

cerebral lesions produced by the injected alumina cream extended from the cortex into the adjacent underlying white matter along the needle track in the "arm-leg" zone. In the animal which failed to develop convulsive attacks a lesion extending into the superior pole of the underlying putamen and adjacent internal capsule was evident.

Discussion. Our findings indicate that multiple intracerebral injections of alumina cream into one hemisphere caused a more rapidly developing convulsive state than that usually induced by a disc containing alumina cream(1). The motor manifestations were initially constant, focal, variable in intensity, and associated with some weakness of the contralateral limbs. In this respect they resembled the form of motor epilepsy in man described as *epilepsia partialis continua* (Koshewnikow)(3).

The only animal which failed to exhibit convulsive movements after the multiple injection treatment of one cerebral hemisphere had received the deepest injections (11 mm),

3. Koshewnikow, A. J., *Neurol. Centralbl.*, 1895, v14, 47.

and the finding of a lesion extending into the superior putamen and internal capsule in this case may be significant. It might be inferred that under these conditions abnormal motor activation from the overlying cortical-subcortical zone was not possible because of interruption at the basal ganglia level.

Summary. 1. Chronic experimental epilepsy was induced in 4 of 5 monkeys (*Macaca mulatta*) by multiple intracerebral injections of alumina cream in the sensori-motor area of one cerebral hemisphere. 2. The early motor convulsive manifestations in the epileptic animals resembled those described in man as *epilepsia partialis continua* (Koshewnikow). 3. In an additional monkey treated by multiple intracerebral injections of alumina cream into both hemispheres death in *status epilepticus* occurred 3 weeks after the onset of seizures. 4. EEG's showed marked spike, sharp, and slow wave activity which was most prominent over the injected hemisphere.

The authors wish to thank Frances Simpson, Elaine Rieders, Frances Zompakos, and Bernice Faigel for their technical assistance.

Received July 27, 1951. P.S.E.B.M., 1951, v78.

Effect of Hyaluronidase on Electrolytes (Sodium and Potassium) of Dog Synovial Fluid. (18971)

K. LEMONE YIELDING AND CARMEN L. ROSANO.
(Introduced by Emmett B. Carmichael.)

From the Biochemistry Department, Medical College and School of Dentistry, University of Alabama, Birmingham.

The action of hyaluronidase as shown by Duran-Reynals(1), is that of depolymerizing a mucopolysaccharide hyaluronic acid, which occurs in high concentration in synovial fluid.

Since hyaluronic acid has been shown to be in a depolymerized state in synovial fluid of arthritic joints(2), and since electrolytes seem to be primarily involved in arthritis(3-5),

tests were undertaken to determine the effect of hyaluronidase on the sodium and potassium content of synovial fluid.

Materials and methods. Twenty mongrel dogs were anesthetized by intraperitoneal injection of 100 mg of sodium phenobarbital per

1. Duran-Reynals, Francisco, *Comp. rend. soc. biol.*, 1928, v99, 6.

2. Ragan, Charles, and Meyer, Karl, *J. Clin. Invest.*, 1949, v28, 56.

3. Jackson, M. D., *Am. J. Med. Sc.*, 1942, v204, 540.

4. Selye, Hans, *Canad. Med. Assn. J.*, 1944, v50, 426.

5. Selye, Hans, *British Med. J.*, 1949, v2, 4637.

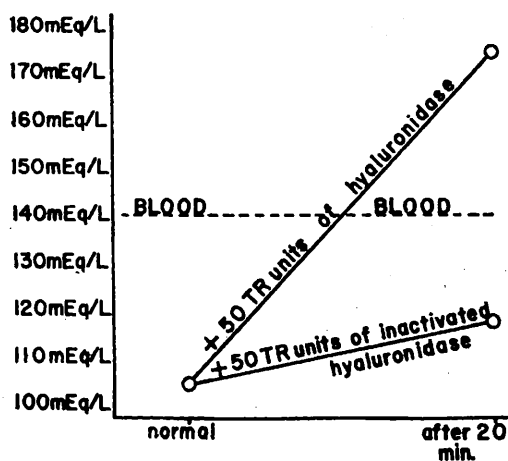


FIG. 1. Effect of hyaluronidase on sodium of synovial fluid.

kilo of body weight. Normal synovial fluid was aspirated from identical joints of both hind legs of each dog in amounts as near to 0.1 cc as possible. The needles were left in place and into one joint of each dog was injected 50 turbidity reducing units of purified bovine testicular hyaluronidase* dissolved in 0.1 cc of distilled water. As a control, the same amount of hyaluronidase which had been inactivated by heating at a temperature of 75°C, was injected into the other joint. After a 20-minute period both knee joints were again aspirated, and the amounts of fluid obtained were similar to those of the previous aspirations. Blood was then obtained by arterial puncture for serum analyses. Sodium and potassium determinations were performed on all specimens by means of the Model DU Beckman Flame Spectrophotometer, using modifications of the Mosher technic(6).

Results. The concentrations of sodium are expressed graphically in Fig. 1. Normal levels of sodium were found to have a mean value of 103.8 mEq/L. Seventy per cent of the samples were within one standard deviation from the mean. After the action of hyaluronidase the mean concentration was 174.6 mEq/L, with 69.2% of the samples falling within one standard deviation from the mean. The difference between these 2 means was

70.8 mEq/L, or 9.1 standard errors. Identical controls run with inactivated hyaluronidase showed a change of only 12.2 mEq/L. Seventy-five per cent of the samples were within one standard deviation from the mean.

The potassium concentrations are expressed graphically in Fig. 2. The values for potassium in normal synovial fluid were found to have a mean of 3.2 mEq/L. Sixty per cent of the samples were within one standard deviation from the mean. Hyaluronidase increased the mean concentration to 7.6 mEq/L, with 50% of the samples being within one standard deviation from the mean. The result was a difference of 4.4 mEq/L, or 4 standard errors between the means. The controls showed a slight decrease of 0.3 mEq/L, probably due to dilution of the normal fluid.

Sodium and potassium concentrations of blood serum were respectively 139.8 mEq/L and 4.3 mEq/L.

It was observed that the viscosity after injection of hyaluronidase was markedly decreased. Because of this difference, the samples were diluted sufficiently so that the accuracy of the instrument would not be affected.

Discussion. It has been shown both *in vivo* and *in vitro* that hyaluronidase depolymerizes the hyaluronic acid of synovial fluid(7), and

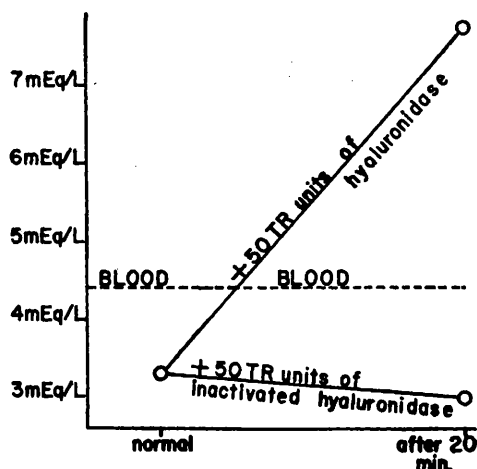


FIG. 2. Effect of hyaluronidase on potassium of synovial fluid.

* The hyaluronidase (Wydase) used in this was kindly supplied by Wyeth, Inc., Philadelphia, Pa.

6. Mosher, R. E., *Am. J. Clin. Path.*, 1949, v19, 461.

7. Ragan, Charles, and De Lamatar, Arlene, *Proc. Soc. Exp. Biol. and Med.*, 1943, v50, 349.

further that the permeability of the synovial membrane is altered by its action(8). Ragan has demonstrated that the synovial fluid of arthritic joints occurs in a depolymerized state(2). There has been some controversy as to the cause of this depolymerization, but Bauer(9) as early as 1940 observed the similarity between arthritic joint fluid and "mucin" after the action of "mucinase."

The importance of electrolyte changes in arthritis has been suggested by experimental evidence. The administration of NaCl solution to rats followed by desoxycorticosterone (DOCA) administration resulted in rheumatic lesions in the heart(4). Selye(5) has produced arthritis in rats by DOCA intoxication, and has further shown that DOCA aggravates formalin induced arthritis. It is interesting to note that DOCA has no effect on the action of hyaluronidase(8). Cortisone, along with certain other compounds used in the treatment

of arthritis, inhibits hyaluronidase(8,10).

In view of all this evidence it seemed of interest to study the relationship of the electrolytes of the synovial fluid to the action of hyaluronidase. The experiments performed showed that hyaluronidase, in addition to decreasing the viscosity of synovial fluid, produced a significant increase in the Na and K levels. Since the resultant concentration was significantly greater than that of the blood it may be assumed that the increase is not due to trauma. Furthermore, the controls failed to show any increase. Analyzing the data statistically it was found that the difference between the means was highly significant.

Conclusions. (1) The action of hyaluronidase increases the concentrations of both sodium and potassium in the synovial fluid of the dog. (2) Inactivated hyaluronidase has no action on the sodium and potassium in dog synovial fluid.

8. Seifter, J., Beader, D. H., and Bagany, A. J., *Proc. Soc. Exp. Biol. and Med.*, 1949, v72, 277.

9. Bauer, W., Ropes, Marion W., and Waine, Hans, *Physiol. Rev.*, 1940, v20, 272.

10. Jaworski, A. A., Farley, John E., Barrett, John, and Jaworski, Rudolf, *A. J. Ped.*, 1950, v37, 597.

Received August 23, 1951. P.S.E.B.M., 1951, v78.

Distribution of Esterase in the Mouse Embryo.* (18972)

ROBERT L. HUNTER. (Introduced by Robert E. Stowell.)

From the Department of Pathology and Oncology, University of Kansas Medical School, Kansas City, Kansas.

The distribution of esterase activity in adult mammalian tissues has been investigated by Gomori and others(1-5). The present study extends these observations to embryonic tissue.

Materials and methods. Strain A mouse embryos measuring 5, 10, 15, and 20 mm in a

crown-rump length were used. Serial sections were made of acetone-fixed embryos for each stage. Adjacent sections were stained with hematoxylin and eosin, and with the Gomori esterase technic(4,6), employing the usual controls.

Results. No esterase activity was demonstrated in the 5 and 10 mm embryos. Activity was shown in both the 15 and 20 mm stages, the reaction being somewhat more intense in the larger embryos. Esterase activity was found in the pancreas, liver, lungs and thyroid, but not in the kidneys. In the pan-

* Aided by a grant from the National Cancer Institute, U. S. Public Health Service.

1. Gomori, G., *Proc. Soc. Exp. Biol. and Med.*, 1945, v58, 362.

2. ———, *Ibid.*, 1948, v67, 4.

3. ———, *Ibid.*, 1949, v72, 697.

4. ———, *Arch. Path.*, 1946, v41, 121.

5. Montagna, W., and Noback, C. R., *Am. J. Anat.*, 1948, v83, 409.

6. Stowell, R. E., and Lee, C. S., *Arch. Path.*, 1950, v50, 519.