REFERENCES

1. Fontès, A., and Thivolle, L. *Compt rend. soc. biol.* **93** (1925) 687.
2. Barkan, G. *Z. physiol. Chem.* **171** (1927) 205.
3. Warburg, O. *Biochem. Z.* **187** (1927) 255.
4. Barkan, G. *Z. physiol. Chem.* **216** (1933) 1, 217; **221** (1933), 241.
5. Starkenstein, E., and Harvalik, Z. *Arch. exp. Path. Pharmakol.* **172** (1933) 74.
6. Barkan, G., and Schales, O. *Z. physiol. Chem.* **248** (1937) 96.
7. Vahlquist, B. *Acta Paediat.* **28** (1941) suppl. 5.
8. Holmberg, C. G., and Laurell, C.-B. *Acta Physiol. Scand.* **10** (1945) 307.
9. Schade, A. L., and Caroline, L. *Science* **104** (1946) 340.
10. Cohn, E. J., Strong, L. E., Hughes, W. L., Mulford, D. J., Ashworth, J., Melin, M., and Taylor, H. S. *J. Am. Chem. Soc.* **68** (1946) 459.
11. Cohn, E. *Experientia* **3** (1947) 125.
12. Oncley, J. L., Scatchard, G., and Brown, A. *J. Phys. and Coll. Chem.* **51** (1947) **184.**
13. Laurell, C.-B. *Acta Physiol. Scand.* **14** (1947) suppl. 46.
14. Svensson, H. *Arkiv Kemi, Mineral., Geol.* **22 A** (1946) no. 10.
15. Svedberg, T., and Pedersen, K. O. *The Ultracentrifuge* Oxford (1940).

Received October 8, 1947.

Preliminary Communications

Isolation of a Phosphorus-rich Substance of High Molecular Weight from *Aspergillus niger*

B. INGELMAN

The Institutes of Biochemistry and Physical Chemistry, University of Uppsala, Uppsala, Sweden

While investigating phosphorus metabolism in moulds, Mann made the very interesting observation that extracts from *Aspergillus niger* contained not only pyrophosphate but also metaphosphate [1]. However, he made no determinations of the molecular weight of this metaphosphate. During investigations of the enzymatic breakdown of synthetic polymetaphosphate of very high molecular weight we made preliminary experiments which showed that a fraction of the naturally occurring metaphosphate (from *A. niger* is not dialyzable and therefore may possess a relatively high molecular weight[2].

In 1936 MacFarlane isolated from yeast a nucleic acid preparation which contained 16—18 % P[3]. Later Wiame also isolated from yeast such preparations which contained 17 % P and 8 % N [4, 5].

In our continued experiments we have now shown that it is possible to isolate from *A. niger* a high molecular, non-dialyzable substance with a very high phosphorus-content. This substance does not contain nucleic acid.

For the experiments we have used a culture of *A. niger* v. Tiegh. (no. 594 from the National Collection of Type Cultures maintained in Britain by the Medical Research Council). The mould was cultivated at 28° C on a medium of the following composition: 10 g glucose, 0.2 g K_2HPO_4, 0.4 g $NaNO_3$, 0.05 g $MgSO_4$, $7H_2O$ and 100 ml water. After 8 days' growth the mould was ground in a »Waring

Blendor» together with active carbon (Norit) in a 2 % solution of sodium carbonate. The extract was then immediately filtered two times through large amounts of active carbon. (The enzyme which breaks down polymetaphosphate has its optimum activity at pH 5—6, which has been demonstrated in our other experiments [6]. It is therefore of importance to keep a rather high pH during the extraction. In the filtration through active carbon the enzyme, which breaks down polymetaphosphate, and of course other organic substances, are removed. At low and high pH the spontaneous breakdown velocity of polymetaphosphate is considerable [2, 7]. It is therefore of importance that the pH of the extract is not made too low or too high.) The clear extract was then dialyzed in a cellophane bag against distilled water for 2 days at 4° C. The extract was then evaporated *in vacuo* at 30° C to about 1/50 of its original volume. The concentrated solution was then filtered again through a small amount of active carbon. Then the solution was dialyzed again one day at 4° C. The solution was then frozen and dried *in vacuo* in the frozen state. A small amount of a white powder was obtained.

This non-dialyzable substance contained 25 % P. (Determined after hydrolysis with sulphuric acid according to Lowry and Lopez [8].) A solution of the substance was investigated in the Beckman spectrophotometer. The characteristic absorption of nucleic acids in ultraviolet could not be observed. (The isolated substance contained less than 0.5 % nucleic acid.) The substance contained 15 % Na (determined by W. Kirsten). The carbohydrate content was less than approximately 5 % determined with the orcinol method. The nitrogen content was low, less than 1 %. A small fraction was heated in a crucible. The ash content was 85 %.

The substance was investigated in the Svedberg ultracentrifuge. The experiments were carried out with 1.5 % and 0.5 % of the substance in a buffer solution with the following composition: 0.025 *M* Na_2HPO_4, 0.025 *M* NaH_2PO_4 and 0.10 *M* NaCl. The »ultracentrifugation diagrams» showed a sedimenting substance, which was polydisperse. Most of the substance could be recovered as sedimenting material under the »peak» of the centrifugation diagram. The calculated sedimentation constants were 2.2 and 2.3 Svedberg units for the 1.5 and 0.5 % solutions.

It seems most probable that the isolated substance is a somewhat impure, high molecular sodium polymetaphosphate, *i. e.* an inorganic colloid.

The author wish to thank Prof. T. Svedberg and Prof. A. Tiselius for the privilege of carrying out this work in their laboratories. I am also grateful to Miss A. Karlsson and Mrs. M. Hårsta for their assistance with the laboratory work and to fil. kand. H. Malmgren for valuable discussions.

1. Mann, T. *Biochem. J.* **38** (1944) 339, 345.
2. Ingelman, B., and Malmgren, H. *Acta Chem. Scand.* **1** (1947) 422.
3. MacFarlane, M. G. *Biochem. J.* **30** (1936) 1369.
4. Wiame, J. M. *Biochim. et Biophys. Acta* **1** (1947) 234.
5. Wiame, J. M. *C. R. Soc. Biol., Paris.* **140** (1946) 895, 897.
6. Ingelman, B., and Malmgren, H. In press.
7. Malmgren, H. In press.
8. Lowry, O., and Lopez, J. *J. Biol. Chem.* **162** (1946) 421.

Received January 20, 1948.

2-(Diphenylmethoxymethyl)-imidazoline, a New Potent Antihistamine Agent

RICHARD DAHLBOM AND BERTIL SJOGREN

The Central Laboratories, Astra, Södertälje, Sweden

Ever since the discovery of the first synthetic therapeutically effective antihistamine agent [1], N-phenyl-N-benzyl-N',