

in proper serological reactions (one of which appears to be the indirect haemagglutination test). Further work will show whether this hypothetical 'C-antigen' is common to all Gram-negative bacteria and of what importance it may be. We have found it up to the present in: *S. typhi*, *S. paratyphi B*, *S. chester*, *S. typhimurium*, *S. cholerae suis* and *Shigella sonnei*.

Thanks are expressed to Prof. O. Westphal and to Dr. O. Lüderitz (Dr. A. Wander Forschungsinstitut, Freiburg) for helpful discussions and support, and to Miss L. Theile for her excellent technical help.

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<sup>1</sup> Walker, J., *Biochem. J.*, **34**, 325 (1940).

<sup>2</sup> Iland, C. N., *Lancet*, **i**, 277 (1953).

<sup>3</sup> Toda, Y., *Ann. Tuberc. (Tsenri)*, **7**, 1 (1956).

<sup>4</sup> Neter, E., Westphal, O., Lucderitz, O., and Gorzynski, E. A., *Ann. N.Y. Acad. Sci.*, **66** 141 (1956). Neter, E., Gorzynski, E. A., Gino, R. M., Westphal, O., and Lucderitz, O., *Canad. J. Microbiol.*, **2**, 232 (1956).

<sup>5</sup> Brodhage, H., *Z. Hyg.*, **148**, 94 (1961).

<sup>6</sup> Weinbach, R., *Schweiz. Z. Path. Bact.*, **21**, 1043 (1959).

<sup>7</sup> Gillissen, G., *Z. Hyg.*, **148**, 105 (1961).

### Utilization of Inorganic Sulphate labelled with Sulphur-35 by *Torulopsis utilis*

It has been established<sup>1</sup> that inorganic and organic sulphurs are utilized by growing yeast. More recently<sup>2</sup> the conditions for the absorption of sulphur-35 by *S. cerevisiae* cells were examined and the necessity of nitrogen and glucose for the utilization of sulphur was demonstrated.

We have in the present experiment examined the utilization of sulphur-35 by *T. utilis* cells which were grown in inorganic salt medium enriched with yeast extract. From this the cells were centrifuged at 0° C., washed with ice-cold water and suspended in sterile water. Aliquots from this suspension were added to the following media (without yeast extract): (a) complete medium without nitrogen; (b) complete medium without sulphur.

Incubation was then carried out aerobically at 25 ± 1° C. for 24 hr. Then the cells were centrifuged and washed with ice-cold water and three suspensions were obtained. Aliquots from each one were added to the following media: (a) without nitrogen and glucose; (b) with nitrogen and without glucose; (c) without nitrogen and with glucose; (d) with nitrogen and glucose.

An amount of K<sub>2</sub><sup>35</sup>SO<sub>4</sub> was added to each medium in order to trace the utilization of sulphur by the cells during absorption periods of 60, 120 and 180 min. The cells were then washed at 0° C., aliquots from the washings being used for determining the radioactivity absorbed.

Table 1

Treatments	Time of incubation (min.)		
	60	120	180
	(c.p.m./gm. dry matter)		
Normal cells			
- N; - glucose	90	120	157
+ N; - glucose	104	194	282
- N; + glucose	78	194	280
+ N; + glucose	200	244	294
N-deficient cells			
- N; - glucose	84	129	139
+ N; - glucose	138	212	216
- N; + glucose	38	144	186
+ N; + glucose	169	185	195
S-deficient cells			
- N; - glucose	61	98	145
+ N; - glucose	119	165	171
- N; + glucose	89	155	243
+ N; + glucose	150	183	255

If the radioactivity of *T. utilis* cells which were grown and allowed to absorb sulphur in complete media during 180 min. is taken as 100 (Table 1), the following conclusions can be drawn:

(1) Glucose and nitrogen are necessary for the utilization of sulphur-35 because:

(a) Cells that were grown in complete medium when allowed to absorb sulphur-35 in medium without nitrogen and glucose showed only 53.4 per cent of absorption.

(b) When these cells were allowed to absorb sulphur-35 in presence of nitrogen or glucose, they showed respectively 95.9 and 95.2 per cent absorption.

(c) Nitrogen-deficient cells when allowed to absorb sulphur-35 in medium without nitrogen and glucose showed only 47.9 per cent absorption; when nitrogen or glucose or both were present, however, the absorption was raised respectively to 73.4, 63.4 and 65.9 per cent. (d) Sulphur-deficient cells which were allowed to absorb sulphur-35 in medium without nitrogen or glucose showed only 49.3 per cent of absorption. On the other hand, when nitrogen or glucose or both were present, the absorption was raised respectively to 58.1, 82.3 and 85.7 per cent.

(2) Nevertheless, nitrogen is not probably necessary for absorption: sulphur-deficient cells when allowed to absorb sulphur-35 in medium with nitrogen or in medium without nitrogen showed in both cases practically the same absorption: respectively 85.7 and 82.3 per cent. This is probably because in sulphur-deficient cells the nitrogen content is increased and thus the sulphur-containing compounds may be formed without any exogenous source of nitrogen<sup>3</sup>.

(3) The utilization of glucose by *T. utilis* cells absorbing sulphur-35 shows that probably the mechanism of sulphur absorption is an active one.

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<sup>1</sup> Schulz, A. S., and McManus, D. K., *Arch. Biochem.*, **25**, 401 (1950).

<sup>2</sup> Kleinzeller, A. A., and Kovac, K. L., *Nature*, **183**, 1402 (1959).

<sup>3</sup> Cowie, D. B., Roberts, R. B., and Bolton, E. T., *Science*, **119**, 379 (1954).

### Susceptibility of *Mycoplasma* (Pleuropneumonia-like Organisms) and Bacterial Protoplasts to Lysis by Various Agents

THE *Mycoplasma* organisms (pleuropneumonia-like organisms, PPLO) are limited by a thin and plastic cell envelope<sup>1</sup>. The absence of diaminopimelic acid and hexosamines in *Mycoplasma*<sup>2,3</sup> indicates the absence of the 'mucoprotein complex' from their cell envelope. The thickness of the *Mycoplasma* cell envelope was found to be ~ 75 Å.<sup>1</sup>, which is similar to that of the cytoplasmic membrane of bacteria<sup>4</sup>. These findings might indicate that the *Mycoplasma* have no cell wall and are limited only by a membrane resembling the cytoplasmic membrane of bacterial protoplasts. In order to elucidate this point, a comparative study of some of the properties of the *Mycoplasma* organisms and bacterial protoplasts was carried out.

*Mycoplasma laidlawii* strain A and *Mycoplasma mycoides* var. *capri* were grown in a modified liquid