

the links between Bulgaria and various parts of the middle-east and north-east Africa provided by the movement of armies during the prolonged Turkish occupation of these regions.

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- ¹ Lehmann, H., and Raper, A. B., *Nature*, **164**, 494 (1949).
² Foy, H., Kondi, A., Timms, G. L., Brass, W., and Bushra, K., *Brit. Med. J.*, **i**, 294 (1954).
³ Roberts, D. F., and Lehmann, H., *Brit. Med. J.*, **i**, 519 (1955).
⁴ Dherte, P., Vandepitte, J., Ager, J. A. M., and Lehmann, H., *Brit. Med. J.*, **ii**, 282 (1959).
⁵ Hall-Craggs, M., Marsden, P. D., Raper, A. B., Lehmann, H., and Beale, D., *Brit. Med. J.*, **ii**, 87 (1964).
⁶ Vella, F., *Sudan Med. J.*, **3**, 16 (1964).
⁷ Ramot, B., Fisher, S., Remez, D., Schneerson, R., Kahane, D., Ager, J. A. M., and Lehmann, H., *Brit. Med. J.*, **ii**, 1262 (1960).
⁸ Kantechev, K. M., Tcholakov, B., Baglioni, C., and Columbo, B., *Nature*, **205**, 187 (1965).
⁹ Lehmann, H., and Vella, F., *Man*, Art. 215 (1962).
¹⁰ Baglioni, C., and Lehmann, H., *Nature*, **196**, 229 (1962).
¹¹ Lehmann, H., Maranjian, G., and Mourant, A. E., *Nature*, **198**, 492 (1963).
¹² Atwater, J., and Regan, J., *Abst. Tenth Congr. Intern. Soc. Haematol.*, Stockholm, **1**, 5 (1964).

Haemoglobin Typing of the Kerry Breed of Cattle

Two types of haemoglobin, designated Bov A and Bov B, have been identified in the blood of cattle by Bangham¹. These are distinguished by their motility in paper electrophoresis; Bov A is the slow-moving and Bov B the fast-moving component. The type of haemoglobin which occurs in an animal is controlled by two simple allelic autosomal genes which give rise to three phenotypes A, AB and B.

In a survey of stud bulls in Britain, Bangham¹ found that with the exception of the Jersey, Guernsey and South Devon breeds all other British breeds exhibited A type haemoglobin exclusively, while in the case of Channel Island and South Devon bulls both B and AB types were encountered in addition to A type haemoglobin. In a subsequent survey of some cattle breeds of Europe and Africa, Bangham and Blumberg² found that B type haemoglobin occurred in breeds located geographically along a line running southwards through France into Africa, while cattle of north-east France, Holland and Denmark showed A type haemoglobin exclusively. This finding tended to support the suggestion of Boston³ that one ancestral line of the Jersey breed originated in the Indus valley and permeated through Africa to Europe. The type of haemoglobin occurring in Kerry cattle—a breed native to Ireland—has not previously been reported and it was felt that haemoglobin typing might throw some light on the views of Moyles⁴, who has suggested that the Kerry breed originated in mid-Asia.

The standards for the different types of haemoglobin were established according to the method of Bangham¹ using blood samples from different breeds, including Friesians, Shorthorns, Herefords, Guernseys and Jerseys, which exhibited the A, AB and B types. Blood samples were then collected from 25 Kerry cattle in a pure-bred Kerry herd and these were run on paper electrophoresis side by side with samples from other breeds. All twenty-five samples from the Kerry cattle contained the commonly occurring A type haemoglobin. This finding has more recently been confirmed by Dabcewski⁵.

Haemoglobin typing, therefore, would suggest that the Kerry breed of cattle has no link with that ancestral line

of Channel Island cattle but would appear to have an origin resembling that of other northern European breeds.

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- ¹ Bangham, A. D., *Nature*, **179**, 467 (1957).
² Bangham, A. D., and Blumberg, B. S., *Nature*, **181**, 1551 (1958).
³ Boston, E. J., *Jersey Cattle*, 32 (Faber and Faber, London, 1954).
⁴ Moyles, M. G., *J. Dept. Agric. Ire.*, **53**, 53 (1956-57).
⁵ Dabcewski, Z. S. (personal communication).

IMMUNOLOGY

Active Immunization of Mice with Chagastoxin

PREVIOUSLY we reported on the immunology, immunochemistry and immunochemotherapy of experimental *Trypanosoma cruzi* infection in mice. *T. cruzi* has at least 9 antigens. Sonic lysates of the cultures contain antigens A, B, C, D, E, F, G and H, and the sediment contains B, E, D and X. Living trypanosomes have antigens B, E and G. Saline supernatant has antigens A, B and F, while distilled water supernatant has A, B and E, and sediment antigens B and X. The recently phenolized cultures have A, B, G and X, and old phenolized cultures have only B. 50 per cent extraction with ammonium sulphate yields A, B, E, G and F. 100 per cent saturation still extracts B, while precipitation with alcohol after full saturation with ammonium sulphate still extracts antigen B. The phenol extract and sediment (Westphal) contain no antigens, but phenol aqueous extract contains antigen B. The antigens B, E and G are major antigens. The agglutinin titre of hyperimmune rabbit serum was 1:10,240 and that of horse serum was 1:327,600. Both rabbit and horse hyperimmune sera had 1:4 titre of agar-gel precipitin. Immunotherapy with hyperimmune rabbit serum prolonged the survival time in fatal *T. cruzi* infection in mice two- to three-fold. Chemotherapy with 1-furaladone, in doses of 50 mg/kg/day for two weeks, followed by 12.5 mg/kg/day for 2.5 months, protected 86.7 per cent of infected mice, while combined immunochemotherapy with hyperimmune rabbit serum plus 1-furaladone protected all mice for at least 5 months¹⁻³.

The present report deals with the toxicity and immunizing properties of the lipopolysaccharide, chagastoxin.

Phenol extraction. Chagastoxin is prepared or extracted using the following modified Westphal method⁴, used for the extraction of lipopolysaccharides from the cell walls of Gram-negative rods. *Trypanosoma cruzi* (Tulahuana) is grown for 10-12 days on brain heart blood agar (Difco), containing 5-8 per cent citrated human blood obtained from the blood bank, in Roux bottles. The cultures are pooled, should contain no bacteria and erythrocytes, and are sonically lysed for 90 sec in Raytheon sonic oscillator model DF01, 10 kc/s, 250 W, 115 V and 60 c/s. After spinning, the sediment and supernatant are treated with an equal volume of 88 per cent phenol for 30 min in the refrigerator. It is then centrifuged at 3,000-4,000 r.p.m., and the supernatant is then dialysed for 3 days in running tap water and for 24 h against distilled water. The sediment is precipitated with alcohol or is directly lyophilized. The phenol fraction is precipitated with 95 per cent alcohol (6-10 volumes); the sediment is washed in saline and then lyophilized. Gentle handling is