rial showed that Sevin constituted 20 percent of the injected radioactivity; metabolite I, 11 percent; metabolite II, 3 percent; and metabolite III, 4 percent. The remainder of the radioactivity appeared in the residue after acetone extraction and as expired carbon-14-O_2. The metabolite I fraction from roaches was further resolved into three components in the same manner as with this fraction from microsomes. Bean and cotton plants injected through the stem with Sevin slowly converted it to a metabolite(s) eluting from the Florisil column with methanol. This metabolite(s) accounted for about 90 percent of the labeled material remaining in the plant 28 days after treatment. There was no indication of metabolites I and II as detected with animals, or large loss of carbon-14 from the plants over a 28-day period.

A second carbamate insecticide, *o-iso*propoxyphenyl *N*-methylcarbamate, was subjected to the same studies with rat liver microsomes, insects, and plants utilizing carbonyl-C^{14} compound. The microsomes and insects yielded metabolites eluting from Florisil in a manner similar to metabolite I and metabolite III of Sevin. A microsome preparation yielded 67 percent of the carbamate precursor, 30 percent metabolite I, and 3 percent of the methanol fraction metabolite. Cockroaches after 4 hours contained 3 percent of the injected dose as the original carbamate, 12 percent as metabolite I, and 7 percent as the metabolite of the methanol fraction, and the remainder of the radioactivity was present in the residue or liberated as $C^{14}O_2$. As with Sevin, this metabolite I peak was resolved into two components by thin-layer chromatography. The metabolite I peak from the Florisil column was further purified on a celite-acetonitrile-hexane column to yield a material with an infrared spectrum nearly identical to that of *o-iso*propoxyphenyl *N*-methylcarbamate except for additional bands at 1030 and 3400 cm^{-1} which might be associated with a primary alcohol group. Degradation of this metabolite fraction yielded formaldehyde and *o-iso*propoxyphenol. Reaction of *o-iso*propoxyphenyl carbamate with formaldehyde in glacial acetic acid produced a 2-percent yield of a material identical to this metabolite fraction in chromatographic characteristics on Florisil and infrared spectrum. These observations suggest, but do not necessarily establish, that the major component of this metabolite I fraction was *o-iso*propoxyphenyl *N*-hydroxymethylcarbamate. The behavior of *o-iso*propoxyphenyl *N*-methylcarbamate in plants was similar to that of Sevin.

Certain carbamate metabolites from microsomes were separated on Florisil columns and the fractions eluted with hexane-ether and ether were assayed for anticholinesterase activity in vitro with homogenates of housefly heads. The metabolite I and II fractions of Sevin and the metabolite I fraction of the *o-iso*propoxyphenyl compound effected cholinesterase inhibition, although in each case these metabolite fractions were less than one-eighth as potent as their N-methylcarbamate precursors.

The mechanism of selective insecticidal activity, acquired resistance to carbamate insecticides, and synergism of insecticidal activity by methylenedioxyphenyl and other compounds appears to be at least partially related to the biological instability of these carbamates based on studies by many different investigators (for reviews see *1* and *8*). This instability may result from oxidative attack on the *N*-methyl group or the ring by enzymes in the microsomes (*9*).

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Homologous Disease Reactivation by X-radiation

Abstract. *Hybrid* F_1 *mice* (BDF$_1$) *that failed to develop homologous disease after injection of parental (C57 Bl/6) spleen cells were treated with 400 roentgens of total body x-radiation 13 months later. Typical homologous disease promptly developed in these mice. This suggests that parental lymphoid cells may acquire immunologic tolerance to "foreign" host antigens and that the tolerant state is disrupted by sublethal doses of radiation.*

The administration of parental lymphoid cells to weanling F_1 hybrid mice results in a lethal disorder characterized by weight loss, dermatitis, diarrhea, hemolytic anemia with a positive antiglobulin test, leukopenia, thrombocytopenia, hyperglobulinemia, and splenomegaly (*1*). Homologous disease is due to an immunologic reaction of the grafted parental cells against antigens present in the host hybrid and hence has also been called the graft versus host reaction. In most parent-hybrid combinations the mortality rate due to the graft versus host reaction is high, and in many it is usually 100 percent.

In our laboratory the combination C57Bl/6→ (C57Bl/6 × DBA$_2$) F_1 usually kills 60 to 65 percent of recipients when 300 to 400 × 10^6 parental spleen cells are injected intraperitoneally into weanling F_1 hybrids. Why 35 to 40 percent of the recipients fail to develop homologous disease is not understood; "exhaustive sensitization" with the ultimate death of the parental cells has been proposed as a possible mechanism (*2*).

We recently had an opportunity to study this phenomenon in our laboratory. Seven BDF$_1$ mice that had been given approximately 300 × 10^6 C57Bl/6 spleen cells failed to develop homologous disease; they were observed for a period of 13 months. They were apparently healthy in every respect. At the end of this period of observation they were treated with 400 r of total body x-radiation. Within 2 weeks four of these mice had died of typical homologous disease. In each case there was weight loss, severe anemia, a strongly positive antiglobulin test, marked leukopenia, and splenomegaly. Two additional mice developed transiently positive antiglobulin tests, and slowly regained their preradiation

weights over a period of 2 months. One mouse was unaffected.

Six normal mice were radiated in the same way and none of these developed signs of homologous disease or had a positive antiglobulin test. There was a slight, transient weight loss and leukopenia, but within 2 weeks these control mice were completely normal.

Three other groups of BDF_1 mice were also given 400 r of total body radiation: group A, six mice that were given 400×10^5 or less C57Bl/6 spleen cells 5 months before radiation; group B, six mice that had received 440×10^6 C57Bl/6 spleen cells 6 months before radiation and that were "cured" of homologous disease by the administration of prednisone (*3*); group C, eight mice that had received 400×10^6 C57Bl/6 spleen cells 9 months before radiation and that were "cured" of homologous disease by the administration of 6-mercaptopurine (*3*). None of these mice died and none developed overt signs of homologous disease, although two mice in group A, one mouse in group B and two mice in group C developed transient, weakly positive antiglobulin tests.

A substantial body of evidence indicates that homologous disease is due to an immunologic attack by grafted cells against the host. Since the disease in the radiated mice of these experiments was identical in every respect to severe homologous disease, the present results indicate that grafted lymphoid tissue may reside dormantly in the hybrid recipient for a long period of time. The cells are not destroyed by "exhaustive sensitization," since they are quickly triggered into action by sublethal doses of radiation. It is not known why the parental cells are inactive, and the role of radiation in activating them is not clear. However, in view of the recent finding that radiation disrupts acquired immunologic tolerance (*4*), it is possible that in some mice the parental cells develop tolerance to host antigens, thus mitigating the development of homologous disease. Radiation might break the state of tolerance in the graft by at least two mechanisms: (i) inhibiting a host versus graft reaction (*5*), and (ii) stimulating proliferation of the grafted tissue. Evidently, a critical number of grafted cells is necessary, since mice given small numbers of parental cells (group A), or recipients in which the grafted cells were presumably destroyed by chemotherapy (groups B and C) failed to develop the reaction.

The analogy between homologous disease and human autoimmune disease has been stated previously (*1*). The present experiments offer a striking model of the exacerbation of chronic lymphocytic leukemia or lymphosarcoma by radiation or alkylating agents. In these cases, not only does the course of the disease accelerate, but autoimmune phenomena frequently appear. Perhaps a similar phenomenon, namely abrogation of "tolerance" of the tumor for the patient's antigens, occurs in some cases of malignant lymphoma treated with these agents (*6*).

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Altruism or Arousal in the Rat?

Abstract. *If rats are exposed successively to white noise, then to recorded rat squeals, or alternatively to squeals first and then white noise, and if, in both situations, they are allowed to shut off the auditory stimulation by applying pressure on a bar, they will touch the bar more frequently when exposed to white noise. The results indicate that the bar-pressing behavior is the expression of increased activity resulting from the increased stimulation.*

Rice and Gainer (*1*) have described behavior in the albino rat which, they suggest, might be homologous to altruism. Their rats would press a bar to lower a suspended animal showing obvious signs of distress, such as "squealing and convulsive wriggling." Since the operating rats in this situation pressed the bar significantly more often than control animals, their behavior was interpreted as being altruistic.

This conclusion does not follow unless it can be demonstrated that the bar is pressed specifically to relieve the distress of the other animal. Indeed, there could be various reasons for the behavior described by Rice and Gainer. The extra stimulation in the experimental, as opposed to the control situation, may raise the general activity level of the organism and hence increase the number of bar presses. This could be considered an arousal phenomenon. For the purposes of the present study, the question is simply: is there some specific component in the squeal of a distressed rat which triggers, in a listening rat, behavior calculated to relieve the distress, or does the increased stimulation result in increased activity?

To resolve this problem an experiment was carried out with ten male albino rats of the Wistar strain, all experimentally naive. From the age of 5 weeks they were handled and weighed daily. At 6 weeks they were introduced to an experimental box constructed according to the specifications of Rice and Gainer (*1*), in two groups of five, chosen at random. Each group remained in the box for 10 minutes. For the next 7 days, each rat spent 10 minutes alone in the experimental box.

Two weeks later each rat of group I was placed in the box, individually, and immediately the white noise was switched on by the experimenter. Whenever the subject touched the bar, the noise was turned off for 15 seconds. If the subject touched the bar again during the 15-second quiet period, an additional 15-second period of silence was allowed before the noise was switched on. Group II was subjected to exactly the same treatment, with the exception that it was exposed to the recorded squeals. Each rat remained in the box for 10 minutes, and the number of times it touched the bar was recorded.

Both groups were tested under the conditions described above for 5 consecutive days. After 2 days of rest, they were tested for another 5 days, with the conditions interchanged, that is, group I was exposed to squeals and group II to white noise.

To obtain "squeals," two other rats were placed in the box and subjected to electric shock. The squeals were recorded on Ampex Instrumentation tape, with an Ampex tape recorder, type 311-2. A loop was made from this recording and analyzed with the Bruel