Lepr. Rev. (1978) **49**, 255–261

Abstracts

47. HARBOE, M., CLOSS, O., BJORVATN, G., KRONVALL, G. & AXELSEN, N. H. **Antibody response in rabbits to immunisation with *Mycobacterium leprae*.** *Infect. Immun.*, 1977, v. 18, No. 3, 792–805.

The authors' summary reads as follows:

"*Mycobacterium leprae* purified from liver tissue of an infected armadillo (the A/10 preparation) was tested for antigenic composition by immunization of rabbits and characterization of the antibody response by crossed immunoelectrophoresis. The rabbit antisera detected 7 distinct components in the *M. leprae* preparation. This number is far lower than in similar experiments with other mycobacteria. The *M. leprae* sonic extract gave far fewer lines after polyacrylamide gel electrophoresis and staining with Coomassie brilliant blue than sonic extracts prepared from BCG, *M. smegmatis,* and *M. phlei* adjusted to the same protein concentration based on the Folin assay. The 7 components detected in *M. leprae* cross-reacted extensively with *M. avium,* BCG, *M. lepraemurium, M. smegmatis,* and *Nocardia asteroides*. The 7 components are involved in immune reactions in leprosy; antibodies against all of them were demonstrated in sera from patients with lepromatous leprosy, but the specificity of the antibodies varied from patient to patient. The reason for the demonstration of so few antigenic components and some of the implications of these findings for the use of armadillo-grown *M. leprae* to develop specific skin test reagents and in other aspects of leprosy research are discussed".

[This article, which has 38 references, describes findings of major importance (for instance, to WHO's IMMLEP programme) and should be read in the original by anyone interested in this field of leprosy research.]

A. C. McDougall

48. KOLLER, W. C., GEHLMANN, L. K., MALKINSON, D. & DAVIS, F. A. **Dapsone-induced peripheral neuropathy.** *Arch. Neurol.,* 1977, v. 34, No. 10, 644–646.

The author's summary reads as follows:

"Peripheral neuropathy is a rare complication of dapsone therapy. This neuropathy appears primarily to be of the motor type, and recovery occurs on discontinuation of the drug therapy. The patient in this report developed a marked motor deficit as well as a selective marked loss of vibration sense shortly after the initiation of a relatively low dose of dapsone. Recovery was rapid on cessation of the therapy. This patient was found to be a slow acetylator of isoniazid, and therefore is probably a slow acetylator of dapsone. The possible mechanisms of the neurotoxicity of dapsone and the role of altered metabolism are discussed."

[Unlike some of the other 9 cases in the world literature, this is an excellently documented and referenced case, highly convincing for dapsone as the cause of the neuropathy reported. The total of 9 world cases is obviously very small indeed for a drug which has been used — in fact since 1950 — for the treatment of many hundreds, perhaps even a few thousand cases of dermatitis herpetiformis, and more recently for the treatment of various dermatological disorders of obscure aetiology. High dosage has often been used over a period of many years. The authors discuss possible mechanisms involved in dapsone neuropathy, drawing attention to the division of patients into fast and slow acetylators for dapsone, similar to that known to exist for isoniazid, adding that "the mechanism of the neurotoxicity of dapsone could be due to impaired metabolism with slow acetylation." They suggest that if slow acetylation is found to correlate

with the development of peripheral neuropathy in other cases, "it would be justifiable to determine the acetylation phenotype prior to initiating dapsone therapy". Apart from the impracticability of doing this, the fact that 50% of people are slow acetylators of dapsone and that the half-life of dapsone is not related to acetylator phenotype casts some doubt on the points made in the final paragraphs of this interesting report.]

A. C. McDougall

49. **Research activities of the National Institute for Leprosy Research, Higashi-murayama-shi, Tokyo, Japan.** Special issue for the 20th Anniversary, July, 1975.

This 257-page document in fact appeared in November 1977, and contains a detailed description of research work carried out at this centre since its foundation in 1955. The main section headings are: Cultivation and Metabolism, Transmission, Host–Parasite Relationships in Leprosy and Anti-leprosy Drugs. Several hundred experiments are described and pages 252–256 list those which have been published in the medical literature. Some of the projects are now of little more than documentary interest, but there is nevertheless a great deal of useful information, particularly under the headings of metabolism, culture and electron microscopy, which should be invaluable to those working in experimental leprosy.

A. C. McDougall

The Abstracts which follow are reprinted from Tropical Diseases Bulletin, *December 1977 and February and March 1978, through the courtesy of the Director, Bureau of Hygiene and Tropical Diseases. They are classified according to subject.*

I. MICROBIOLOGY

50. DESAI, A. C., APTE, S. N. & BHIDE, M. B. **The infectivity of drug resistant cases.** *Lepr. India,* 1977, v. 49, No. 1, 54–58.

Growth curves in the mouse footpad of *Mycobacterium leprae* derived from untreated patients with lepromatous leprosy are compared with those of proved dapsone-resistant *M. leprae* derived from patients after more than 5 years of dapsone therapy. The close similarity between the respective growth curves suggests to the authors that dapsone-resistant bacilli are as infective as their dapsone-sensitive counterparts.

T. F. Davey

51. DHOPLE, A. M. & HANKS, J. H. ***In vitro* growth of *Mycobacterium lepraemurium*, an obligate intracellular microbe.** *Science.* Washington, 1977, July 22, v. 197, 379–381.

This study of the limited *in vitro* growth of *Mycobacterium lepraemurium* was undertaken in the hope that the results would be of value to the culture of *M. leprae*. Firefly luminescence was used to measure the percentage of adenosine triphosphate (ATP) in a suspension or culture. Results with this ultrasensitive method confirmed that *M. lepraemurium* is capable of extra-cellular growth in diffusion chambers implanted in the peritoneal cavities of mice, and that growth is obtained in Nakamura's system. A 17-fold increase in mass was obtained after adaptation of the organism to growth *in vitro* in an improved modification of the latter method, though growth ceased after 6 weeks. A merit of ATP estimation as an index of metabolic activity for organisms such as *M. leprae* would be that data can be obtained immediately from unwashed host-grown organisms.

D. S. Ridley

52. KOHSAKA, K., MORI, T. & ITO, T. **Lepromatoid lesion developed in nude mouse inoculated with *Mycobacterium leprae*. Animal transmission of leprosy.** *Lepro,* 1976, v. 45, No. 3, 177–187.

Eight nude mice (BALB/c-*nu/nu*) were inoculated when 5 weeks old in the right hind footpad with 1×10^4 *Mycobacterium leprae* obtained from a relapsed lepromatous patient. The mice were kept under specific pathogen free (SPF) conditions in a Vinylplastic-isolator. The generation time of *M. leprae* was similar to that in control mice (in which Shepard-type limited multiplication occurred). However, spread outside the footpad was detected in a mouse killed at 13 months after inoculation. The 3 mice which survived to be killed at 17 to 22 months, all showed swelling of their inoculated footpads. Histological examination of the latter revealed lepromatous lesions with foamy histiocytes full of acid-fast bacilli; dermal nerves were involved and the histological figures published are compatible with *M. leprae* (compared with *M. lepraemurium*) infection. Lepromatous lesions were also detected in the eyelids, ears, nose and tail. Further proof that the lesions were due to infection with *M. leprae* was obtained in a number of ways, including failure of growth on artificial medium, and the results of the reaction in tuberculoid and lepromatous patients to lepromin prepared from a swollen footpad. Passage experiments are in progress in both *nu/nu* and normal mice.

[This is an important paper. Although the claim by Rees *et al.* to obtain lepromatous leprosy in thymectomized-irradiated mice (*Nature*, 1967, v. 215, 599) was confirmed by Job *et al.* (*Trop. Dis. Bull.,* 1975, v. 72, abstr. 1391), many workers have experienced great difficulty in keeping such mice alive for a sufficient length of time. Immunologically-deficient animals are required to study persisting viable *M. leprae* in treated leprosy patients, and the nude mouse kept under SPF conditions could prove to be an acceptable alternative.]

M. F. R. Waters

53. NAKAMURA, M., ITOH, T. & WAKI, C. **[Isolation of a cultivable mycobacterium from an armadillo subcutaneous tissue infected with *M. leprae* and characterization of this isolated strain.]** *Lepro,* 1976, v. 45, No. 4, 217–222. [In Japanese.]

The English summary appended to the paper is as follows:

"A strain of acid fast bacillus was isolated from a leproma of armadillo infected with *M. leprae* during the cultivation trial. Colonies were easily formed on Ogawa egg medium 1–2 weeks after inoculation, and were yellow.

"This isolated mycobacterium was identified as a type of Scotochromogen, which belonged to Group II atypical mycobacterium, by biological and biochemical characterizations."

54. ELLISTON, E. P. & TAYLOR, C. E. **Separation of *M. leprae* from human leproma and the development of a cytoplasmic skin test antigen from purified bacilli.** *Int. J. Lepr.,* 1976, v. 44, No. 3, 319–331.

With the object of producing a purified leprolin antigen that could be used for the epidemiological study of leprosy, leprosy bacilli were isolated without heating from a homogenized lesion by a flotation technique, digested enzymatically and sonicated. After repeated processing a dialysate (leprolin) was obtained that was free of cell wall material, and also a separate particulate fraction. These 2 antigens elicited positive skin test reactions in people with tuberculoid leprosy and negative reactions in lepromatous leprosy. The positive reactions were enhanced by the purification procedure. In a group of children not exposed to leprosy, the particulate antigens gave stronger reactions after BCG vaccination than in tuberculin negative children. Leprolin gave a low level response in both groups. Full evaluation was handicapped by limitation of supplies of antigen.

D. S. Ridley

55. MATSUO, E. & SKINSNES, O. K. **Immunologic identification of *M. leprae*. Immunofluorescence and complement fixation.** *Int. J. Lepr.,* 1976, v. 44, No. 3, 301–314.

An attempt was made to identify *Mycobacterium leprae* specifically by indirect immunofluorescent techniques. The antibody was the purified IgG fraction of serum of patients with lepromatous leprosy, adsorbed against *M. tuberculosis* and conjugated with fluorescein isothiocyanate. The organism, cultured by the authors in a hyaluronic acid based medium (LA-3) and thought to be *M. leprae,* gave strong fluorescence, especially about the periphery of the bacilli, but not all bacilli fluoresced. A similar result was obtained with bacilli in a cryostat section of a nodular lesion of lepromatous leprosy, but not with *M. lepraemurium, M. tuberculosis* or 12 other species of mycobacteria grown in LA-3 medium. *M. avium* produced faint fluorescence.

With complement fixation, a method based on antigen dilution gave more promising results than methods using serum dilution. The results seemed to correspond to those with immunofluorescence.

It was concluded that these methods were a means of specific identification of *M. leprae.* The specific antigen was considered to be a surface antigen with a lecithin-phospholipid component. The results strongly reinforced the claim that *M. leprae* is readily cultivated in LA-3 medium (*Trop. Dis. Bull.,* 1976, v. 73, abstr. 2053). Many technical details are given in the paper.

D. S. Ridley

56. OGAWA, T. **[Attempt at growth of *M. leprae* in mice.]** *Lepro,* 1976, v. 45, No. 4, 223—229. [In Japanese.]

The English summary appended to the paper is as follows:

"The technical procedures of experiment were explained briefly in Table 1. Bacterial suspension prepared from lepromas was injected into mice once or several times at weekly intervals by subcutaneous or intravenous route, or by both routes. Animals were killed at various intervals 2–16 months after injection. At necropsy, lesions were sought by gross inspection. Portions of various organs were removed and ground in mortar to make the homogenates. Smears made from the homogenate were stained by Ziehl-Neelsen's method and examined microscopically. The homogenate treated with 1% sodium hydroxide solution and then inoculated onto the egg yolk medium (for *M. lepraemurium*; also for *M. leprae* (?)) and Ogawa 1% egg medium (for cultivable mycobacteria). The tubes were incubated at 37°C for over 3 months. The details of single inoculation experiments and multiple inoculation experiments were shown in Tables 2 and 3.

"Ten experiments containing four with single inoculation and the other six with multiple inoculation were carried out. But one experiment, Expt. (4), exhibited a probable contamination and its results will be described in the following paper separately.

"In nine experiments, gross findings were all negative. Cultivation trials showed a few, smooth, and buff colonies, supposedly atypical mycobacteria, from two specimens only, but no colonies of mycobacteria, especially suspected of *M. leprae,* have been isolated.

"On the other hand, microscopic examination revealed the presence of acid-fast bacilli in the tissues of various organs. Among the two experiments, Expts. (3)–1 & –2, showed remarkable microscopical findings summarized in Table 6. As shown, in the subcutaneous experiment Expt. (3)–1, acid-fast bacilli found were in the injection site and superficial lymph nodes, and none of the tissue of viscera. In the intravenous experiment, Expt. (3)–2, acid-fast bacilli were detected in the spleen, liver and lungs, but smaller in number, comparing with those of the injection site just mentioned. No bacilli were found in the superficial lymph nodes. In both of the experiments the acid-fast bacilli had a tendency to decrease in number steadily. Where numbers of bacilli were present, globi were often seen, but these were usually small in size and loose in arrangement. And, indeed, it was uncertain whether the bacilli had multiplied within the tissue or not. The microscopic findings obtained in all the experiments were summarized in Table 7.

"As the materials of leproma used differed from experiment to experiment, it was impossible to compare directly the values of percentage for smear-positive specimens. As a whole, however, it seemed fairly justified in concluding that the microscopic findings were superior in the multiple

inoculation than in the single inoculation. This fact was accorded with the observations reported by previous workers."

[The tables are in English.]

57. NAKAMURA, M. **Multiplication of *Mycobacterium lepraemurium* in cell-free liquid medium. 10. Factors involved in the starting material of *M. lepraemurium* for the growth *in vitro* and *in vivo*.** *Lepro,* 1976, v. 45, No. 4, 203–210. **11. Establishment of the ND-5 medium.** *Ibid.,* 211–216. [In Japanese.]

The English summaries appended to the papers are as follows:

10. "Factors involved in the inoculum of *M. lepraemurium* for the growth in NC-5 medium as well as in mice were studied and the results obtained are as follows:

"1. Significant multiplications of *M. lepraemurium* obtained from infected subcutaneous tissue, liver, and spleen in NC-5 medium were observed. Therefore, it is obvious that the growth of bacilli in NC-5 medium is independent from the sources of the materials used. The multiplication ability of the bacilli in NC-5 medium is kept for 2 months at −20°C (in a freezer).

"2. No effects of treatments with 0.1% trypsin, 0.2% pronase, and 0.1% desoxycholate at 37°C for 60 min on the potentiality of the growth in NC-5 medium were recognized. The treatment with petroleum ether somewhat destroyed the ability.

"3. The growth rate of purified bacilli was superior than that of crude material.

"4. The potentialities of the growth of *M. lepraemurium* in NC-5 medium as much as in mice were completely destroyed by the treatment with below pH 6 at 37°C for 60 min and by heating at 50°C for 30 min. On the other hand, a complete destruction of the growth potentiality of bacilli in NC-5 medium was resulted by UV irradiation for 2.5 min, whereas the leproma producing ability in mice was maintained even by irradiation for 60 min."

11. "In order to improve the NC-5 medium, the Dubos medium (pH 7.3) was used as a basal medium, instead of Kirchner medium. The complete medium thus prepared is referred to as ND-5 medium. In this medium, *Mycobacterium lepraemurium* quickly multiplies in the form of binary fission without an extraordinary elongation. Possible generation times could be calculated by repeated experiments as 1.4–2.6 days. A slightly degenerative change in the cells during prolonged cultivation was observed by electron microscopy. This medium has some advantages for inhibiting other bacterial contaminations. Serial subcultivation is not tested yet."

[For earlier parts see *Trop. Dis. Bull.,* 1976, v. 73, abstr. 897.]

2. IMMUNOLOGY, PATHOLOGY

58. NIRMALA, V., CHACKO, C. J. G. & JOB, C. K. **Tuberculoid leprosy and tuberculosis skin—a comparative histopathological study.** *Lepr. India,* 1977, v. 49, No. 1, 65–69.

"Since it has been found hard to differentiate histopathologically tuberculoid leprosy from tuberculosis of the skin, a study of 20 biopsies from each of those conditions was undertaken to identify if possible some of their characteristic features.

"In tuberculoid leprosy along with tuberculoid granulomata there is always selective involvement and destruction of nerves, lack of fibrosis, absence of caseous necrosis and often epidermal atrophy. In cutaneous tuberculosis, on the other hand, in addition to tuberculous granuloma, there is often a proliferative reaction of the epidermis, areas of ulceration, absence of nerve destruction, marked increase in the reticulin, significant fibrosis and occasionally caseous necrosis."

59. WAHAL, P. E. *et al.* **Nephrotic syndrome complicating erythema nodosum leprosum (E.N.L.).** *J. Ass. Physns India,* 1977, v. 25, No. 6, 423–426.

"A case of lepromatous leprosy who developed renal amyloidosis with nephrotic syndrome as a complication of Erythema nodosum leprosum (ENL) reaction has been described. The remission

and exacerbation of clinical and biochemical picture of nephrotic syndrome coincided with subsidence and recurrence of lepra reaction. The case report emphasises the importance of early detection and treatment of E.N.L. episodes in lepromatous leprosy in an attempt to possibly prevent the development of this irreversible grave complication in these cases."

60. SINGH, T., KAUR, S., KUMAR, B., SAWHNEY, B. B. & CHOPRA, J. S. **A study of motor and sensory nerve conduction in leprosy.** *Indian J. Med. Res.,* 1977, v. 65, No. 5, 632–639.

"Motor and sensory nerve conduction velocities were studied in ulnar, median, lateral popliteal and posterior tibial nerves in 40 patients with leprosy and compared with 50 age-matched controls. The conduction velocity was found to be decreased in all varieties of leprosy and in all segments of the nerves. Lateral popliteal nerve was found to be the most frequently involved nerve. A clinico-electrophysiological correlation was found between nerve involvement clinically in the form of thickening of nerve weakness and wasting of muscles supplied by the nerve and the degree of conduction abnormality. Motor and sensory nerve conduction velocities were found to be equally affected in the neuropathy of leprosy. The study did not substantiate the presumption that sensory nerve conduction is more affected than motor conduction. It is suggested that for an evaluation of the severity of leprosy polyneuritis, nerve conduction velocity and distal delay especially for the motor nerves should both be tested."

61. KARAT, A. B. A. & RAO, P. S. S. **Haematological profile in leprosy. Part I—General findings.** *Lepr. India,* 1977, v. 49, No. 2, 187–196.

"Haematological studies in 904 adult leprosy patients with different types of leprosy, in various stages of the disease and treatment are described. Haemoglobin, packed cell volume, serum albumin and serum iron are significantly lower among lepromatous leprosy patients as compared with non-lepromatous patients. The serum B12 levels were significantly higher among the lepromatous group. Acid fast bacilli have been demonstrated in skin smear negative leprosy patients with indeterminate and tuberculoid leprosy, suggesting occurrence of bacillaemia in these groups of patients."

The discovery of acid-fast bacilli in bone marrow in 5% of men with indeterminate leprosy and in 4.3% of men with tuberculoid leprosy is of particular interest.

T. F. Davey

62. EL SHIEMY, S., EL HEFNAWY, H., FATTAH, A. A., EL HAWARY, M. F. & FARES, R. **Muscle invovlement in leprosy and its correlation with serum aldolase activity.** *Int. J. Derm.,* 1977, v. 16, No. 7, 587–593.

"Thirty-six leprosy patients underwent muscle biopsy; the specimens were studied for serum aldolase activity. The authors concluded that muscle degeneration occurs only in lepromatous leprosy due to direct invasion by leprosy bacilli increasing serum aldolase activity during the active degenerative phase of the muscle fibers."

3. CLINICAL

63. VACHHARAJANI, S. D., RASTOG, D. S., ARORA, P. N. & SOHI, A. K. **Leprosy in tuberculosis.** *Indian J. Tuberc.,* 1977, v. 24, No. 3, 135–136.

"The present article reports four cases of leprosy one lepromatous and three tuberculoid types. In all these cases, the leprosy was detected in confirmed cases of pulmonary tuberculosis who were on antituberculous drugs for varying intervals of 8–20 weeks.

"It is presumed that leprosy became active and manifest while pulmonary tuberculosis was active and being treated. This perhaps casts doubt about the antigenic similarity between the

tubercle and leprosy bacilli. It is emphasised that in a TB Hospital, careful search for detecting leprosy among its patients should be made periodically, even though the association of the two diseases is not very frequent."

4. THERAPY

64. HAMILTON, J. M. **The place of electrical stimulation in the physiotherapy of leprosy.** *Lepr. India*, 1977, v. 49, No. 2, 197–206.

"The production of nerve and muscle impulses by faradic and interrupted direct current, and the 'strength-duration curves' plotted for normal, denervated, and partially denervated muscles, are described. The advantages and disadvantages of such electrical stimulation in the testing of recent paralysis, the treatment of recent paralysis, and following tendon transfer surgery, in leprosy patients, are discussed. In the light of these, electrical stimulation is concluded to have only a minor role in the physiotherapy of leprosy."

65. KERHARO, J. La pharmacopée sénégalaise: note sur quelques traitements antilépreux traditionnels pratiqués dans le Baouar (préfecture de Kebemer). **[Senegalese pharmacopoeia: notes concerning certain traditional antileprous treatments practised in Baouar (prefecture of Kebemer).]** *Bull. Soc. Méd. Afr. Noire Lang. Fr.,* 1977, v. 22, No. 3, 321–329. English summary.

5. EPIDEMIOLOGY, PREVENTION, CONTROL

66. NAIK, S. S. & GANAPATI, R. **Regularity of dapsone intake by leprosy patients attending urban treatment centre.** *Lepr. India,* 1977, v. 49, No. 2, 207–215.

"Dapsone/Creatinine in urine ratios were determined in statistically randomised samples of 965 leprosy patients attending out-patient department of Acworth Leprosy Hospital and 44 inmates of the Hospital. The percentage of irregularity in DDS treatment was found in 43 and 22.6 respectively in out-patients and inmates of the Hospital. The need to assess the possible response for irregularity in treatment is stressed and the hazard of infectious cases remaining without treatment or with incomplete treatment is pointed out."

[The authors followed the methods described by Low and Pearson, *Trop. Dis. Bull.,* 1974, v. 71, abstr. 2797.]

6. REHABILITATION AND SOCIAL ASPECTS

67. MUTATKAR, R. K. **Health education in leprosy. An evaluation.** *Lepr. India,* 1977, v. 49, No. 2, 234–239.

This is an evaluation by the University of Poona of health education programmes related to leprosy pioneered in that city by the Gandhi Memorial Leprosy Foundation, and undertaken on behalf of the Foundation. Two areas are compared, and random sampling methods employed. The study indicates that the methods employed have effectively changed both attitudes and behaviour of the people towards leprosy, but the process is slow and needs repeated contact between educator and people in a continuous programme.

T. F. Davey