

Fig. 1. Percentage dose lysine labelled with carbon-14 incorporated into total plasma proteins at different times in eight experiments

Incorporation of the label was shown to occur mainly into γ - and α -globulins. Negligible values were obtained for albumin; but results for β -globulin were inconclusive because of technical difficulties in The obtaining an uncontaminated preparation. highest specific activity was found for the slower γ-globulin as shown by paper electrophoresis.

These experiments show that the perfused isolated rat spleen synthesizes α - and γ -globulins; but calculation of the actual amount of proteins synthesized on the basis of plasma protein specific activities was not attempted because of lack of information about intracellular amino-acid specific activities¹².

The finding that spleen as well as liver synthesizes α -globulins suggests that the reticulo-endothelial system, in which liver and spleen are both rich, may play a part.

These experiments and others related to them will be published in full in the near future.

I would like to acknowledge Dr. A. H. Gordon's help and supervision in this work and Dr. S. Cohen's advice on chromatographic separations.

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- Gitlin, D., Gross, P. A. M., and Janeway, C. A., New Engl. J. Med., 260, 21 (1959).
 Askonas, B. A., Humphrey, J. H., and Porter, R. R., Biochem. J., 63, 412 (1956).
- 412 (1900).
 Miller, L. L., Bly, C. G., and Bale, W. F., J. Exp. Med., 99, 133 (1954).
 Bradley, J. E. S., Holloway, R. C., and McFarlane, A. S., Biochem. J.,
- ¹⁰ Bradley, J. E. S., Holloway, R. C., and McFarlane, A. S., *Biocnem. J.*, 57, 192 (1954).
 ⁶ Miller, L. L., Bly, C. G., Watson, M. L., and Bale, W. F., J. Exp. Med., 94, 431 (1951).
 ⁶ Askonas, B. A., and Humphrey, J. H., *Biochem. J.*, 70, 212 (1958).
 ⁶ Peters, T., T. Biol. Chem., 200, 461 (1953).
 ⁶ Askonas, B. A., and Humphrey, J. H., Biochem. J., 68, 252 (1958).
 ⁸ Peterson, E. A., and Sober, H. A., J. Amer. Chem. Soc., 78, 751 (1956).
 ¹⁰ Sober, H. A., Gutter, F. J., Wyckoff, M. M., and Peterson, E. A., J. Merer. Chem. Soc., 78, 756 (1956).
 ¹¹ Sober, H. A., and Peterson, E. A., Fed. Proc., 17, 1116 (1958).
 ¹³ McFarlane, A. S., "Prog. in Biophys.", 7, 116 (1957).

Occurrence of 4-O-Methyl Glucuronic Acid in Rhizobium Gums

DURING a survey of the extracellular polysaccharides of Rhizobium species¹, six of seven strains of Rhizobium trifolii were shown to include a component which moved very quickly in acid solvents ($\tilde{R}_{glucose}$ about 3) and, on spraying with p-anisidine hydro-chloride, rapidly developed in the cold a bright pink colour. I have obtained evidence that this compound is 4-O-methyl glucuronic acid. Seven strains of R. meliloti and the one of R. phaseoli that was tested lacked this component which has however since been found in one of two strains of R. leguminosarum.

One mgm. of beech xylan² was hydrolysed in 0.2 ml. 1N hydrochloric acid in a sealed tube at 100°C. for 20 hr. Rhizobium gums were similarly hydrolysed in the proportion of 10 mgm. gum in 0.5 ml. 1N hydrochloric acid. After evaporation to dryness in vacuo in the presence of potassium hydroxide, aliquots were spotted on Whatman No. 1 paper, and developed in the following solvents: (a) butanol:acetic acid:water, 6:1:2; (b) ethyl acetate:acetic acid:formic acid:water, 18:3:1:4; (c) phenol:water, 4:1; (d) propanol:water, 3:2; (e) butanol:pyridine:water; 10:3:3. In solvents (a) and (b) the beech xylan hydrolysates showed two spots. One of these corresponded to xylose, and the other, which moved with the same R_F as the fast-moving components in the gum hydrolysates, also developed its pink colour in the cold. Aspinall et al. state² that beech xylan is composed of xylose and 4-O-methyl glucuronic acid. Hough et al³ found that the methylated uronic acids tested by them with p-anisidine hydrochloride rapidly produced red colours of high brilliance. In solvent (c) the unknown component of the gums ran with the same R_F as xylose and the beech xylan hydrolysate showed only a single spot at the xylose level. In alkaline solvents (d) and (e), the component was held at the starting point in both beech xylan and the gum hydrolysates. Chromatograms run in solvents (a) and (b) and sprayed with brom-cresol blue showed that the component had an acid reaction.

Rhizobium gum (100 mgm.) was hydrolysed in 10 mgm. lots and the hydrolysates run as a band in solvent (a). The fast-moving component was eluted from the paper chromatogram and found to give a positive carbazole reaction for uronic acids with a peak in the absorption curve at 530 m μ (Dische⁴). A similar curve was given by the beech xylan.

4-O-methyl glucuronic acid, a common constituent of hemicelluloses, has not to my knowledge been reported previously from a bacterial source.

My thanks are due to Dr. A. D. Brown of the Commonwealth Scientific and Industrial Organization Marine Laboratory, Sydney, for much helpful advice and for a sample of beech xylan originally obtained from Dr. G. O. Aspinall.

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- ¹ Humphrey, B. A., and Vincent, J. M., J. Gen. Microbiol., 21 (in the press). ⁴ Aspinall, G. O., Hirst, E. L., and Mahomed, R. S., J. Chem. Soc.,
- 134 (1954). 19, L., Jones, K. N., and Vadham, W. H., J. Chem. Soc., 1702
- 113* (1997).
 Hough, L., Jones, K. N., and Vadham, w (1950).
 Dische, Z., J. Biol. Chem., 167, 189 (1947).