It is to be noted that tests with 1,000 p.p.m. sulphur dioxide gas (or 1,300 p.p.m. sodium bisulphite), and more, preserved the original colour almost to the very end of the experiment (more than three months), the test being carried out at 25–28° C. Control tests and citrus concentrates containing less than 1,000 p.p.m. sulphur dioxide darkened progressively with storage.

It seems to us that larger amounts of preservatives are required for colour preservation than for microbiological protection. The limit for the former has been found to be 1,000–1,200 p.p.m. sulphur dioxide gas, or 1,200–1,350 p.p.m. bisulphite solution (equivalent to 700 p.p.m. sulphur dioxide).

The relative efficiency of gaseous sulphur dioxide and sodium bisulphite solution may be explained by the different behaviour of the two forms of preservatives when dissolved in the citrus juice concentrate³.

These experiments have been carried out in the Department of Biological and Colloidal Chemistry, Hebrew University, Jerusalem, with the generous co-operation of Prof. A. Fodor, director of the Department. We acknowledge with thanks the interest taken by the Scientific Advisory Committee, War Supply Board, Government of Palestine, which has also sponsored this investigation.

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Vitamin Content of Tubercle Bacilli

MANY chemical compounds which inhibit the growth of certain bacteria are structurally related to compounds which are growth stimulators for the same bacteria. This relationship makes of interest the vitamin content of pathogenic bacteria grown on vitamin-free media, since this may give a clue to the structure of compounds which are important in the metabolism of the micro-organism under study and indicate a starting point for the synthesis of possible growth inhibitors. Thus, Ekstrand and Sjögren have reported on the production of *p*-aminobenzoic acid by the tubercle bacillus¹.

Recently we have had occasion to assay a tubercle bacillus culture for several vitamins by microbiological methods. The culture was an H37Rv strain of virulent human type tubercle bacilli (supplied by Dr. Guy P. Youmans, Northwestern University Medical School, Chicago) which had been grown for 21 days on Proskauer – Beck medium. Determinations were made on the carefully suspended whole culture and on the clear filtrate after removing bacterial cells by centrifugation. The results are indicated in the table.

Vitamins determined	Culture filtrate* (γ/c.c.)	Whole culture† (γ/c.c.)
Vitamin Bc	0.05	—
<i>p</i> -Aminobenzoic acid	—	0.0216
Pantothenic acid	1.04	—
Riboflavin	0.2	1.5
Nicotinic acid	37	61
Biotin	0.03	0.057

* Autoclaved 30 min. at 15 lb. pressure before assaying.

† Autoclaved 30 min. at 15 lb. pressure in *N* sulphuric acid before assaying.

A sample of the basal medium was also assayed for the vitamins indicated. Negative results, indicating less than 0.002 γ per c.c., were obtained for all except nicotinic acid. The amount of nicotinic acid found was 0.071 γ per c.c.

Of interest in these results are the rather high content of nicotinic acid and the low content of vitamin Bc and *p*-aminobenzoic acid. Ekstrand and Sjögren¹, using a chemical method of assay, also found only a small amount of *p*-aminobenzoic acid in tubercle bacilli. This is in contrast to a value of 0.63 γ *p*-aminobenzoic acid per c.c. which we recently obtained for a 17-hr. culture of *E. coli*.

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¹ Ekstrand, T., and Sjögren, B., *Nature*, **156**, 476 (1945).

Antibacterial Titre in Wood-destroying Fungi

FROM exploratory surveys of the production of bacteriostatic substances by wood-rotting fungi both in Great Britain and in the United States, it appears that the authors, Wilkins¹ and Robbins *et al.*², have tested the antibacterial activity of the metabolism-solution periodically up to a maximum of 30 days and 22 days respectively, and that their test-organisms have always been *Bacterium coli* and *Staphylococcus aureus*. On the basis of my experiences stated below, I suggest that: (1) this period of test should be extended, and that (2) some other organisms should also be employed as test-organisms from time to time.

In the course of my work on the antibacterial activity of 'polyporin' from a wood-destroying fungus, *Polystictus sanguineus*, I³ have observed that the antibacterial titre persists for a much longer time (in some cases more than three months), with periodic rise and fall of the titre, and that the activity which appears very early in the course of a week or so goes down soon and does not seem so stable as the one which appears later. For example, in an inoculated flask where the composition of the medium was of MnSO₄, FeSO₄, pea seed-extract, KH₂PO₄ and 4 per cent glucose, the antibacterial activity of the fungus which appeared on the ninth day disappeared on the eleventh, while the activity appearing on the fifteenth day continued until the thirtieth, and the activity reappearing on the sixty-fifth day continued until the eighty-sixth; in another case I have a record of continuing activity from the seventieth to the ninety-eighth day, with five rises and falls from the initial start. Kent⁴ has distinguished two kinds of substances in the metabolism solution, one as staling substances which appear earlier in the course of growth of the organism and are not very stable, and the other as inhibitors which are fairly stable and are slow in appearing in culture media. It has been further observed that a fungus inactive against *Staph. aureus* and *B. coli* forms substances antibiotic against other organisms (typhoid, cholera, etc.).

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