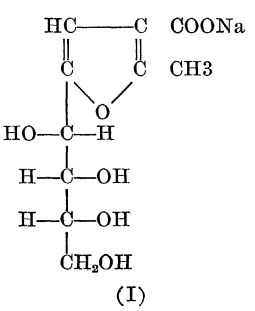
sulfhydryl group of glutathione. Bruckman and Wertheimer (*3*), on the other hand, have observed that substitution of alloxan in both imino groups or in the C_5 position obviates its diabetogenic effect. Although glucose as well as acetoacetate have been found to cause immediate rise in blood sugar on injection, the condensation product of the two, namely 2-tetrahydroxy butyl, 5-methyl, 4-carbethoxy furan (*4, 5*) is without any such effect (*6*). Acetoacetate has been shown to reduce glucose tolerance in rabbits on gram diet (*Cicer arietinum*) (*7*) and to cause a marked increase in the susceptibility to alloxan diabetes (*8*). Studies on the effect of this condensation product on alloxan diabetes were undertaken to see if this substance could have any effect on alloxan diabetes.

This substance was prepared according to the method of West (*9*), slightly modified in our laboratory (*10*). The intraperitoneal injection of the condensation product was given in fine suspension with distilled water to male albino rats weighing between 110 and 160 g, about 45 min before the injection of the diabetogenic dose of alloxan (20 mg/100 g body weight, injected intraperitoneally in a 5-percent solution). In all the 24 rats studied, the condensation product in the molecular proportion of about 15:1 with alloxan was found to prevent completely the development of alloxan diabetes, and when such proportion was reduced to 10:1, complete prevention was observed in about 50 percent of the animals taken. No prevention could, however, be observed when this substance was injected after alloxan was administered in 6 rats.

In the subsequent experiments, where the ethyl ester of the condensation product was transformed into the more soluble Na salt (I) by treatment with alkali (2 *N* NaOH) it has been observed that a much lower amount of this product (i.e., the Na salt) in the proportion of 1.5:1 with alloxan could bring about complete prevention of alloxan diabetes. No glycosuria or hyperglycemia could be noticed in any of the 8 rats studied, even up to a period of 10 days, and the amount of blood GSH was also found to be retained to the normal level (i.e., in the neighborhood of 34 mg/100 ml blood), and all the animals seemed to be perfectly normal.



(I)

The condensation product has recently been shown in this laboratory (*11*) to undergo oxidation by alkaline H_2O_2 to form some bisulfite binding substance which gives a semicarbazone melting at 248° C, and it seems likely that the protective effect of the condensation product might be either through the formation of a complex between it or one of its breakdown products and alloxan, or through the substitution of alloxan in both imino groups as suggested by Bruckman and Wertheimer (*3*).

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The Li^7 (n,t) Reaction

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The well-known Li^6(n,t)α reaction has been utilized for neutron detection, Li^6 detection, and tritium production. The extent of undetected Li^7(n,t) contributions in some of these applications has been questioned (*1*). With the development of equipment for tritium recovery and counting at sufficiently low activity levels (*2*), it became possible, in highly enriched Li^7, to detect the (n,t) reaction.

Pile Irradiation Procedure. The high thermal cross section (*ca.* 900 barns) and $1/v$ dependence of the Li^6(n,t)α reaction required extensive shielding and the purest possible Li^7. Boron and uranium were chosen as the primary shields because of their $1/v$ absorption and because of the additional fission neutrons produced in the uranium. A small cadmium shield was added as an extra precaution. A uranium sleeve location was available in the Oak Ridge National Laboratory graphite pile[1] where the proportions of fission and moderated neutrons were nearly equal. The purest Li^7 available contained 55 ppm Li^6. For a pure fission spectrum, the Li^6(n,t) reaction for this material was computed to yield tritium equivalent to about 0.4 mb Li^7(n,t) cross section, the latter computed for the flux above 2.8 Mev. To reduce the slow neutron Li^6(n,t) contribution to the same value in the location available required an average attenuation of 10^8. The boron powder shield (Fig. 1) was increased to about 10^{10}

[1] Courtesy of the ORNL Solid State Division.

attenuation along the axis of the uranium sleeve as most of the slow neutrons were expected to stream in from the ends.

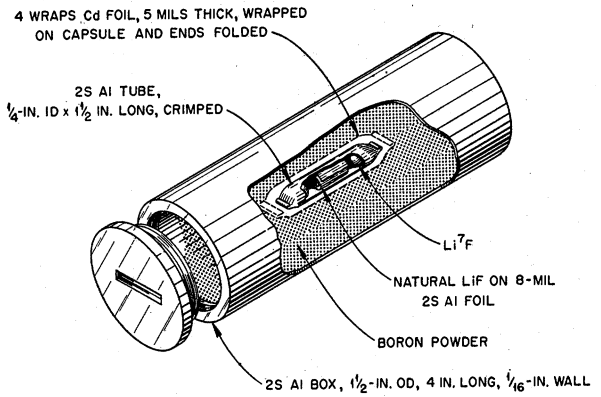

FIG. 1. Experimental arrangement for pile exposure.

Two pills of fused Li^7F (0.2 g each) were irradiated for 10 hr, with a small sample of natural LiF between them, along the axis of the uranium sleeve. The axial attenuation at one end of the assembly was slightly too low and in consequence one of the samples was not usable. The central natural LiF sample indicated a $Li^6(n,t)$ contribution equivalent to 4.2 mb. This is probably several times the effective value at the well-shielded sample, but, in any case, negligible.

Polonium-Beryllium Irradiation Procedure. The high energy of polonium-beryllium neutrons (maximum near 4.5 Mev) made $Li^6(n,t)$ interference less important than in the pile irradiation and, therefore, allowed the use of 9988 percent Li^7 which was available in larger quantities.[2] The capacity of the tritium extraction apparatus (about 0.5 g per run) limited the exposed sample to 2 g Li^7F; 2 g of natural LiF were exposed at the same time as a $Li^6(n,t)$ monitor. The powdered samples were wrapped in aluminum foil and strapped to a thin-walled aluminum tube without shielding. The neutron source was slipped inside the tube and the assembly hoisted up in an attic about 5 m from possible neutron moderating walls or beams for 9 days.

Tritium Yield and Cross Sections. The tritium was recovered by fusing the samples repeatedly in a 1-percent hydrogen-argon atmosphere. The activity was detected by introducing the gas into a proportional counter and adding 10 percent methane-argon as the counting gas. This apparatus had been previously calibrated (*2*), giving 1 c/s for 6.70×10^8 tritons. The pile sample gave 377 c/s, and portions of the Po-Be irradiated sample gave 1.5 c/s. Accidental contamination on unexposed samples ran about 0.5 c/s, so the second result has a rather large uncertainty.

The pile flux above 2.8 Mev, the calculated threshold for $Li^7(n,t)\alpha+n$, has been computed as 2.0×10^{10} n/cm² sec (*3*), with an uncertainty of perhaps 25 percent. This flux decreases exponentially with energy (*4*), so the computed cross section, 72 ± 18 mb[3] is essentially that near 3 Mev. The average neutron loss in the boron shield is about 7 percent at this energy.

The total Po-Be flux computed from the manufacturer's measurements was 2.7×10^6 n/cm² sec. The computed cross section is 30 ± 20 mb.

A more complete investigation of the $Li^7(n,t)$ cross section should be possible through the use of monoenergetic neutrons from charged particle reactions, and much larger samples of Li^7 isotope. The poor recovery of tritium from large samples might be circumvented by fusion as a thin layer or by stirring.

[2] Courtesy of J. Schenck, ORNL Physics Division.

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[3] Tritium produced by the reactions $Li^7(\gamma,t)\alpha$ and $F^{19}(n,t)N^{17}$ is considered negligible.

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Intracellular Distribution of Acid and Alkaline Ribonuclease in Normal Rat Liver[1]

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While investigating the intracellular distribution of ribonuclease (RNase), the presence of RNase activity at both acid and alkaline pH was observed. Extensive survey of the literature[3] revealed that alkaline RNase was never reported present in rat liver.

RNase activity was determined by the spectrophotometric method of Schneider and Hogeboom (*1*). The succinate buffer was replaced by ABC (acetate-borate-cacodylate) (*2*) and PBC (phosphate-borate-cacodylate) buffers, as these buffers could be employed over a broad range of pH and did not seem to affect the RNase activity.

The pH-activity curves (Fig. 1), determined with constant concentrations of ions in ABC and PBC buffers, reveal two peaks of activity, one at 5.8 and another at 8.2. The peak at pH 8.2 is beyond the optimal pH and the isoelectric point of crystalline RNase, which are pH 7.7 and 7.8 respectively (*3*). A proportional relation of enzymatic activity to the amount of tissue used as well as to the length of incubation was found at both pH 5.8 and 8.2. Optimal concentration of substrate was also determined. For 0.1 ml of 10-

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[2] Fellow of the Damon Runyon Memorial Fund for Cancer Research, Inc.

[3] At the time this paper was sent for publication we were not aware of Roth's results on RNase activity of rat liver at different pH values (*Biol. Bull.*, Oct. 1953, p. 359). These results appear to confirm the presence of two peaks of RNase activity.