Radiat. Res. 30:515-524, (1967).
4. Albert, R. E. and F. J. Burns. Tumor and Injury Responses of Rat Skin After Sieve Pattern X-Irradiation. Radiat. Res. 67:142-148, (1976).
5. Burns, F. J., R. E. Albert, I. P. Sinclair and P. Bennett. The Effect of Fractionation on Tumor Induction and Hair Follicle Damage in Rat Skin. Radiat. Res. 53:235-240, (1973).
6. Burns, F. J., R. E. Albert and I. P. Sinclair. The Effect of a 24-Hour Fractionation Interval on the Induction of Rat Skin Tumors by Electron Radiation. Radiat. Res. 62:478-487, (1975).

7. Burns, F. J., R. E. Albert and R. D. Heimbach. The RBE for Skin Tumors and Hair Follicle Damage in the Rat Following Irradiation with Alpha Particles and Electrons. Radiat. Res. 36:225-241, (1968).
8. Burns, F. J., R. E. Albert, M. Vanderlaan and P. Strickland. The Dose-Response Curve for Tumor Induction with Single and Split Doses of 10 Mev Protons. Radiat. Res. 62:598, (1975).
9. Burns, F. J., R. E. Albert, P. Bennett and I. P. Sinclair. Tumor Incidence in Rat Skin Following Proton Irradiation in a Sieve Pattern. Radiat. Res. 50:181-190, (1972).
10. Albert, R. E., F. J. Burns and P. Bennett. Radiation-Induced Hair Follicle Damage and Tumor Formation in Mouse and Rat Skin. J. Natl. Cancer Inst. 49:1131-1137, (1972)
11. Albert, R. E., F. J. Burns and R. D. Heimbach. The Association between Chronic Radiation Damage of the Hair Follicles and Tumor Formation in the Rat. Radiat. Res. 30:590-599, (1967).
12. Vanderlaan, M., F. J. Burns and R. E. Albert. A Model Describing the Effects of Dose and Dose Rate on Tumor Induction by Radiation in Rat Skin. Conference Report-- Presented to International Atomic Energy Agency (IAEA), November, 1975.

13. Burns, F. J. and M. Vanderlaan. Split Dose Recovery for Radiation Induced Tumors in Rat Skin. Inter. J. Radiat. Biol. 32(2):135-144, (1977).
14. Burns, F. J., I. P. Sinclair, R. E. Albert and M. Vanderlaan. Tumor Induction and Hair Follicle Damage for Different Electron Penetrations in Rat Skin. Radiat. Res. 67:474-481, (1976).
15. Burns, F. J., E. V. Sargent and R. E. Albert. A Model of DNA Double Strand Breakage and Repair as a Basis for the Quadratic Dose-Response and Recovery in Radiation Carcinogenesis. (Manuscript in Preparation)
16. Burns, F. J. and E. V. Sargent. The Induction and Repair of DNA Breaks in Rat Epidermis Irradiated with Electrons. Radiation Res. (Submitted) COO-3380-31
17. Burns, F. J., E. V. Sargent, M. Vanderlaan and R. E. Albert. Age-Dependence of Ionizing Radiation Oncogenicity in Rat Skin. (Manuscript in Preparation) COO-3380-32
18. German, J. Chromosomes and Cancer, John Wiley, New York, 1974.

19. Leenhouts, H. P. and K. H. Chadwick. Radiation Induced DNA Double Strand Breaks and Chromosome Aberrations. Theor. Appl. Genet. 44:167-172, (1974).
20. Burns, F. J., E. V. Sargent and R. E. Albert. The Dose-Response for Rat Skin Tumor Induction Estimated from Multiple Doses. Radiat. Res. 83:435, (1980).
21. Shipley, W. W., M. M. Elkind and W. B. Prather. Potentiation of X-Ray Killing by 5-Bromodeoxyuridine in Chinese Hamster Cells: A Reduction in Capacity for Sublethal Damage Accumulation. Radiat. Res. 47:437-449, (1971).
22. Barendsen, G. W. Fundamental Aspects of Cancer Induction in Relation to the Effectiveness of Small Doses of Radiation. In: Late Biological Effects of Ionizing Radiation, Vol. 2, International Atomic Energy Agency, Vienna, 1978, pp. 263-274.
23. Verma, A. K., B. G. Shapas, H. M. Rice and R. K. Boutwell. Correlation of the Inhibition by Retinoids of Tumor Promoter-induced Mouse Epidermal Ornithine Decarboxylase Activity and of Skin Tumor Promotion. Cancer Res. 39:419-425, (1979).

24. McCormick, D. L., F. J. Burns and R. E. Albert. Inhibition of Rat Mammary Carcinogenesis by Short Dietary Exposure to Retinyl Acetate. Cancer Res. 40:1140-1143, (1980).
25. Becci, P. J., H. J. Thompson, C. J. Grubbs, C. C. Brown and R. C. Moon. Effect of Delay in Administration of 13-cu-retinoic Acid in the Inhibition of Urinary Bladder Carcinogenesis in the Rat. Cancer Res. 39:3141-3144, (192()).
26. Smith, D. M., A. E. Rogers and P. M. Newberne. Vitamin A and Benzo(a)pyrene Carcinogenesis in the Respiratory Tract of Hamsters Fed a Semi-Synthetic Diet. Cancer Res. 35:1485-1488, (1975).
27. Protkin, L. The Effect of Vitamin A Acid on Hyperkeratinization and the Keratoacanthoma. J. Invest. Dermatol. 49:165-172, (1967).
28. Bollag, W. Prophylaxis of Chemically-induced Benign and Malignant Epithelial Tumors by Vitamin A Acid (Retinoic Acid). Eur. J. Cancer 8:689-693, (1972).