solution of a phenol. At 0.01 M potassium laurate the slope of the death-time curve alters sharply, first flattening, then rising to a maximum at about 0.04 M potassium laurate and finally decreasing again with concentration. In the range 0.01-0.04 Mpotassium laurate, the amount of the phenol which the soap will solubilize increases rapidly; but as the phenol soap ratio remains constant in the solutions used, the degree of saturation of the micellar material actually decreases from about 0.015 M to 0.04 M, and it is this fact that is responsible for the rise of death-time in this range. That activity is greatest at approximately 0.01 M soap is due to the concentration of phenol in the micelles being greater at this point than elsewhere along the line AB. Death-times along line CD decrease rapidly as the amount of phenol per molecule of soap increases, indicating that maximum bactericidal activity is attained where the micelles are fully saturated with the phenol.

Experiments using sparingly soluble phenols like 2-chloro-5 hydroxy-1: 3-dimethylbenzene and watersoluble phenols such as the cresols are in progress, and results will be published in due course.

> H. BERRY H. S. BEAN

College of the Pharmaceutical Society, University of London, 17 Bloomsbury Square, W.C.1. Nov. 27.

<sup>1</sup> McBain, Merril and Vinograd, J. Amer. Chem. Soc., 63, 670 (1941). <sup>a</sup> Hartey, J. Chem. Soc., 1968 (1938).

## Calcium-Deficient Media: Their Effect on Phage Action

While typing staphylococci by the bacteriophage method of Wilson and Atkinson<sup>1</sup>, I encountered three batches of nutrient agar that were strongly inhibitory to phage action; several of the phage filtrates had to be used in a concentration one hundred times greater than normal.

Stassano and de Beaufort<sup>2</sup>, Asheshov<sup>3</sup>, Bordet<sup>4</sup>, Burnet<sup>5</sup>, and others have reported on the inhibitory effect of sodium citrate when added to media used for the demonstration of phage action. This suggested that a deficiency of calcium in the medium might be responsible for the inhibition of phage action referred to above. On the suggestion of Dr. G. P. Gladstone of the Lister Institute, calcium chloride in concentrations of M/100 were added to two of the batches of inhibitory media; the third was unfortunately discarded. The effect was striking. The media were now found to be completely satisfactory for the demonstration of bacteriophage activity.

As a result of this experience, it is considered advisable to add calcium chloride to solid culture media that are to be used for the typing of staphylococci by the bacteriophage method.

H. WILLIAMS SMITH

Dept. of Bacteriology, London School of Hygiene and Tropical Medicine, Keppel Street, London, W.C.1. Dec. 1.

<sup>1</sup> Wilson, G. S., and Atkinson, J. D., Lancet, i, 647 (1945).

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Asheshov, I. N., C. R. Soc. Biol., 94, 687 (1926).

4 Bordet, J., C. R. Soc. Biol., 94, 403 (1926).

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## Production of Itaconic Acid and Kojic Acid by a Species of Aspergillus

THE observation reported in the present note relates to an Aspergillus with yellow- to ochraceouscoloured conidia. The mould does not appear to correspond entirely with any published description.

Grown upon the surface of a 20-25 per cent sucrose solution containing the nutrient salts of Kardo-Ssyssojewa's medium<sup>1</sup> at 28° C. in the presence of calcium carbonate, this mould produces both itaconic acid and kojic acid. The relative proportions in which the two compounds appear may be varied by varying the conditions of the fermentation. At higher temperatures itaconic acid preponderates and may amount to more than 20 per cent of the weight of the sugar provided.

The itaconic acid was prepared from the calcium salt which is present in solution and often also separates as nodules on the underside of the mould. Its crystals showed characteristic cleavage; m.p. 163-166° C., not altered by admixture with authentic itaconic acid; analysis: C, 46.41, 46.53; H, 4.92, 5.00 per cent (calc. for  $C_5H_6O_4$ : C, 46.15; H, 4.61per cent); equivalent, 65; the calcium content of the calcium salt was 21.5 per cent.

The kojic acid was prepared via the insoluble copper salt. It melted at 152-153° C.; its solution gave an intense blood-red colour with ferric chloride.

Itaconic acid has hitherto been shown to be produced by A. itaconicus<sup>2</sup> and by A. terreus<sup>3</sup>. The itaconic acid fermentation by A. terreus has since been extensively investigated in the United States in the Fermentation Division of the Northern Regional Research Laboratory4-7.

The work now reported is being continued, and it is hoped to publish it fully elsewhere in due course.

It is a pleasure to acknowledge the assistance of my colleague Mr. J. H. Morrison and of a former colleague, Mr. C. W. Hutchinson.

JOHN L. YULL

John and E. Sturge (Citric), Ltd., 1 Wheeleys Road, Birmingham. Dec. 5.

<sup>1</sup> Kardo-Ssyssojewa, H., Z. Bakt., 93, 265 (1936).

Kinoshita, K., Acta Phytochim., 5, 271 (1930-31).
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<sup>4</sup> Lockwood, L. B., Raper, K. B., Moyer, A. J., and Coghill, R. D., Amer. J. Bot., 32, 214 (1945).

<sup>5</sup> Moyer, A. J., and Coghill, R. D., Arch. Biochem., 7, 167 (1945). Lockwood, L. B., and Ward, G. E., Ind. and Eng. Chem., 37, 405

<sup>7</sup> Thom, C., and Raper, K. B., "A Manual of the Aspergilli", 297 for bibliography (1945).

## Presence of Substances Inhibitory to Acid Phosphatase in Normal Human Urine

In adult bilateral cryptorchid human subjects, a correlation has been found between androgen excretion in the urine and the amount of acid phosphatase in the semen1. The suggestion was made that if a similar correlation was found in normal subjects, measurement of acid phosphatase in semen might suffice for appraisal of urinary androgen content. Since specimens of urine are usually more easily obtained than those of semen, it seemed to be worth while to determine whether a correlation exists between urinary androgens and urinary acid phosphatase.