gelatin.) In an earlier communication<sup>3</sup> dealing mainly with inhibition of gelation of gelatin by alkaline copper (II), it was reported that a cobalt (II) complex prepared at pH 11.5 also prevented gelation. The correct reading is that a pH 11.5 nickel (II) complex,  $\lambda_{max}$ . 435 mµ, prevented gelation, and that on acidification gelation occurred. The cobalt complex reported earlier was formed at higher alkalinity and was actually the cobalt (III) complex. At that time the cobalt (II) complex was not observed because the very slow adjustment of pH permitted oxidation to complex B without forming a noticeable amount of complex A.

JAKE BELLO HELENE R. BELLO

Department of Biophysics,

Roswell Park Memorial Institute.

Buffalo, 3.

<sup>4</sup> Martin, R. B., Chamberlin, M., and Edsall, J. T., J. Amer. Chem.Soc., 82, 495 (1960).
<sup>2</sup> Manyak, A. R., Murphy, C. B., and Martell, A. E., Arch. Biochem. Biophys., 59, 373 (1955).
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## Formation of 2-Heptanone from Caprylic Acid by Spores of Various Filamentous Fungi

IT was shown recently that washed, non-germinating spores of Penicillium roqueforti are capable of producing 2-heptanone from caprylic acid<sup>1</sup>. In examining the mechanism of this reaction<sup>2</sup>, other fungal spores were examined for their ability to produce a ketone from the fatty acid. It is significant that spores from almost half the fungi selected converted caprylic acid to heptanone, thus indicating the more general occurrence of a reaction thought to be limited to only a few moulds.

The fungi were grown on tomato juice-agar slants in Roux bottles, and after incubation at 25° C. for 3-10 days (depending on the growth-rate of a particular fungus) the spores were collected by adding water to the bottle and scraping the surface of the mycelium with a flattened metal rod. This mixture was then filtered through several layers of cheesecloth to remove bits of agar and hyphal cells. The spores were removed from suspension by centrifugation and then 'washed' several times in distilled water The spore concentration was by centrifugation. adjusted to 10<sup>9</sup>/ml. by using a conventional hæmacytometer. Ketone-forming activity remained con-stant over a period of several months when the spores were stored in water at 4° C.

To avoid using large amounts of spores, which required more time in culturing and collection, the reaction was conducted in Warburg vessels. The reaction mixture consisted of 1.0 ml. of spore suspension, 1.0 ml. of 0.5 M phosphate buffer at pH 6.0 or 7.0 and 0.2 ml. of 0.1 M sodium caprylate;0.2 ml. of 20 per cent potassium hydroxide was placed in the centre well to absorb carbon dioxide. Thus, the oxygen consumption of the spores could be followed manometrically and the general direction and rate of the reaction could be observed. Heptanone production was proportional to the consumption of oxygen; if no uptake of oxygen was observed, no heptanone was produced. After 3-5 hr. the Warburg vessels were removed from the bath and the contents of the vessels were steam-distilled directly into 1.0 ml. of a 0.1 per cent solution of 2,4-dinitrophenylhydrazine. The samples, collected in the hydrazine solution, were stored at 4° C. over-night. The following day the hydrazone crystals were washed, recrystallized from hot 95 per cent ethanol and the melting points The hydrazones were also chromatodetermined. graphed at room temperature in a closed glass cylinder with absolute methanol/water (9:1 by volume) as the developing solvent. The compounds were spotted on a sheet of Whatman No. 1 filter paper and after 6 hr. development the  $R_F$  of the unknown was compared with that of the known hydrazone of 2heptanone.

It is noteworthy that spores from the majority of the Aspergillus sp. (9 out of 11) and Penicillium sp. (9 out of 12) and some related moulds including Paecilomyces varioti and Scaputariopsis brevicqulis rapidly converted caprylate to 2-heptanone. However, none of the spores of Mucorales (Mucor hiemalis, Mucor mucedo, Phycomyces blakesleanus, Rhizopus stolonifer, Thamnidium elegans, and others) was capable of the reaction.

Spores of Penicillium roqueforti and Aspergillus ochraceus were found to produce identically the same amount of 2-heptanone. Some of the fungal spores produced more ketone at pH 6.0 and others more at pH 7.0. Although the reaction with some of the spores may have occurred at a lower or higher pH, our only interest in these particular investigations was to determine the range of the reaction among the fungi The pathway for the conversion of fatty selected. acids to ketones by spores of filamentous fungi is being investigated and the results of these experiments will be published shortly.

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> R. F. GEHRIG S. G. KNIGHT

Department of Bacteriology, **University** of Wisconsin, Madison, 6.

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## Isolation of a Cystine Derivative from Silk

CONSIDERABLE doubt exists as to whether silk fibroin (Bombyx mori) contains a small amount of cystine or whether this amino-acid is entirely absent. Apart from any biological significance, even a very small quantity of cystine, depending on its mode of incorporation, could have a considerable influence on the molecular weight of fibroin.

Schroeder and Kay<sup>1</sup>, using ion-exchange chromatography, reported the presence of 0.17 per cent cystine in silk, and later Zuber, Ziegler and Zahn<sup>2</sup> estimated the cystine by an electrophoretic method after oxidation of the fibroin as 0.12 per cent. In recent reviews on silk<sup>3,4</sup>, however, no reference is made to its cystine content and it has been asserted that sulphur-containing amino-acids are entirely absent and the claim that silk fibroin contains 0.12 per cent cystine cannot be reconciled with the analytical results so far available<sup>4</sup>. In these laboratories it has been found possible apparently to detect cystine in silk hydrolysates by both paper and ion-exchange chromatography and the Shinohara method (Earland, C., Stell, J. G. P., Lodge, R. S., and Raven, D. J.,