may be assumed to transform into its inverse the given invariant operator of order 4 of the latter subgroup of order 32. If G exists it can be obtained by extending the group of order 64 generated by the two given subgroups of order 32 by means of an operator of order 4 which is commutative with the product of the two operators of order 4 which are respectively invariant under the two given subgroups of order 32. Since this extending operator of order 4 could not have the same square as the operator of order 4 with which it is assumed to be commutative this is impossible and it therefore results that if a group has the property that its squares generate the four group and that each of its remaining operators is of order 4 then the order of the group cannot exceed 64. It is known that there are at least three such groups of order 64.

When the commutator subgroup of G is of order 2^{n_1} then all the operators of order 4 whose squares are in the commutator subgroup together with its operators of order 2 generate an invariant subgroup of G such that each of the remaining operators of G is of order 4 and has a square which is not contained in the commutator subgroup of G . This theorem results directly from the fact that the quotient group with respect to the commutator subgroup is abelian and that the operators of order 2 in this quotient group generate a subgroup which involves no operator of order 4. The index of the given invariant subgroup under G is equal to the index of the commutator subgroup under the group generated by the squares of the operators of G. When the commutator subgroup of G is of order 2 the given invariant subgroup belongs to one of the three infinite categories of groups which are characterized by the fact that each of them is composed of groups which separately involve two and only two operators which are squares under it.¹

¹ G. A. Miller, Amer. Jour. Math., 55, 417-430 (1933).

COMPARATIVE EFFECTS OF X-RAYS AND NEUTRONS ON NORMAL AND TUMOR TISSUE

By JOHN H. LAWRENCE, PAUL C. AEBERSOLD AND ERNEST 0. LAWRENCE

DEPARTMENT OF INTERNAL MEDICINE, YALE UNIVERSITY SCHOOL OF MEDICINE, DIVi-SION OF ROENTGENOLOGY, UNIVERSITY OF CALIFORNIA MEDICAL SCHOOL AND THE RADIATION LABORATORY, DEPARTMENT OF PHYSICS, UNIVERSITY OF CALIFORNIA

Communicated July 23, 1936

In a previous paper¹ it was demonstrated that per unit of ionization, neutrons were approximately five times as effective as x-rays in producing a lymphopenia in white rats. This result was not surprising in view of the totally different character of the ionization produced in tissue by neutrons when compared with x-rays. Unlike x-rays which resemble light, neutrons

544 PATHOLOGY: LAWRENCE, AEBERSOLD AND LAWRENCE PROC. N. A. S.

are swiftly moving particles of matter which are absorbed by striking the nuclei of atoms, an effect which is greatest for hydrogen (and therefore great for substances rich in hydrogen such as many biological materials), and results in localized and very dense regions of ionization. Figure ¹ is a photograph of the ionization tracks produced by neutrons and gamma rays in a Wilson Cloud Chamber filled with hydrogen.* The very dense long tracks represent recoil protons which are the result of the collisions of

FIGURE ¹ Photograph of ionization tracks in Wilson Cloud Chamber.

neutrons with hydrogen nuclei. The finer lines in the background represent the ionization produced by gamma rays, the result of secondary electron emission. When compared with electrons of equal energy, protons are very heavy and slowly moving charged particles and hence produce ionization with a density of 100 to 1000 times that produced by x-rays or gamma rays. The photograph was taken in $1/100$ th of a second, and represents qualitatively what is taking place in tissue cells exposed to the neutron rays during this period of time.

VOL. 22, ¹⁹³⁶ PATHOLOGY: LA WRENCE, AEBERSOLD A ND LA WRENCE ⁵⁴⁵

Zirkle and Aebersold,² studying the inhibitory effect on the growth of wheat seedlings, found that neutrons were relatively even more effective than the results on rats had indicated. These apparent varying biological sensitivities of two kinds of material, i.e., lymphocytes and wheat seedlings, prompted us to study further the effects of x-rays and neutrons on animal tissues. During a limited period of time, when the cyclotron was available for biological investigation, we have studied the relative effects of filtered 200 kv. x-rays and neutrons on normal albino mice and on Sarcoma 180, an easily transplantable tumor of mice. The lethal doses for mice weighing 20 grams were determined in addition to the effects on the circulating white blood cells in another group of mice. The tumor tissue was irradiated in vitro and then transplanted into normal susceptible mice. Failure to

TABLE ¹

RESULTS OF IRRADIATING MICE WITH 200 KV. FILTERED X-RAYS

grow indicated destruction of the tumor tissue by the respective forms of radiation.

In the experiments previously carried out, using white rats, the neutron dosage measurements were only roughly accurate. It was necessary to place the animals close to the source of neutrons, making it difficult to determine accurately the dosage in r units. Therefore, in the present studies we have used white mice and tumor tissue, these being much smaller test objects, making it possible to obtain quite accurate measurements of dosage.

Methods.—It was the primary purpose of this investigation to determine the relative doses of both neutron rays and x-rays required to kill mouse Sarcoma 180 and to kill healthy mice. In other words, these experiments were concerned with dosage ratios rather than absolute values and a consideration of the relative ionization by neutrons and x-rays did not enter

⁵⁴⁶ PA THOLOG Y: LA WRENCE, AEBERSOLD AND LA WRENCE PROC. N. A. S.

specifically into the problem. It was a question only of comparing the radio-sensitivity of the tumor with that of the healthy mouse for the two kinds of radiation. Accordingly, for the measurement of dosage it was only necessary to use a procedure which yielded dosage values that could be relied upon as being proportional to the amounts of neutron (or x-ray) radiation absorbed in tissue.

Schematic drawing of chamber used in neutron exposures.

Thus it was permissible and convenient to measure the neutron radiation with the same standard apparatus and procedure used in x-ray measurements. A Victoreen condenser type ^r meter having ^a thimble chamber with walls of tissue-like composition served for the measurements of both radiations. The instrument gave directly the x-ray dosages in roentgens. The neutron measurements were also expressed in ^r units, that is to say, the unit of neutron dosage was taken as the convenient and presumably arbitrary amount of radiation that produced the same ionization in the ^r meter as ¹ r of x-rays.

VOL. 22, ¹⁹³⁶ PATHOLOGY: LAWRENCE,AEBERSOLDANDLAWRENCE ⁵⁴⁷

Although knowledge of the ionization produced in the tissues by neutrons and by x-rays was not essential for the investigation, information in this regard was readily obtained. The walls of the dosimeter ionization chamber being thick and similar in atomic composition to tissue, and the dimensions of the chamber being small in comparison to the range of the secondary electrons, the ionization produced in the dosimeter by x-rays was practically due to an equilibrium distribution of secondary electrons from the chamber walls and therefore was proportional to the equilibrium ionization in tissue. Similar considerations indicated that the r meter gave directly equivalent

FIGURE ³ The cyclotron, which produces the intense neutron radiation.

neutron dosages also, for it was probable that most of the ionization in both tissue and the ionization chamber was due to recoil protons having ranges large in comparison to the chamber dimensions. That the neutron ionization in the dosimeter chamber was largely an equilibrium wall effect and therefore that the ionization by the neutron rays in the chamber was an indication of ionization in tissue directly equivalent to that of x-rays, was established by the following experimental test. Observations of ionization by a neutron beam of constant intensity were made with the ionization chamber filled first with air and next with methane. The ionization in the latter gas containing four hydrogen atoms to each molecule was only 20 per cent greater than in the air, indicating that recoil nuclei originating in the gas were relatively small in number and that ionization was largely due to ionizing particles from the chamber walls. Thus the expression of neutron dosages in r units was not only a matter of convenience but was found also to have a certain validity.

Figure 2 shows in some detail the arrangement for the neutron irradiation of the tumors and mice and for the dosage measurements. The biological objects were placed in a hole A in a wooden block about 5 cm. from the source of neutrons, i.e., a target of Be. metal bombarded by several microamperes of 5 million volt deuterons (the apparatus for generating these deuterons of high energy, called the magnetic resonance accelerator, or more conveniently, the cyclotron, has been previously described³ and is shown in general view in Fig. 3). The thick walled wood chamber contributed to an equilibrium of radiation on the biological material. The cylindrical hole A in the block of wood (another at B was also used for some of the smaller dosages) was just large enough to accommodate one 20-gm. mouse and alongside a smaller cavity C was provided for the integrating dosimeter. The tumor particles were contained in a small cavity (equal in dimensions to the dosimeter ionization chamber) in a subsidiary wooden plug which fitted into the mouse hole A . The cylindrical wooden plug in its effect on radiation was substantially equivalent to a mouse, so that the radiation on the tumors was the same as at the center of a mouse in A . other words, the arrangement was such as to insure in first approximation the same quality of radiation in the tumors as in the mice.

In order to calibrate the reading of the dosimeter at C in terms of the r units of radiation at A , a calibrated Victoreen condenser ionization chamber was placed in the plug A in the mouse hole A and thereby the proportionality factor between A and C was readily ascertained.

The dosimeter C, therefore, was calibrated to read directly the dosage in r of the irradiated tumor particles in the cavity in the wooden plug A or the dosage in the central region of a mouse in the cylindrical hole Λ . Inasmuch as the mouse was not small in comparison to its distance from the neutron source it was necessary to make correction for the variation of intensity of the radiation over the region occupied by the mouse. Direct observations of radiation intensity at various places in A (special wooden plugs with ionization chamber cavities off center were used) were made showing an angular distribution of radiation intensity in addition to the inverse square variation with distance and these, by interpolation and numerical integration, led to an estimate of the average value of the total dosage through the body of the mouse in terms of the dosage in the central region. This correction is clearly not accurate and herein lies a possible significant error in the measurements. The tumor measurements on the other hand, do not

involve this correction because the tumors occupied the same space as the calibrating ionization chamber and in all probability the physical measurements in this instance are more precise than the biological observations.

The neutron intensity at A was about 7 r per min. per microampere of deuterons bombarding the beryllium target. Bombarding currents up to about 12 microamperes were available and accordingly the exposure times were not inconveniently long. Although in these first experiments we have not given particular attention to a possible time factor in the neutron effects, the exposure times for the various neutron dosages were kept of the same order of magnitude as the x-ray exposure times which varied from about 2 minutes for the small dosages to fifty minutes for the largest amounts of irradiation.

TABLE ²

RESULTS OF IRRADIATING MICE WITH NEUTRONS

It is hardly necessary to describe the x-ray measurements in detail inasmuch as the procedure is established and well known. The mice were contained in a cardboard box 50 cm. from the target of a 200 kv. (constant potential) x-ray tube and the radiation was filtered by 0.2 mm. tin, 0.25 mm. copper and 2.0 mm. aluminum. Under these circumstances with ¹⁵ m.a. emissions, 32 r per min. was obtained.^{**} The tumors, wrapped in wet filter paper, cellophane and a cellulose capsule, were exposed nearer the x-ray tube, about 36 cm. from the target giving 77 ^r per minute.** The x-ray dosages were recorded by the Victoreen r meter suitably placed alongside the biological specimens.

The mice exposed to x-rays were irradiated in groups of ten, while mice were exposed singly to neutrons. In all of the experiments with Sarcoma 180, tumor particles weighing approximately 8 mgm., were wrapped in groups of 20 in filter paper moistened with a buffered physiological solution

of pH 7.2 and then wrapped in cellophane. The tumor tissue was obtained from freshly killed animals having tumors two to three weeks of age. These were removed under aseptic precautions. After irradiation, the particles were implanted into both axillary regions of susceptible mice. All tumor tissue was implanted at the end of the complete exposure, the control tissue always being implanted last. On the average 3 hours were necessary for the completion of one experiment.

Results.-In table 1 are tabulated the results of irradiating five small groups of animals to various dosages of x-rays. The animals receiving 800 r or more died within a relatively short time from acute radiation intoxica-

TABLE ³

RESULTS OF TRANSPLANTING MOUSE SARCOMA 180 AFTER EXPOSURE TO FILTERED ROENTGEN RAYS (200 KV. 0.2 Sn + 0.25 cu. + 2 ALUM. FILTER-65 R PER MINUTE)

* Numbers in parentheses indicate converted percentages.

tion, nearly all of them developing diarrhea which often was bloody. The animals receiving the smaller doses died after a longer period of time, losing weight and apparently dying of starvation, secondary to injury from radiation. The results of neutron irradiation on four groups of mice is shown in table 2. All animals receiving 185 r or more died in a shorter or longer time, with occasional deaths below this dosage. Diarrhea was less common in the animals exposed to the higher doses of neutrons, loss of weight being the most striking sign. In animals exposed to the larger doses of both x-rays and neutrons, there was a latent period of two or three days, after which the animals were obviously sick, with ruffled fur and arched backs.

In a separate study we shall report the pathological findings in the irra-

Curve indicating that neutrons have the same lethal effect on mice with one-third x-ray dose.

⁵⁵² PA THOLOG Y: LA WRENCE, AEBERSOLD AND LA WRENCE PROC. N. A. S.

diated animals, which may shed light on the mechanism of death for the respective forms of radiation. Warren and Whipple⁴ have shown that the acute x-ray intoxication from large doses is the result of rapid destruction of the mucosa of the small intestine, and acute destruction of the bone marrow. In animals dying from smaller doses, the fatal outcome is apparently due to inability to absorb food from a damaged intestinal mucosa. Our preliminary pathological studies bear out this conclusion, but whether the same mechanism holds in the case of neutron irradiation, we cannot at present say. However, after irradiation with neutrons as with x-rays, degenerative

Showing the fall in leucocytes after irradiation with 100 r neutrons and 300 r x-rays. Each point represents average of 3 animals.

lesions of the spleen and small intestine develop, and the bone marrow becomes aplastic.

In figures 4 and 5 the lethal effects of various doses of x-rays and neutrons are compared. -192 and 260 ^r of neutrons seem equivalent to 541 and 802 r, respectively, of x-rays, indicating that neutrons produce the same lethal effect with one-third the dose. This is also indicated in figure 5, where the neutron scale is one-third that of the x-ray scale.

Blood studies were carried out on a group of 30 mice, exposed to various dosages of x-rays and neutrons. In view of the great variability in the number of leucocytes in white mice, the number of animals exposed is too few- for quantitative analysis. However, as pointed out in the previous paper,' a leukopenia predominantly affecting the lymphocytes, results from neutron irradiation. In figure 6 are charted the average total number of white cells of three mice exposed to 102 r units of neutrons and of three mice irradiated with 300 r units of x-rays. With both forms of radiation, the greatest fall occurs four days after irradiation. In the case of the animals irradiated with neutrons, the fall in the lymphocytes seems relatively greater in relation to the fall in the polymorphonuclears, although the data are too meager to place much weight on this observation. After exposure to these doses the number of white cells returns to a normal level after two weeks.

TABLE ⁴

RESULTS OF TRANSPLANTING MOUSE SARCOMA 180 AFTER EXPOSURE TO NEUTRONS

* Numbers in parentheses indicate converted percentages.

In figure 7 the ratio (per cent) of the lowest absolute number of polymorphonuclear cells and lymphocytes after irradiation to the number before irradiation is plotted against the dose, with the neutron scale one-third the scale for x-rays. These curves show the greater radiosensitivity of lymphocytes to both forms of radiation, and also that neutrons produce roughly the same degree of leukopenia, with one-third the dose necessary with x-rays.

In tables 3 and 4 are shown the results of the irradiation in vitro of par-

554 PATHOLOGY: LAWRENCE, AEBERSOLD AND LAWRENCE PROC. N. A. S.

ticles of Sarcoma 180, followed by transplantation into normal mice. After implantation, the animals were examined weekly and the tumors, if present, were measured and charted. If there were a palpable implant at the end of three weeks it was considered a "take"; likewise, if no implant were palpable at the end of this time it was considered negative. All of the animals were followed for a period of at least six weeks during which time most of the animals developing tumors died of cachexia or ipfection. The presence or absence of a tumor was always checked at autopsy.

The percentage of original cells for minimum after irradiation are plotted against ^r units. The solid dots represent lymphocytes after x-ray, the circles, lymphocytes after neutron irradiation. The solid squares represent polymorphonuclears after x-ray, the open squares, polymorphonuclears after neutron irradiation. Each point represents the average of three animals. The neutron scale is one-third the x-ray scale. The solid line represents lymphocytes, the dashed line polymorphonuclears.

In figure 8 are plotted the percentage implants which grew against the various doses of neutrons and x-rays. It is evident that the lethal dose of x-rays for Sarcoma 180, lies somewhere between 2800 and 3000 r while the dose required to kill half the tumors is in the neighborhood of 2000 r. These results agree fairly closely with the findings of Wood,⁵ Packard⁶ and Sugiura.⁷ In the case of neutrons, the lethal dose seems to lie somewhere around 700-750 r while for 50 per cent the value is near 500 r. It was also generally noted that with the higher doses of neutrons the tumors grew less rapidly when compared to tumors irradiated with equivalent doses of xrays. Thus from the results it appears that neutrons produce the same lethal effect with one-quarter the x-ray dose.

Discussion.-These experiments which have been carried out on a limited number of animals, do not allow us to arrive at any broad conclusions.

Results of transplantation of Sarcoma 180, after irradiation with various doses of neutrons and x-rays.

The work was done in the Radiation Laboratory during a limited period of time and facilities for complete biological study were lacking. In the studies of the lethal effects of the respective forms of radiation on normal animals, it is at present not safe to draw extensively quantitative conclusions. The mechanism of death after neutron irradiation is not yet well understood. In the cases of the few animals studied bacteriologically, in neither group was death due to infection, and it is our belief that, like xrays, neutrons cause death by tissue destruction, resulting in "radiation intoxication." We hope to clear up this point in the near future by complete bacteriological and histological study of a large number of irradiated animals. In the colony of animals used in this study, an occasional death occurred spontaneously from a disease characterized by numerous small liver abscesses. Bacteriological study of one of these animals indicated that this disease fits into the pseudo-tuberculosis group and not into mouse typhoid. Although none of the irradiated animals had the disease grossly, there is no assurance that it may not have been present in a sub-clinical form. With these reservations in mind, the indications nevertheless are that neutrons will kill normal mice with one-third the dose necessary with x-rays. Again, in producing a leukopenia this ratio is also suggested, although the number of animals is too few for quantitative conclusions.

In the relative effects on Sarcoma 180, the dosage ratio of neutrons to x-rays seems to be about 1:4, suggesting that on this tumor tissue, neutrons are relatively more effective than are x-rays. In other words, the results suggest that in comparison with x-rays neutrons may be more selectively effective on the tumor tissue.[†] If further studies on this tumor and other animal tumors bear out these indications, then we have here a new form of radiation, which may have important clinical applications. Other laboratories will soon have facilities for the production of neutrons, and it seems important that the biological effects of this new form of radiation be thoroughly studied. We are now planning further studies when the cyclotron is again available for biological investigations.

 $Conclusions. -1.$ Per unit of ionization, neutrons are much more effective than x-rays in destroying normal mice in vivo, and Sarcoma 180 in vitro.

2. The preliminary results indicate that neutrons are three times as effective in destroying normal mouse tissue, and four times as effective in destroying Sarcoma 180 in vitro.

* This photograph was taken with the cloud chamber at a distance of 20 ft. from the Be. target of the cyclotron which emits both gamma rays and neutrons. The gamma rays, however, are not present in sufficient intensity to produce appreciable biological effects.

** Different doses were obtained by changing the duration of exposure.

^t It may be helpful to express the results by alternative and equivalent dosage ratios, i.e., the tumor lethal dose divided by the mouse lethal dose for the two forms of radiation. Thus, for neutrons the dose required for 100 per cent killing of tumors *in vitro* apparently was about 750 ^r and the dose required to kill mice in seven days was 260 r, while on the other hand, the corresponding values for x-rays were found to be 2800 r and 800 r, re-

spectively. In other words for x-rays $\frac{2800}{0.000}$ = 3.5 times the lethal dose for the mouse is

tumor.

required to kill the tumor while in the case of neutron rays only $\frac{750}{260}$ = 2.9 times the mouse lethal dose produces the same tumor effect. This comparison of dosage ratios indicates that neutron rays may be more selective than x-rays in their effects on this

We are indebted to Dr. Francis Carter Wood for specimens of Sarcoma 180, and to Dr. R. S. Stone for his interest and for the x-ray facilities. One of the authors (P. C. A.) participated in this work through the award of a Fellowship by the Committee of the Christine Breon Fund of the University of California Medical School. We gratefully acknowledge also that these experiments have been made possible by the support of the Josiah Macy, Jr., Foundation, the Research Corporation and the Chemical Foundation.

¹ Lawrence, John H., and Lawrence, Ernest O., Proc Nat. Acad. Sci., 22, 124 (1936).

² Zirkle, Raymond E., and Aebersold, Paul C., *Ibid.*, **22**, 134 (1936).

³ Lawrence, E.O., and Livingston, M.S., *Phys. Review*, 40, 19 (1932).

⁴ Whipple, Geo. W., and Warren, Stafford L., Jour. Exp. Med., 35, 187 (1922). -

⁵ Wood, F. C., Radiology, 5, 199 (1925).

* Packard, C., Am. Jour. Cancer, 16, 1257 (1932).

7Sugiura, Kanematsu, Am. Jour. Roent. and Radium Therapy, 31, 614 (1934).

CHARACTERISTIC TEMPERA TURES IN SUPER-NOVAE

By F. ZWICKY

NORMAN BRIDGE LABORATORY OF PHYSICS, CALIFORNIA INSTITUTE OF TECHNOLOGY

Communicated August 15, 1936

A. Introduction.—Some time ago Baade and I called attention to the existence of certain temporary and extremely luminous objects in extragalactic nebulae. $¹$ We suggested:</sup>

1. These temporary objects are individual stars which behave like giant analogues of common novae and which, therefore, may appropriately be called super-novae.

2. The visual brightness of super-novae, on the average, is comparable to the brightness of the nebulae themselves.

3. The average frequency of occurrence of super-novae is one per extragalactic nebula per several centuries.

Recently the above suggestions have been verified to a great extent. A thorough investigation of past photographic records of temporary objects in nebulae by Baade, as well as the discovery in January, 1936, of a super-nova in N. G. C. 4273 have contributed much new evidence toward the verification of our super-nova hypothesis. It has also proved possible to establish between certain physical characteristics of temporary stars a number of relations which enable us to satisfactorily bridge the gap between common novae and super-novae.²