growth hormone may be to accentuate the action of insulin in promoting the entry of glucose into the cell.

I wish to thank the Medical Research Council for personal and expenses grants in aid of this investigation.

P. J. Randle

Department of Biochemistry
and Addenbrooke's Hospital,
University of Cambridge.
Aug. 30.

[1] Milman, A. E., and Russell, J. A., *Endocrin.*, **47**, 114 (1950).
[2] Park, C. R., Brown, D. H., Cornblath, M., Daughaday, W. H., and Krahl, M. E., *J. Biol. Chem.*, **197**, 151 (1952).
[3] Ottaway, J. H., *Brit. Med. J.*, ii, 357 (1953).
[4] Wilhelmi, A. E., Fishman, W. H., and Russell, J. A., *J. Biol. Chem.*, **176**, 735 (1948).
[5] Gey, G. O., and Gey, M. K., *Amer. J. Cancer*, **27**, 45 (1936).
[6] Somogyi, M., *J. Biol. Chem.*, **16**), 69 (1945).
[7] Bulbrook, R. D., and Ottaway, J. H., *J. Physiol.*, **123**, 57 *P* (1954).
[8] Young, F. G., Abst. Comm. XIX Inter. Physiol. Cong., Montreal, 1954, p. 18.
[9] Gemmill, C. L., *Bull. Johns Hopkins Hosp.*, **66**, 232 (1940).

An Improved Resolution of Cytoplasmic Proteins of Rat Liver after *n*-Butanol Treatment

In an earlier communication[1], a technique was described for obtaining consistent separations of the cytoplasmic proteins of rat liver using electrophoresis on filter paper. A notable improvement in resolution has been observed following the treatment of the protein mixtures with *n*-butanol. The use of this reagent was suggested by the experiments of Morton[2] on the extraction of various enzymes from the particulate fractions of the cell structure.

The cytoplasm of rat liver (5 volumes), prepared as previously described[1], was shaken at room temperature with *n*-butanol (1 volume) and separated in a centrifuge. The lower aqueous layer was dialysed at 4° C. against distilled water to remove butanol and dried in the frozen state. The cytoplasmic proteins were obtained as a readily soluble powder. A 6–7 per cent solution of this powder subjected to electrophoretic analysis on filter paper as previously described[1] gives a characteristic reproducible pattern of seven bands; the correspondence between the patterns with and without the butanol treatment is illustrated in Fig. 1. Band No. 2 of the untreated sample has become resolved after butanol treatment into three distinct bands (2*a*, 2*b* and 2). Band No. 4 in both patterns corresponds to an esterase activity identified by the diacetyl-fluorescein reagent of Jermyn[3]. The esterase band is sometimes split into two close but distinct components.

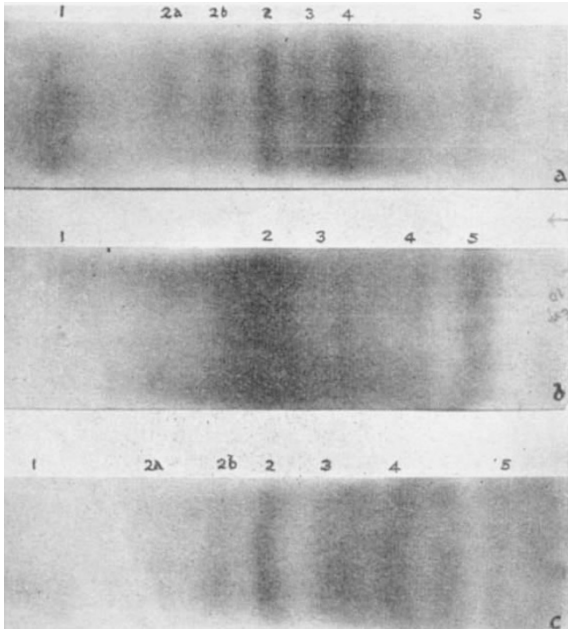

Fig. 1. Electrophoretic pattern of rat liver cytoplasm. *b*, Before, *a* and *c*, after butanol treatment. The numbering of the bands corresponds with those previously described (ref. 1)

Even when butanol is used, a borate buffer is required to obtain satisfactory resolution of the protein components.

G. Adjutantis

Department of Chemical Pathology,
Postgraduate Medical School of London,
Ducane Road,
London, W.12.
Aug. 23.

[1] Adjutantis, G., *Nature*, **173**, 539 (1954).
[2] Morton, R. K., *Nature*, **166**, 1092 (1950).
[3] Jermyn, M. A., *Aust. J. Biol. Sci.*, **6**, 77 (1953).

Inhibition by Cystine of Lecithinase (α-Toxin) Production in *Clostridium welchii* (*perfringens*) BP6K

The highest yields of lecithinase per ml. of medium in cultures of *Clostridium welchii* were reported by Logan *et al.*[1]. In the course of investigations of factors present in this medium which increase toxin production, we have encountered specific instances of the inhibition of the production of lecithinase.

The medium was a pancreatic digest of beef heart prepared according to Logan[1], used at a level of 7·5 per cent of solids. Optimal iron and dextrin concentrations were determined for each fresh preparation of digest. Culture tubes containing 10 ml. of medium, final *p*H 7·5, were inoculated as described by Logan[1] and incubated for 16 hr. at 34° C. The tubes were then centrifuged and the supernatant tested for enzyme activity. The lecithinase activity was measured by the development of turbidity when the culture filtrate was incubated with lecithovitellin in the manner described by van Heyningen[2]. The final turbidities were read in a Coleman spectrophotometer at a wave-length of 590 mμ. From these turbidities the *LD*50 was obtained by comparison with a reference curve given by a stabilized control toxin solution which had been standardized in mice by intravenous injection. Such a standard curve was set up with each experiment.

The pancreatic digest of beef heart was fractionated by means of a 'Dowex 2' cation exchange resin; some of the fractions thus obtained, when added to the original medium, stimulated, while others inhibited, lecithinase production. The inhibition of lecithinase production appeared to be due in part to free amino-acids.

The effect of adding to the normal medium each of the common amino-acids (final concentration of $1{\cdot}5 \times 10^{-3}$ *M* L-amino-acid) was studied. Each