ABSTRACTS

Isoniazid Resistant and Dependent Strains of Mycobacterium Lepraemurium Studied in Vivo and in Vitro. P. D'ARCY HART, R. J. W, REES and R. C. VALENTINE. The J. of Path. & Bact., **84,** No. 1. 1962, pp. 105-111.

Since *Mycobacterium lepraemurium* does not multiply in cell-free medium the usual techniques for producing and demonstrating drug-resistant strains are not practicable in this species.

Mice were infected with *Myco. lepraemurium* and treated with isoniazid. The proportion of bacilli in the liver that were degenerate was assessed with electron microscope. This porportion increased at first but, in some animals, later decreased, suggesting that healthy drug-resistant bacilli were becoming dominant. A substrain of the bacilli that was passaged three times in further groups of treated and untreated mice appeared, from the trend of numbers of stainable bacilli in the liver and from the animals' survival times, to be not only drug resistant but also, to some degree, drug dependent; the dependence was strongest in the first passage from an isoniazid-treated animal.

A substrain from the third passage in isonizaid-treated mice showed elongation of the bacilli *in vitro* when these were incubated in a nutrient medium containing isoniazid in concentrations up to 5μg. per ml. This strain required a concentration of 25μg. per ml. for complete inhibition of the elongation. In contrast, an isoniazid-sensitive strain was completely inhibited by 1μg. per ml. The resistant strain showed evidence of slight isoniazid dependence *in vitro*.

Studies on Mycobacterium Lepraemurium in Tissue Culture: II. *The Production and Properties of Soluble Antigens from Myco. Lepraemurium in Tissue Culture.* R. J. W. REES and ROSEMARY D. TEE. Brit. J. exp. Path., **43,** No. 5, October 1962, pp. 480-487.

The present studies were undertaken on cultures of rat fibroblasts in which continuous multiplication of the rat leprosy bacillus *Myco. lepraemurium*, was maintained by subculturing the infected cells to fresh flasks every 20-30 days and changing the media every ten days. Filtered media were concentrated approximately 12-fold and screened, by agar-gel diffusion tests, for the presence of soluble mycobacterial antigens using a test rabbit anti-*Myco. lepraemurium* serum. From a total of 36 individual or pooled 10-day culture filtrates from five cultures of *Myco. lepraemurium* tested, 23 contained antigens which precipitated with the *Myco. lepraemurium* serum and which also reacted, though less strongly, with rabbit anti-*Myco. tuberculosis* serum.

Present investigations suggest that the mycobacterial antigen in the culture filtrates is predominantly polysaccharide since the

precipitation lines stain with periodic-acid Schiff reagent and the antigenicity of the culture filtrates is only slightly reduced by heating to 65° or by exposure to papain.

The evidence suggests that the mycobacterial antigen present in the culture filtrates is being actively produced by the multiplying bacilli because no antigen is present in filtrates harvested from comparable numbers of non-multiplying *Myco. lepraemurium*. Although antigen is present in the media none has been detected in the rat fibroblasts, wherein the bacilli are multiplying, indicating that the antigen rapidly diffuses out of the cells.

The culture filtrates which contain soluble mycobacterial antigens also elicited a delayed type reaction in the skin of guinea-pigs sensitized with BCG.