some animals survived oral doses of 50 mg/kg. Rats tolerated oral doses of 100 mg/kg daily for 15 days. Oral administration to dogs of 5 mg/kg daily for 6 to 17 weeks was well tolerated. Gross and histopathological examination of the vital organs of the medicated rats and dogs revealed no significant abnormalities. Isuprel did not interfere with growth when incorporated into a standard diet to make 0.5% by weight and given as the only source of food to weanling rats.

From the above data it is concluded that Isuprel has a comparatively low toxicity.

The authors are indebted to Dr. J. O. Hoppe for the intravenous toxicity data on mice included in this communication.

16533

Radiation-Induced Hemorrhagic Disease in Chickens.*

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Several articles describing the hemorrhagic disease in animals and man following radiation have recently appeared.[1-3] In these subjects hemorrhage is one of the predominant features in the postradiation syndrome. Allen and Jacobson[1] concluded that, contrary to the prevalent idea, thrombocytopenia plays only a secondary role in producing hemorrhage following radiation in dogs. They felt that the most significant change was the presence in the blood of an increased amount of free heparin. Liebow[2] and Cronkite[3] mentioned the presence of increased numbers of mast cells in tissues of victims of the atomic bombings, a finding of possible significance in view of the suggested relationship between mast cells and heparin production.

The blood changes and hemorrhagic disease we observed in chickens following continuous internal radiation from P^{32} administered subcutaneously were significantly different from those reported above in animals. In comparing the results of our work with the previous work, 3 factors should be kept in mind. First, the radiation differed in two respects from the external radiation of the above experiments. Throughout our experiment, radiation by P^{32} was continuous in contrast with the brief interval of exposure above; while widespread throughout the body, the P^{32} did not give the uniformity of body radiation which results from external total body gamma radiation. Second, the platelets or thrombocytes of the chicken are nucleated cells therefore differing in origin and character from the non-nucleated mammalian forms. Third, compared to the mammals studied, the chicken is relatively resistant to radiation.[4]

Our course of P^{32} injections killed 25% of the 20 experimental chickens and made those surviving at the end of the 17-day observation period moribund. The course of the blood counts and the coagulation times as measured by the capillary tube method using venous blood are recorded on the accompanying graph. The surviving birds were killed after 17 days of radiation, and clot retraction and blood chemistry determinations were made. Gross and microscopic autopsy studies were also done.

Observations. RBC counts decreased stead-

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1 Allen, J. G., and Jacobson, L. O., *Science*, 1947, **105**, 388.

2 Liebow, A. A., and Warren, S. (Abstract), *Am. J. Path.*, 1947, **23**, 888, 892.

3 Cronkite, E. P. (Abstract), *Am. J. Path.*, 1947, **23**, 891.

4 Warren, S. L., and Whipple, G. H., *J. Exp. Med.*, 1923, **38**, 741.

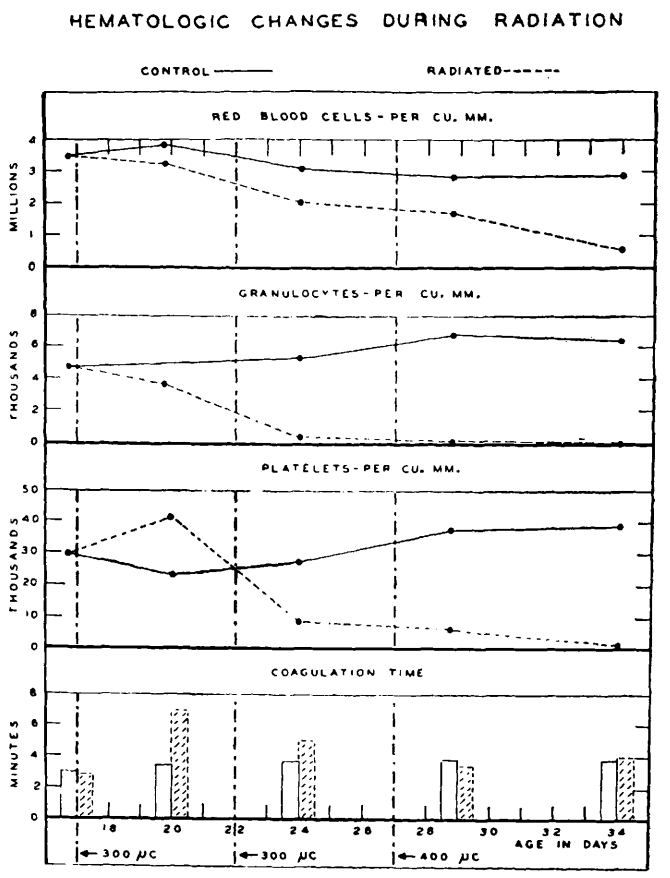

FIG. 1.

ily during radiation reaching 500,000 at the end of the 17-day observation period. Virtual agranulocytosis was reached after seven days of radiation and was maintained throughout the remainder of the experiment. The platelets showed a slight transitory rise and then fell off to insignificant levels after seven days and remained there. These changes in blood counts are comparable to the changes following total external body radiation.

The blood clotting time in both man and dog may be greatly prolonged or the blood rendered entirely incoagulable after radiation.[1] However, the radiated chickens had only a transitory increase in coagulation time over their control mates. This increase was of doubtful significance since the values were within the normal range for chickens reported by other workers.[5,6] In the latter half of the experiment, when the radiation injury was most severe, the coagulation time was not increased.

The character of the blood clot changed markedly in the radiated birds. After the mid-point of the observation period, when the platelets and granulocytes had reached very low levels, the blood clots were jelly-like in consistency and had completely lost the ability to retract. Even after ringing the clot and prolonged centrifuging very little serum could be separated from the clot.

Serum protein determinations, albumen globulin ratios, and blood cholesterol values were not altered in any of the radiated birds.

At autopsy these birds showed little hemorrhagic tendency relative to that described in mammals. Petechiae not exceeding 1 mm in

[5] Wirth, *Folia haemat.*, 1942, **68**, 279.

[6] Amendt, K., *Arch. f. d. ges. Physiol.*, 1922-1923, **197**, 556.

diameter were found scattered over the epicardium, thymus, and fascial planes of the muscles and tendons. Occasionally petechiae were found on the pleural surfaces of the lungs and on the peritoneal surfaces of the bowel. There were no large hemorrhages.

Microscopic study revealed the usual radiation injuries of the lymphoid tissue, bone marrow, and gonads, while other tissues showed little or no change. We failed to find any increase in the number of mast cells in our birds.

Conclusion. The thrombocytopenia developing after radiation and the associated poor clot and loss of clot retraction would seem to be the factors of importance in the hemorrhagic disease in chickens following P^{32} radiation. Since the blood coagulation time was not significantly altered even after severe radiation injury, it seems that the presence of a heparin-like substance similar to that found in dogs would be unlikely.

Summary. In order to study the hemorrhagic disease resulting from radiation, we produced severe radiation injury by repeated subcutaneous injections of P^{32} into young chicks. Agranulocytosis and thrombocytopenia developed rapidly and during the same period the clotting mechanism of the blood was altered. However, there was no significant increase in coagulation time as found in animals and the hemorrhagic disease which developed was much less severe than that seen in animals. The increased number of mast cells associated with atomic bomb radiation injury in humans was not seen in the chickens.

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16534 P

Inulin Volume of Distribution as a Measure of Extracellular Fluid in Dog and Man.

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Current methods for measuring extracellular volume of water utilize substances which enter the tissue cells in a variable proportion (thiocyanate,[1-3] sodium,[4] chloride,[5,6] bromide[7,8]), are very rapidly excreted (sulfate,[1] sucrose[1]), or are partially utilized by the organism (sucrose,[9] mannitol[10,11]).

Inulin has several advantages over any of the above substances: it is not metabolized to

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1 Lavietes, P. H., Bourdillon, J., and Klinghoffer, K. A., *J. Clin. Invest.*, 1936, **15**, 261.

2 Gregersen, M. I., and Stewart, J. D., *Am. J. Physiol.*, 1939, **125**, 142.

3 Elkinton, J. R., and Taffel, M., *Am. J. Physiol.*, 1942-43, **138**, 126.

4 Kaltreider, N. L., Meneely, G. R., Allen, J. R., and Bale, W. F., *J. Exp. Med.*, 1941, **74**, 569.

5 Manery, J. F., *Am. J. Physiol.*, 1940, **129**, 417.

6 Amberson, W. R., Nash, T. P., Mulder, A. G., and Binns, D., *Am. J. Physiol.*, 1938, **122**, 224.

7 Wallace, G. B., and Brodie, B. B., *J. Pharm. and Exp. Therap.*, 1939, **65**, 214.

8 Weir, E. G., and Hastings, A. B., *J. Biol. Chem.*, 1939, **129**, 547.

9 Keith, N. M., and Power, M. H., *Am. J. Physiol.*, 1937, **120**, 203.

10 Smith, W. W., Finkelstein, N., and Smith, H. W., *J. Biol. Chem.*, 1940, **135**, 231.

11 Dominguez, R., Corcoran, A. C., and Page, I. H., *J. Lab. and Clin. Med.*, 1947, **32**, 1192.