

In the focal necrosis the average was 28 per cent., in the diffuse necrosis 21 per cent.

6. No difference could be observed in the rapidity with which the necrotic liver underwent autolysis, the maximum was apparently reached in four weeks. This phase of the subject will be discussed in a later paper.

50 (193)

**The action of nitric acid on the phosphorus of nucleoproteids and paranucleoproteids.**

By **A. B. MACALLUM.**

[*From the Physiological Laboratory of the University of Toronto.*]

The manner in which phosphorus is combined in the true nucleoproteids and in those known as the psuedo (para) nucleo-compounds or phospho-proteins has not as yet been definitely ascertained nor has it been determined that the phosphorus in both classes of compounds is similarly or otherwise combined. Burian<sup>1</sup> has, it is true, suggested that in true nucleic acids phosphorus is the bond between the No. 7 nitrogen of the purin bases and the remainder of the nucleic acid molecule, but this view is founded on the fact that the nucleic acids do not give the diazo reaction which he regards as characteristic of those purins in which there is no substitution of the imide hydrogen of nitrogen No. 7 of the purin skeleton, an explanation of the reaction that is rejected by Steudel who has pointed out that thymin gives the diazo reaction of Burian although it does not contain nitrogen in the No. 7 position.<sup>2</sup>

If Burian's suggestion were accepted it would establish a radical distinction between the manner in which phosphorus is held in nucleic acids and that obtaining in paranucleic acids, for in the latter there are no purin bases.

Whether we do or do not accept Burian's view, it is possible on other grounds to establish a distinction between the two classes of compounds in regard to the manner in which the phosphorus is combined in them. For this purpose nitric acid may be allowed

<sup>1</sup> *Ber. d. d. chem. Ges.*, vol. 37, p. 708 (1904).

<sup>2</sup> *Zeit. für physiol. Chem.*, vol. 42, p. 165 (1904).

to act for periods of varying length on pure preparations of the compounds.

In the observations of which the present paper is the result, nucleic acid from yeast and Hammarsten's nucleoproteid of the pancreas were employed as representatives of the true nucleoproteids while caseinogen exemplified the para compounds.

The yeast nucleic acid and the pancreatic nucleoproteid were prepared in such a way as to free them from lecithin and inorganic phosphates. The nucleic acid was dissolved in dilute sodic hydrate solution (1 per cent. strength) and precipitated therefrom with dilute hydrochloric acid. This solution and precipitation was repeated three times. The precipitate was finally extracted with ether in a Soxhlet apparatus to remove all traces of lecithin. The pancreatic nucleoproteid was prepared by extracting the minced pancreas with boiling water, filtering and adding to the filtrate dilute acetic acid, when the nucleoproteid was precipitated. The precipitate was carefully washed with very dilute acetic acid solution, then dissolved in very dilute ammonium hydrate and the solution rendered acid with acetic acid. The precipitate so obtained was again carefully washed, dissolved and once more precipitated. It was then extracted with ether to remove all traces of lecithin.

In order to determine the absence of phosphates portions of the nucleic acid and of the nucleoproteid so obtained were treated with a solution of ammonium molybdate in nitric acid prepared according to Fresenius' method and the addition of the reagent was followed immediately by that of a solution of phenylhydrazin of 2 per cent. strength. This gave no change of color, indicating the total absence of phosphates. As the nitric-molybdate reagent when employed with phenylhydrazin solution shows one part of P in 2,600,000, the test is an exceedingly sensitive one and consequently it may be relied on to indicate whether phosphates are wholly absent.

When, however, nitric acid of 30 per cent. strength was allowed to act on portions of either the nucleic acid or nucleoproteid for twenty four hours at 35°C., the addition of the nitric-molybdate reagent at once produced a precipitate which is immediately reduced to green or greenish-blue on the addition of the phenylhydrazin solution. That the yellowish precipitate is molybdo-

phosphate of ammonia was shown again and again by dissolving it in ammonia and precipitating it from the latter solution by adding concentrated nitric acid, when the characteristic crystals of the molybdo-phosphate, as shown under the microscope, were formed. The phosphate of this precipitate was also obtained as ammonio-magnesian phosphate. When the nitric acid was allowed to act for a longer time, *e. g.*, from two to six days, at 35°C., the quantity of phosphorus liberated as phosphoric acid was increased.

Quite a different result was obtained with caseinogen. The used quantity of the latter was purified by dissolving and precipitating five times and by extracting with ether to free it from lecithin. The material so prepared did not give the slightest evidence of the presence of phosphates when the nitric-molybdate reagent was added and immediately thereafter some phenylhydrazin solution (1 per cent.). When portions of the pure caseinogen were dissolved in nitric acid of 1.2 sp. gr., and kept at 35° C., for two weeks not the slightest trace of phosphoric acid was demonstrated with the nitric-molybdate reagent and phenylhydrazin, and even after two months only the slightest possible trace of phosphoric acid was present.

It is, therefore, to be concluded that phosphorus is combined in caseinogen in a manner very different from that which obtains in true nucleoproteids and that nitric acid may be employed to distinguish nucleic acids and the typical nucleoproteids from paranucleic compounds.

51 (194)

**Does the stomach of the dog contain free hydrochloric acid during gastric digestion?**

By **LAFAYETTE B. MENDEL.**

[*From the Sheffield Laboratory of Physiological Chemistry, Yale University.*]

In a recent contribution to the physiology of digestion from the Physiological Laboratory of the University of Vienna, Albert Müller<sup>1</sup> has made the announcement that the digestion of meat regularly proceeds in the stomach of healthy, normal dogs in the

<sup>1</sup> Albert Müller: *Archiv für die gesammte Physiologie*, 1907, cxvi, 163.