CURRENT LITERATURE

This department carries selected abstracts of articles published in current medical journals dealing with leprosy and other mycobacterial diseases.

General and Historical

Irgens, L. M. The discovery of *Mycobacterium leprae.* A medical achievement in the light of evolving scientific methods. Am. J. Dermatopathol. **6** (1984) 337–343.

The discovery of *Mycobacterium leprae* by G. H. Armauer Hansen (1841–1912) in 1873 represents a link in a chain of development in international medicine that was influenced by two main concepts, namely, that germs may be causes of disease and that social conditions may be related to disease, either as causes or consequences or both. Hansen's work is also a link in a chain of research on leprosy in Norway. Hansen met with serious challenges in addition to those that were purely scientific. To prove the causative effect of the microorganism according to principles that later came to be known as Koch's postulates, Hansen felt compelled to conduct experiments that were deemed illegal by the authorities. At the same time, he was fighting to establish priority for his discovery. Later, Hansen was honored as the discoverer of the bacillus of leprosy and was privileged to see benefits in the public health as a consequence of the discovery. Hansen's epoch-making achievements may serve as an inspiration to all who aspire to combat disease by seeking answers to questions about causes of disease, modes of prevention, and social consequences of disease.—Author's Abstract

Manchester, K. Tuberculosis and leprosy in antiquity: An interpretation. Med. Hist. **28** (1984) 162–173.

The increased tuberculous infection in the community, probably of the human-type pulmonary lesions, induced an immunity in the survivors which prevented the superinfection by the allied *Mycobacterium leprae.* In the past, just as today, the majority of individuals, mostly young children, would overcome the primary tubercle invasion, would recover, and would henceforth be immune to leprosy. Pulmonary tuberculosis, being a population density-dependent disease, owes its medieval increase to urbanization or at least to the development of aggregate population groups. Both diseases, therefore, may owe their changing incidence in medieval England to a phenomenon of human social development.

Leprosy affects an estimated 12–15 million people today, and tuberculosis is also very prevalent. That they were of collective and individual significance in earlier communities is without question. It is proposed that these two infectious diseases were closely interrelated in antiquity.—(*From* the Article)

Riley, D. N. Treatment of leprosy in rural India as seen on a medical student elective. Lepr. Rev. **55** (1984) 397–402.

A personal report on the community and hospital treatment of leprosy in the rural villages of South India. Details are given of the present drug regimens and the practical difficulties encountered in implementing them successfully.—Author's Summary

Trautman, J. R. A brief history of Hansen's disease. Bull. N.Y. Acad. Med. **60** (1984) 689–695.

Thousands of historical events have been related to Hansen's disease, some more important than others, but it is not possible to rank them in the order of importance or to achieve anything resembling a consensus. However, four milestones which most persons knowledgeable about the disease deem of great importance are the discovery of *Mycobacterium leprae* by Dr. G. Armauer Hansen of Norway in 1874; initial use of

sulfone therapy by Dr. Guy Faget of Carville in 1941; discovery that the mouse foot pad supported the multiplication of *M. leprae* by Dr. Charles Shepard of the Centers for Disease Control in 1959; and the demonstration that the nine-banded armadillo is highly susceptible to developing disseminated Hansen's disease after inoculation with *M. leprae* by Dr. Waldemar Kirchheimer of Carville and Dr. Eleanor Storrs of the Gulf South Research Institute in 1968.—(*From* the Article)

Chemotherapy

Anderson, R. Enhancement by clofazimine and inhibition by dapsone of production of prostaglandin E_2 by human polymorphonuclear leukocytes *in vitro*. Antimicrob. Agents Chemother. **27** (1985) 257–262.

The effects of the antileprosy agents clofazimine and dapsone (1 to 10 μg/ml) on the spontaneous and stimulated release of prostaglandin E_2 (PG E_2) by human polymorphonuclear leukocytes (PMNL) *in vitro* have been investigated. PMNL were obtained from normal adult volunteers and three patients with leprosy (two borderline lepromatous and one subpolar lepromatous leprosy). The synthetic chemotactic tripeptide *N*-formyl-L-methionyl-L-leucyl-L-phenylalanine (FMLP) at a concentration of 10^{-7} M was used as the stimulant of PG E_2 synthesis. None of the test agents at the concentrations used inhibited the binding of radiolabeled FMLP to PMNL. However, dapsone at 5 and 10 μg/ml inhibited the spontaneous and FMLP-induced release of PG E_2 by PMNL. Clofazimine, on the other hand, significantly increased both the spontaneous and the FMLP-induced synthesis of PG E_2 by PMNL. The enhancing effects of clofazimine on FMLP-mediated synthesis of PG E_2 were particularly striking and were observed at concentrations of 1 to 10 μg of the drug per ml. Measurements of PMNL spontaneous and FMLP-induced synthesis of PG E_2 in the presence of both clofazimine and dapsone (5 μg/ml) indicated that the two drugs are mutually antagonistic. PMNL from both normal control subjects and patients with leprosy were equally sensitive to these effects of clofazimine and dapsone. The immunostimulatory and immunosuppressive properties of dapsone and clofazimine, respectively, may be related to the opposite effects of these agents on PG E_2 synthesis in human leukocytes.—Author's Abstract

Bhatia, V. N., Balakrishnan, S., Seshadri, P. S., Neelan, P. N. and Roy, R. G. Dapsone resistance in patients attending Central Leprosy Teaching and Research Institute, Chengalpattu (South India). Indian J. Lepr. **56** (1984) 587–594.

The Central Leprosy Teaching and Research Institute (CLTRI), Chengalpattu, took up studies on dapsone resistance in *Mycobacterium leprae* from 1974. From 1978, the study was further strengthened by a project under THELEP (TDR) for eliciting information on the efficacy of certain drug regimens. The THELEP studies were to be conducted only on the dapsone-sensitive untreated cases and, therefore, directed toward the detection of primary resistance; while the non-THELEP institutional studies were concentrated on the secondary dapsone-resistance problem. These two studies together detected 99 cases of dapsone resistance in the patients who attended CLTRI Hospital during 1974–1981; 23 of them were of primary origin, 16, 6 and 1 showing mild (RI), moderate (RII), and high (RIII) grades, respectively. Of the remaining 76 cases of secondary resistance, 7 and 69 were of RII and RIII grades, respectively. The need for vertical and horizontal monitoring of the drug resistance problem has been pointed out.—Authors' Abstract

Foucauld, J., Uphouse, W. and Berenberg, J. Dapsone and aplastic anemia. (Letter) Ann. Intern. Med. **102** (1985) 139.

There are several reports of agranulocytosis and erythrocyte aplasia after administration of dapsone for treatment of leprosy and for prophylaxis against falciparum ma-

laria. We report what we believe is the first reported case of aplastic anemia due to administration of dapsone.

In our review of the literature, we could find no cases of aplastic anemia due to dapsone. Direct toxicity to the bone marrow as shown by aplastic anemia is postulated. Dapsone is widely used throughout the world. We recommend that frequent hematologic monitoring be done and the drug discontinued at first sign of toxicity.—(*From* the Letter)

Gelber, R. H., Henika, P. R. and Gibson, J. B. The bactericidal activity of various aminoglycoside antibiotics against *Mycobacterium leprae* in mice. Lepr. Rev. **55** (1984) 341–347.

The killing potential of various aminoglycoside antibiotics for *Mycobacterium leprae* infection of the mouse foot pad was studied, utilizing daily intraperitoneal therapy. Kanamycin (100 mg/kg), streptomycin (150 mg/kg), and amikacin (100 mg/kg) resulted in impressive killing of bacilli (99.7%, 97% and 96% bactericidal, respectively). Gentamicin (20 mg/kg) and tobramycin (20 mg/kg) were much less active (60% and 37% bactericidal). The bactericidal activity of these very high doses of kanamycin, streptomycin, and amikacin compared favorably with those of other agents previously studied in a similar manner at relatively lower dosage levels.—Authors' Summary

González Vázquez, R. Sulfonorrestencia y recaida en lepra. [Sulfone resistance and relapse in leprosy.] Dermatol. Rev. Mex. **28** (1984) 41–50. (in Spanish)

Dapsone (DDS) is still considered the basic treatment in leprosy, which is why appropriate measures should be taken in order to avoid the development of primary or secondary resistance to the drug using combined therapy.

The resistance to DDS and the relapse of leprosy are problems in the management of lepromatous leprosy, mostly in developing countries.

Two mechanisms through which lepromatous leprosy under sulfone treatment can recur are proposed: endogenous and exogenous reinfection. The first one occurs as a result of multiplication of bacilli resistant to DDS or mutant bacilli with partial resistance to it or due to persistent (dormant) bacilli present in the patient's organs. The second mechanism is a second infection, but this is less likely because previous cure is implied.—Author's English Abstract

Gupta, P. K., Luniya, A. K., Gupta, N. K. and Tiwari, M. L. Acute severe peripheral neuropathy due to thiacetazone. Indian J. Tuberc. **31** (1984) 126–127.

A patient with pulmonary tuberculosis developed acute peripheral neuropathy within 15 minutes of starting chemotherapy. Subsequently, thiacetazone was found to be the offending drug.—AS/D. J. Girling (*From* Trop. Dis. Bull.)

Joshi, J. V., Maitra, A., Sankolli, G., Bhatki, S. and Joshi, U. M. Norethisterone and ethinyl estradiol kinetics during dapsone therapy. J. Assn. Physicians India **32** (1984) 191–193.

Single dose kinetics of norethisterone (NET) and ethinyl estradiol (EE) were investigated in ten female leprosy patients following the ingestion of a contraceptive pill (NET acetate 1 mg + EE 30 μg). They had been under treatment with dapsone (100 mg daily) for 5–20 years. Six young healthy women were similarly studied as controls. Serum concentrations up to 24 hours were measured by sensitive and specific radioimmunoassays, the intra- and interassay coefficients of variation being less than 13%. Peak levels and area under concentration time curve (AUC 0–24 hr) for NET were similar in leprosy patients and in healthy controls (peak level 11.3 $\pm$ 6.4 ng/ml vs 10.5 $\pm$ 4.7 ng/ml; AUC 89 $\pm$ 38 ng/ml·hr vs 67 $\pm$ 9 ng/ml·hr). Peak levels and AUC (0–8 hr) for EE were higher in leprosy patients as compared to the controls (peak levels 184 $\pm$ 66 pg/ml vs 128 $\pm$ 25 pg/ml; AUC 1041 $\pm$ 319 pg/ml·hr vs 681 $\pm$ 123 pg/ml·hr). There was a tendency toward higher concentrations in leprosy patients although it was significant only with respect to AUC for EE. Chronic treatment with dapsone does not appear to reduce the bioavailability of NET or EE. Oral contraceptives therefore may be effectively used in

leprosy patients under dapsone therapy.—Authors' Summary

Keeler, R. F. Multidrug therapy for leprosy in Trinidad and Tobago: A preliminary report. Lepr. Rev. **55** (1984) 391–396.

Dual chemotherapy for multibacillary patients was introduced in Trinidad and Tobago in 1971, using clofazimine and dapsone. Since 1973, newly diagnosed multibacillary patients have received triple therapy, at first only for a few weeks, but later for three months, adding rifampin 600 mg daily to the clofazimine and dapsone already in use. In January 1982, the short-course regimens recommended by the World Health Organization (WHO) (Technical Report series, 1982) were introduced and after a period of 21 months, 531 patients had completed their courses of treatment. This paper reports preliminary results in this group.—Author's Summary

Levis, W. R. Treatment of leprosy in the United States. Bull. N.Y. Acad. Med. **60** (1984) 696–711.

Current recommendations for treatment of leprosy in the United States differ from World Health Organization (WHO) guidelines. Daily rifampin and dapsone in combination are recommended in the United States, and mouse foot pad antibiotic sensitivity testing should be routinely obtained on all multibacillary cases when possible. Clofazimine is used in reactional premenopausal women, drug-resistant patients, and patients for whom either rifampin or dapsone are contraindicated. The emergence of increasing drug resistance requires multidrug antibiotic therapy. Drug resistance is the underlying factor behind both the American and WHO recommendations. There is a need for new antibiotics in the treatment of leprosy as well as improved methods of screening and monitoring. Current recommendations for multidrug therapy may provide effective control of leprosy. Alternatively, dapsone resistance may continue to increase. Secondary resistance followed by primary resistance to other antileprosy antibiotics may also develop, as it has for dapsone. Despite the limited number of effective antibiotics and emerging drug resistance at the present time, it is possible effectively to treat most patients with leprosy to the point that they can lead productive lives in an ambulatory care setting. While the duration of therapy, coexistent social problems, variable degrees of disability, and associated medical problems will challenge health care personnel, results are unusually rewarding for both the patients and the health care team.—Author's Summary

Mester de Parajd, L. and Garnier, J.-P. Nutritional aspects of leprosy. Acta Leprol. (Genève) **2** (1984) 293–303.

Deoxyfructo-serotonin (DFS) being a naturally occurring metabolite, which, like serotonin, has its origin in tryptophan in food, one can ask about the role of nutrition in leprosy.

The unique situation, that a human metabolite shows antileprosy activity, confirmed *in vitro* and *in vivo,* makes it possible to develop a "built in" antileprosy therapy. This is now realized through a so-called "AntiLeprosy Nutriment" (NAL). The effectiveness of this diet was confirmed by the fact that a daily dose of 0.5 g NAL per mouse has a similar effect to 20 mg/kg body weight dapsone per day in the conventional mouse foot pad test.

The biosynthesis of DFS in man has been demonstrated by high performance liquid chromatography (HPLC), spectrofluorometry and mass spectrometry. The activity of NAL (rich in tryptophan, unsaturated fatty acids and glucose) is due to an increased biosynthesis of DSF. The latter may be considered as a "physiological protecting agent" against leprosy. This type of food may play a role in the prevention of leprosy in endemic areas.—Authors' Summary

Mohamed, K. N. Hypersensitivity reaction to dapsone: Report from Malaysia. Lepr. Rev. **55** (1984) 385–389.

Three patients who developed hypersensitivity reaction at varying intervals after the initiation of dapsone therapy are described. Two of them had leprosy whereas the other had dermatitis herpetiformis. Their clinical manifestations were not uniform. Although fatal complications have been reported, the unique role played by dapsone in the treatment of leprosy and the

importance of recognizing this reaction are discussed.—Author's Summary

Nêgrel, A. D., Chovet, M., Baquillon, G. and Lagadec, R. Clofazimine and the eye: Preliminary communication. Lepr. Rev. **55** (1984) 349–352.

Fifty-seven patients, admitted to the Marchoux Institute in Bamako, Mali, were treated with clofazimine for periods varying from between 3 and 26 months. Detailed ophthalmological examination was carried out in all cases, including visual acuity; conjunctival smears for crystals; corneal sensation; the ocular fundus after pupillary dilatation. In addition, slit-lamp examination was carried out in all cases. In this preliminary study, apart from brown-red subepithelial pigmentation in the cornea, no untoward effects on the eye were recorded.—Authors' Summary

Niwa, Y., Sakane, T., Miyachi, Y. and Ozaki, M. Oxygen metabolism in phagocytes of leprotic patients: Enhanced endogenous superoxide dismutase activity and hydroxyl radical generation by clofazimine. J. Clin. Microbiol. **20** (1984) 837–842.

We examined the generation of active oxygens (O_2^-, H_2O_2, and OH·) and the superoxide dismutase (SOD) activity of polymorphonuclear leukocytes (PMNs) and monocytes from 14 leprosy patients manifesting a bacterial index above 2.2. Patients with disease of more than four years in duration showed significantly enhanced SOD activity and a decrease in O_2^- and OH· production. The antileprotic agent, clofazimine, significantly increased the generation of OH· in a dose-dependent manner, with a subsequent decrease in H_2O_2, but had no effect on the SOD activity of the PMNs and monocytes. In medium containing $FeSO_4$ or Fe^{2+}-EDTA, the drug elevated OH· production markedly further. Phagocytic SOD in PMNs and monocytes of leprosy patients was both host and bacillus derived, because the presence of cyanide, to which human-derived cuprozine SOD is susceptible, did not completely abrogate SOD activity. The difficulty in treating leprosy may be partly ascribable to decreased phagocytic OH· generation, which in leprosy patients is apparently due to the uptake of Hansen bacillus-derived SOD. Clofazimine may be effective in leprosy by chelating Fe^{2+}, with the resultant potentiation of the catalyzing activity of Fe^{2+} in the Haber-Weiss reaction increasing OH· formation from H_2O_2.—Authors' Abstract

Pattyn, S. R., Yada, A., Sansarricq, H. and Van Loo, L. Prevalence of secondary dapsone-resistant leprosy in Upper Volta. Lepr. Rev. **55** (1984) 361–367.

A secondary dapsone-resistance survey was performed in three health sectors of Upper Volta in 1981–1982, among a total population of 994 lepromatous patients. Prevalence of secondary dapsone resistance was found to be 7%, 4%, and 1%, respectively. Analysis of the results reveals that considerable loss of viability of *Mycobacterium leprae* occurred in the specimens during transportation and was much more pronounced in the two sectors with the lower dapsone-resistance prevalence figures. It is therefore strongly suspected that the 7% prevalence is also representative of the sectors where the low prevalence figures were found. For other reasons discussed, the 7% figure must in its turn be a minimum.—Authors' Summary

Schwab, B. W., Paldino, A. M., Matthiessen, T. and Spencer, P. S. Rabbit sural nerve responses to chronic treatment with thalidomide and supidimide. Muscle Nerve **7** (1984) 362–368.

Chronic treatment of rabbits with thalidomide (3-(N-phthalimido) glutarimide) produced progressive decrements in sural nerve conduction velocity (NCV) that were unassociated with qualitative or quantitative morphological changes of nervous tissue. Three groups of eight rabbits received 100 mg/kg/day thalidomide (group I), 200 mg/kg/day supidimide (a related drug) (group II), or a carboxymethylcellulose vehicle (group III) 5 days/week for 40 weeks. At 6 months of treatment, a noninvasive determination of the mean maximum sural NCV for group I was significantly reduced relative to the conduction velocities of groups II and III. Direct measurement of conduction velocity when treatment was terminated confirmed these findings and

demonstrated similar conduction deficits in proximal and distal portions of the sural nerve in group I animals. At 6 months, rabbits in group II showed a significant reduction in mean conduction velocity and, at the termination of treatment, they displayed mean values similar to those of group III and significantly greater than those of group I. Morphological findings were unremarkable in 20 regions of the central nervous system (CNS) and the peripheral nervous system (PNS) known to display changes early in toxic neuropathies. Morphometric estimation of unmyelinated and myelinated fibers in the sural nerve at the heel revealed no between-group differences in axon diameter, fiber diameter, g-ratio (the ratio of inside/outside diameters), or internodal length. In conclusion, chronic treatment with thalidomide produces selected decrements in sural nerve function that have an unknown relationship to the poorly reversible sensory neuropathy reported in humans receiving this drug.—Authors' Abstract

Clinical Sciences

Agarwal, S. and Aggarwal, S. K. Electrocardiographic changes in multibacillary leprosy. Indian J. Lepr. **56** (1984) 569–574.

Fourteen cases of multibacillary leprosy (BL and LL) were investigated biochemically and EKG recording was done. Serum cholesterol was found to be raised in 4 out of 14 cases and SGOT was raised in 2 cases. P wave, PR interval and QRS complex did not show any changes; ST segment was depressed in 2 cases; T wave was flat in 2 cases and inverted in 1 case and Q-Tc interval was prolonged (more than 0.40 secs) in 3 cases.—Authors' Abstract

Bobhate, S. K., Sisodia, S. M. and Kherdekar, M. A case of squamous cell carcinoma in plantar ulcer in leprosy. Indian J. Lepr. **56** (1984) 653–655.

A case report of squamous cell carcinoma in plantar ulcer of a leprosy patient is presented.—Authors' Abstract

Ekambaram, V., Naidu, B. and Prabhakara Rao, V. Differential diagnosis between leprosy and post-kala-azar dermal leishmaniasis. Indian J. Lepr. **56** (1984) 641–646.

A case of post-kala-azar dermal leishmaniasis misdiagnosed and treated as a case of leprosy is reported. It is stressed that to avoid such errors, it is essential that even in so-called "typical cases" of leprosy a thorough routine clinical and bacteriological examination is necessary for diagnosis of leprosy.—(*From* the Article)

Fleury, R. N. and Opromolla, D. V. A. Carcinoma in plantar ulcers in leprosy. Lepr. Rev. **55** (1984) 369–378.

The authors studied carcinoma arising in plantar ulcers (PUs) in 16 patients having leprosy, admitted to the Lauro de Souza Lima Hospital (Bauru, São Paulo, Brazil) from 1970 to 1982. In a study of the evolutional, clinical, and pathological aspects, the main conclusions of interest were:

The apparent rarity of these neoplasms must be related to the fact that many observed cases are not recorded. The carcinomas occur chiefly in patients of the borderline group. The carcinomas have in general an ulcero-vegetating appearance, large extension and depth, and preferential situation on the proximal third of the sole of the foot. Having in mind the low frequency of lesions of PUs in the foot proximal third, the incidence of carcinoma in this site can be considered relatively high. From the histological point of view, the majority of tumors were well differentiated, and showed peculiarities similar to those in verrucous carcinoma, in giant condyloma acuminata and in epithelioma coniculatum plantare. The great tendency to extension of these neoplasms and the possibility of regional metastasis justify treatment by amputation.—Authors' Summary

Gonén, B., Blank, A., Shochina, M., Wolf, J. and Sheskin, J. El calculo "T menos H" como indicativo del estado sensorial del segmento distal del nervio tibial en la hanseniasis. [Calculating "T minus H" as an indicator of sensory conduction of the

distal segment of the tibial nerve in hanseniasis.] Actas Dermosifiliogr. **75** (1984) 247–250. (in Spanish)

The tendon reflex (T) provides an estimate of the sensory and motor function of the tibial nerve, between the Achilles tendon and the spinal cord. The Hoffman reflex (H) provides an estimate of sensory and motor function of the segment of the tibial nerve between the popliteal fossa and the spinal cord. From the results of these two measurements, one may calculate "T minus H" which provides an estimate of the sensory function, only, of the segment of the tibial nerve that lies between the Achilles tendon and the popliteal fossa. These procedures are easily applied: they are less traumatic than those procedures previously employed to measure the function of the tibial nerve because one of them employs simply a reflex-hammer and the second, superficially placed electrodes rather than needles.—Authors' English Summary

Hobbs, E. R. and Hempstead, R. W. Cutaneous coccidioidomycosis simulating lepromatous leprosy. Int. J. Dermatol. **23** (1984) 334–336.

This patient demonstrated an unusual cutaneous presentation of disseminated coccidioidomycosis: diffuse inflammatory infiltration of the pinnae, which in some respects mimics the clinical presentation of lepromatous leprosy. The presence and nature of the plaques, papules, and nodules, as well as slight madarosis, contributed to the ruse. We suggest that the differential diagnosis of leprosy, particularly in areas where *Coccidioides immitis* is indigenous, include disseminated coccidioidomycosis.—(*From* the Article)

Jain, A. P., Gupta, O. P., Jajoo, U. N. and Kumar, K. A study of leprosy in relation in HBsAg. Indian J. Lepr. **56** (1984) 575–577.

In the present study presence of HBsAg in the serum of 130 leprosy patients (LL 50, ENL 30, TT 50) and its relationship with various complications has been studied. The HBsAg positivity in controls and various types of leprosy did not show any significant difference. Similarly, complication also had no relation with HBsAg positivity, so we conclude that leprosy has no relation with HBsAg.—Authors' Abstract

Kale, H. D., Zawar, P. C., Chawhan, R. N. and Kulkarni, G. R. Cardiac dysautonomia in lepromatous leprosy. Indian J. Lepr. **56** (1984) 563–568.

Cardiovascular autonomic functions were studied in 32 patients with lepromatous leprosy, including 12 patients with lepra reaction. Fifty age- and sex-matched healthly subjects served as controls. Variable degrees of autonomic dysfunction were observed in the study group. The derangement of autonomic functions in patients with leprosy can be explained on the basis of neurotrophic action of lepra bacilli that infiltrate the sympathetic and the parasympathetic fibers.—Authors' Abstract

Kanwar, A. J., Bharija, S. C. and Belhaj, M. S. Renal functional status in leprosy. Indian J. Lepr. **56** (1984) 595–599.

Renal functional status was evaluated in 27 patients with lepromatous leprosy. Renal functions were found to be significantly impaired in lepromatous patients with erythema nodosum leprosum (ENL) in the active phases. Patients in quiescent phases and uncomplicated lepromatous leprosy patients did show renal impairment, however the degree of impairment in such cases was less than that in the reactive cases.—Authors' Abstract

Kumar, A., Durai, V. and Sirumban, P. Diagnostic efficiency of paramedical workers involved in leprosy case detection program. Indian J. Lepr. **56** (1984) 626–632.

The diagnostic efficiency and accuracy in classification of leprosy and its activity status, by four senior, trained paramedical workers (PMWs) involved in the leprosy case detection program was assessed on 1394 cases detected by them and concurrently confirmed by an experienced medical officer. The inter-observer variation between two experienced PMWs in diagnosis and classification of leprosy on 216 patients was also studied. Of the 1394 cases detected by PMWs, 257 (18.44%) were wrongly diag-

nosed as leprosy, mostly as nonlepromatous (N) type. Although all lepromatous (L) and 98% of N-type cases were correctly classified by PMWs, 25.64% of borderline (N ? L) cases were either under-diagnosed as N-type (17.95%) or over-diagnosed as L-type (7.69%). The activity status of 19% of the cases was wrongly assessed by PMWs, including 8% active lesions assessed as inactive. The discrepancy between two PMWs in diagnosis, classification and assessment of activity status of leprosy was found in 1.39%, 7.41%, and 25.67% cases, respectively. The implications of these observations, and the suggestions to improve the technical skills of workers for an efficient and effective implementation of the leprosy control program, are discussed.—Authors' Abstract

Kumar, K. and Kant, M. Squamous cell carcinoma developing in trophic ulcers in leprosy—a case report. Indian J. Lepr. **56** (1984) 656–657.

Malignant transformation of trophic ulcer in leprosy, though not very uncommon, is ill-reported in the literature. Not more than a dozen cases have been reported so far. A case of squamous cell carcinoma developing in a trophic ulcer in leprosy is presented. The histopathology has been discussed.—Authors' Abstract

Laing, A. B. G., Millard, P. R. and McDougall, A. C. *Mycobacterium leprae* within a squamous cell carcinoma. (Letter) Am. J. Dermatopathol. **6** (1984) 413–414.

An Asiatic Indian was found to have lepromatous leprosy in 1949 at the age of 27 years and was treated with dapsone by mouth. By 1954, his disease was apparently well controlled and therefore he was discharged from the hospital but advised to continue treatment as an outpatient. In 1979, he relapsed with extremely active and extensive lesions of lepromatous leprosy that were positive in slit-skin smears from several sites.

He was treated with rifampin, thiambutosine, and clofazimine and responded well, but in 1980 was found to have a huge growth that was clinically suspected of being malignant at the site of a preceding ulcer on the right heel. Biopsy revealed squamous cell carcinoma and a below-knee amputation was done. Within a very short period of time new masses appeared at the very site, which were once again malignant in appearance. Again biopsy of one of the new lesions showed well-differentiated squamous cell carcinoma. Sections of this material were also stained with the Fite–Faraco modification of the Ziehl–Neelsen stain for acid-fast bacilli. To our surprise, lepra bacilli were found not only in the adjacent dermis but also in the actual substance of the squamous cell carcinoma. Many bacilli lay between neoplastic cells, usually singly, but occasionally in small groups. An examination of serial sections under oil immersion by several observers suggested that occasional organisms lay in the cytoplasm of neoplastic cells.

The close association of lepra bacilli with the cells of a malignant growth has not, to our knowledge, been described in the literature.—(*From* the Letter)

Misra, R. S., Jain, R. K. and Mukherjee, A. Vitiligo on tuberculoid patches—a case report. Indian J. Lepr. **56** (1984) 658–661.

A case of tuberculoid leprosy with complete depigmentation on its active margins in a predisposed individual to vitiligo is presented. The postulated mechanism of hypopigmentation vis-à-vis leprosy lesions is discussed in brief in the context of the unusual presenting association.—Authors' Abstract

Nsibambi, J., Berhan, T. Y. and Warndorff van Diepen, T. Multibacillary leprosy in an 18-month-old child: A case report. Lepr. Rev. **55** (1984) 379–383.

A case of multibacillary leprosy, proven on slit-skin smears and skin biopsy, is reported in a child aged 18 months in Ethiopia. The father and two other children were not available for examination, but the mother was a registered case of mid-borderline (BB) leprosy of five years' duration. The clinical, bacteriological, and histopathological findings are described and discussed in relation to the accepted incubation period of leprosy and the possibility of intrauterine infection—Authors' Summary

Pattyn, S. R., Eyckmans, L. and Gigase, P. Late reversal reaction after combined treatment of a patient with multibacillary leprosy. Ann. Soc. Belg. Med. Trop. **64** (1984) 291–294.

A patient with a histoid form of BL leprosy was treated with the combination rifampin-prothionamide-dapsone. He developed hepatitis at day 130 and later on received rifampin-dapsone for another 50 days after which dapsone 50 mg daily was administered. Four years after the start of treatment when all symptoms and signs had receded, new lesions appeared. These were first thought to be due to relapsing disease. They were, however, due to a reversal reaction.

The rational interpretation of such cases and the necessary examinations to be performed to elucidate the difference between relapse and reversal reactions are explained.—Authors' Summary

Roy, R. G., Kar, H. K. and Murthy, N. N. Gynaecomastia in leprosy in three districts of Tamil Nadu. Indian J. Lepr. **56** (1984) 578–586.

The prevalence of gynecomastia (GM), a well-known complication of leprosy in adult male patients, was studied in 790 cases of whom 641 were the inmates of five leprosy hospitals and the remaining 149 mostly from the clinics of the field area attached to the Central Leprosy Teaching and Research Institute, Chengalpattu, during 1982–1983. The overall prevalence rate was found to be 19.24%. Among the hospitalized patients, it was 22.15% against 6.71% among the patients attending the clinics in the field area. The youngest and the oldest patient in this study were 16 years and 83 years, respectively. The highest rate of 32.89% was in the 36–45 age group. Only 152 GM cases were detected; the rates in the lepromatous, borderline lepromatous, borderline tuberculoid were 29.21%, 9.64% and 3.68%, respectively. Those who gave a history of frequent ENL reactions had a higher rate of GM, i.e., 34.55 as against 21.52 without or with very infrequent ENL reaction.

Early treatment had a remarkable effect in the reduction of GM. Only 14% developed GM when the treatment was started within 2 years after the onset of leprosy as against 46.9% when the same was started after 16 years. In the latter group, the longer duration of the disease could also play a contributory role. Sterility rate was more than double in those with GM, i.e., 34.14% against 14% without it.—Authors' Abstract

Sandhu, H. S., Naik, G., Chandorkar, R. K. and Bose, C. A study of knowledge, attitude and practice of leprosy among doctors of Bhopal, India. Indian J. Lepr. **56** (1984) 633–640.

A study of knowledge, attitude and practice of leprosy among doctors of Bhopal has found that junior doctors had more exposure to leprology compared to all other groups. Medical college doctors had better knowledge of leprology compared to non-medical college doctors. Knowledge and attitude about leprosy among doctors were influenced by qualification, age, cultural and environmental factors. A strong association was observed between knowledge and attitude about leprosy of doctors and their practice of treating leprosy cases.—Authors' Abstract

Immuno-Pathology

Akiyama, T. Molecular biology of host-parasite relationship in infectious diseases, especially in salmonellosis, tuberculosis, and leprosy. In: *Microbiology—1984.* Leive, L. and Schlessinger, D., eds. Washington, D.C.: American Society for Microbiology, 1984, pp. 355–362.

Labeled DNA from *in vivo*-grown *Mycobacterium lepraemurium* Hawaiian, which was harvested from the livers of infected CBA/J mice and then purified by the method of Prabhakaran, *et al.*, was tested for its homology to cold DNAs extracted from the livers of two strains of uninfected mice with

distinct susceptibility to murine leprosy. The homology between the microbial DNA and the DNA from CBA/J mice, which are highly susceptible to infection, was markedly greater than that between the same *M. lepraemurium* DNA and DNA from C57BL/6N mice, which are relatively resistant to murine leprosy. Similar results were obtained when mutant bacilli carried through 81 serial cultures on Ogawa egg yolk medium were used as a source of labeled DNA. From these data it may be suggested that the susceptibility of animals to infection due to intracellular parasites is predetermined genetically by the degree of relatedness between the DNAs of microbe and host.—(*From* the Article)

Bloom, B. R., Mehra, V., Grosskinsky, C. and Brosnan, C. Immunology of leprosy. In: *Progress in Immunology V.* Tokyo: Academic Press, 1983, 1279–1293.

The increasing availability of specific monoclonal antibodies should make it possible to identify new *Mycobacterium leprae*-specific antigens which, along with the unique glycolipid, should permit worldwide, accurate, and inexpensive epidemiological testing for: 1) infection by *M. leprae,* 2) studying the mode of transmission, 3) characterizing the latent period, and 4) identifying individuals at high risk for developing leprosy, hopefully predicting the form of disease to which they are prone. In addition, with such reagents it should be possible to engineer clones of *Escherichia coli* or other microbial hosts expressing *M. leprae*-specific epitopes, which could then be used as antigens for standardized epidemiologic testing.

It is possible that some of these specific epitopes may be important for developing protective immunity. Because of the limitations on vaccine production in the armadillo, one hopes that it may be possible to produce effective vaccines by recombinant DNA technology in the future. The difficulty, however, is that polypeptide antigens alone are unlikely to be as effective as mycobacteria in inducing cell-mediated immunity, as they would lack the extraordinary adjuvant activity possessed by the mycobacteria. Further development of effective adjuvants for inducing cell-mediated immunity in man is urgently required. Finally, it is not totally unrealistic to conceive of the transfer of genetic information for leprosy-specific protective antigens into cultivable, nonpathogenic mycobacteria, such as BCG vaccine strains, that could provide an ideal vaccine, i.e., one that contains specific protective antigens, lacks tolerogenic determinants, and possesses a potent adjuvant for cell-mediated immunity. Were such a vaccine strain to be developed and engineered with genes for protective antigens against other infectious agents, it could have enormous usefulness for immunizing against many diseases for which cell-mediated immunity is critical to resistance.—(*From* the Article)

Buchanan, T. M. Prospects for early diagnosis, immunotherapy, and vaccination for leprosy. In: *Microbiology—1984.* Leive, L. and Schlessinger, D., eds. Washington, D.C.: American Society for Microbiology, 1984, pp. 314–315.

Effective strategies for early diagnosis, immunotherapy, or vaccination to prevent leprosy will rationally be based upon a greater knowledge of the immunochemistry of *Mycobacterium leprae.* Specifically, early diagnosis will be facilitated by an understanding of the antigens or epitopes of the leprosy bacillus that are found only in *M. leprae,* which can be used as a basis for immunoassays to detect these antigens or antibodies to them in patients or their contacts who are incubating leprosy. Immunotherapy and vaccination to prevent leprosy will be facilitated by including within the immunogen the specific antigens of *M. leprae* that stimulate effective cellular immunity to the organisms and by minimizing antigens that promote suppressor cell responses and inhibit an effective cellular immune response. One may hypothesize that these antigens will be specific for the leprosy bacillus since the immune defect seen in leprosy patients is specific for leprosy and does not in general extend to other infectious diseases. This paper reviews what is currently known about the antigens or epitopes that are specific for *M. leprae* and the potential uses of these antigens for early diagnosis of leprosy, immunotherapy, or vaccination to prevent the disease.—(*From* the Article)

de Chastellier, C. and Lang, T. Exchange of material between the extracellular medium and macrophage phagosomes containing different species of bacteria including mycobacteria. Acta Leprol. (Genève) **2** (1984) 249–257.

Pathogenic mycobacteria survive and multiply once they have infected macrophages. The aim of the present work was to determine whether the persistence of pathogenic bacteria such as *Mycobacterium avium* inside the host cell phagosomes had any effect on the exchanges that normally occur between the extracellular medium and the macrophage vacuolar compartment.

Our results indicate that fusions between phagosomes and lysosomes or/and incoming pinosomes appear to be slowed down by the presence of mycobacteria and even partially inhibited when phagosomes contain viable pathogenic mycobacteria.—Authors' Summary

Duncan, M. E., Fox, H., Harkness, R. A. and Rees, R. J. W. The placenta in leprosy. Placenta **5** (1984) 189–198.

Eighty-one placentae from women with leprosy and 17 placentae from healthy controls were subjected to a detailed macroscopic, light microscopic, ultrastructural, immunopathological, microbiological and biochemical study. The placental morphology and immunohistology were normal, and there was no morphological evidence of infection of the placenta due to *Mycobacterium leprae.* No acid-fast bacilli or acid-fast bacillary granules were seen on light microscopy of any of the placentae from leprous women, although homogenates from two out of seven placentae from women with very active lepromatous leprosy contained acid-fast bacilli in very small numbers. The small placental size of women with leprosy, most marked in those with lepromatous leprosy, appears to be due to a decrease in placental cell size, rather than to a reduced number of cells in the placenta. It is postulated that the small placenta and reduced fetal birth weight observed in lepromatous leprosy are a consequence of depressed maternal immune reactivity.—Authors' Summary

Dyachina, M. N., Sukhenko, L. T., Yushchenko, A. A. and Ermolin, G. A. [Diagnostic test systems on the basis of ELISA in leprosy.] Zh. Mikrobiol. Epidemiol. Immunobiol. **1** (1985) 64–69. (in Russian)

Diagnostic test systems for the detection of IgG and IgM to *Mycobacterium leprae* in the blood sera of leprosy patients and armadillos experimentally infected with *M. leprae* have been developed on the basis of the indirect immunoperoxidase assay. The possibility has been shown of prognosing the activity of the leprotic process in leprosy patients and the results of the experimental infection of armadillos by the dynamic increase of antibody reactions with the development of the infection.—Authors' English Abstract

González-Abreu, E., González-Segregdo, A. and de la Cruz, F. Anti-*Mycobacterium leprae* antibodies induced by lepromin injection as demonstrated by indirect immunofluorescence. Lepr. Rev. **55** (1984) 337–340.

In Cuba the Mitsuda test is carried out on all household contacts of leprosy patients as a measure of epidemiological control. Hence, we need to know whether positivity in the FLA-ABS test can be caused by lepromin testing. The present study shows that sera from healthy individuals were positive by the FLA-ABS test up to 180 days after lepromin testing. The highest positivity rate was reached by day 21, and the highest antibody level by day 45.—Authors' Summary

González-Abreu Castells, E., de la Cruz Castillo, F. and González-Segredo, A. Immunofluorescencia indirecta para la detección de anticuerpos anti-*M. leprae.* Aplicación a un grupo de pacientes y convivientes. [Indirect immunofluorescence for detecting anti-*M. leprae* antibodies. Its application to a group of patients and people living together.] Rev. Cub. Med. Trop. **35** (1983) 251–256. (in Spanish)

A study of the indirect immunofluorescence technique for detecting anti-*Mycobacterium leprae* antibodies, described by M. Abe, in order to evaluate its usefulness

in our environment is performed. Results obtained were comparable to those from Abe and coworkers, which demonstrates possible replication of the method, as well as its usefulness for seroepidemiologic studies.—Authors' English Summary

Jones, R. L. and McDougall, A. C. Three histologic fixatives for the demonstration of *Mycobacterium leprae.* Am. J. Dermatopathol. **6** (1984) 379–380.

We undertook to fix sections of biopsies from four patients known to have lepromatous leprosy in either 10% Formal-saline, 10% Formal-calcium, and "F.M.A.," which consists of 10 parts formalin, 2 parts mercuric chloride, 3 parts acetic acid, and 90 parts distilled water, and is our routine fixative for leprous tissue. The specimens were subsequently cut and stained in parallel and the results compared. All three fixatives appear to allow equally bright staining of acid-fast bacilli in wax-embedded material, and the slides showed no evidence of fading even after 12 months' storage. We conclude, therefore, that fixatives containing calcium chloride do not have a deleterious effect on the acid-fast staining properties of mycobacteria by the Fite–Faraco technique.—(*From* the Article)

Kaplan, G. and Cohn, Z. Hansen's disease as a research model. Bull. N.Y. Acad. Med. **60** (1984) 712–721.

These observations suggest that the severity of the disease is related directly to the extent of macrophage activation. An important factor which complicates the disease is the chemical complexity of the *Mycobacterium leprae* bacteria. Even after they are killed, the bacteria are digested only very slowly and a storage-like disease ensues. It is important to establish the nature of the vacuolar content in the bacteria-laden phagosomes of lepromatous infiltrate cells. That treatment does not cure the disease and a viable bacterial reservoir remains, adds to the complexity of leprosy. Determining where and how the viable bacteria survive awaits the discovery of a rapid method to identify viable as compared to dead bacteria.

It would be of interest to establish the exact phenotype of the unusual lymphocytes found in the tuberculoid lesions and to identify the cause of the micronecrosis in these lesions.—Authors' Conclusions

Khandke, L., Salgame, P. R. and Mahadevan, P. R. *In vitro* lymphocyte stimulating ability of cell components of *Mycobacterium leprae.* IRCS Med. Sci. **12** (1984) 1012–1013.

Mononuclear cells from lepromatous patients, that do not respond to *Mycobacterium leprae,* sonicate of *M. leprae,* cytoplasm, cell wall or lipid fractions of *M. leprae,* do respond strongly to the delipidified cell wall. Thus lepromatous patients are not totally anergic to all the antigens of *M. leprae.* Delipidification could be exposing certain *M. leprae* antigens which are recognized by the lepromatous macrophage and presented in an immunogenic form. An alternative hypothesis is that lipids themselves are immunosuppressive and their removal allows cellular proliferation to occur. In all probability the lipids of *M. leprae* mask the antigenic components and thus the cellular response is not elicited. Whatever the actual mechanism, the present study demonstrates that some components of the organism can elicit an effective cellular proliferation in the defective cells of lepromatous patients.—(*From* the Article)

Kingston, A. E. and Colston, M. J. Concentration-dependent effects of mycobacteria on the stimulation of murine T-cell clones. Acta Leprol. (Genève) **2** (1984) 369–377.

Mycobacterium leprae produced concentration-dependent bimodal effects in cultures of *M. leprae*-immune lymphocytes. At low to intermediate concentrations, *M. leprae* and other species of mycobacteria stimulated lymphoproliferation of *M. Leprae* T-helper cell clones, whereas at high concentrations responses were reduced. Lymphokine production by *M. leprae*-immune T-cell hybridomas also showed bimodal responses to different concentrations of *M. leprae.* These results indicate that mycobacterial antigen may directly induce tolerance of responding lymphocytes.—Authors' Summary

Kolk, A. H. J., Minh, H. L., Klatser, P. R., Eggelte, T. A., Kuijper, S., De Jonge, S. and Van Leeuwen, J. Production and characterization of monoclonal anltibodies to *Mycobacterium tuberculosis, M. bovis* (BCG) and *M. leprae.* Clin. Exp. Immunol. **58** (1984) 511–521.

Thirty-two monoclonal antibodies (MoAb) to *Mycobacterium tuberculosis* H37 Rv, *M. bovis* BCG and *M. leprae* were produced. The spleen cells of BALB/c mice immunized with sonicated or intact bacilli were fused with Sp2/0-Ag-14 myeloma cells. Many more antibody producing hybridomas were found when *M. tuberculosis,* rather than *M. leprae,* was used as the immunogen. The MoAb were characterized by an enzyme immunoassay and immunofluorescence on 16 mycobacterial species. The sodium dodecylsulphate polyacrylamide gel electrophoresis immunoperoxidase assay was used to determine the molecular weight of the antigens detected by the MoAb. Antigens of high, low, and intermediate molecular weight were found. Some of the antigens were proteinaceous; others, of a glycolipid nature. The immunofluorescence assay proved to be essential for the selection of MoAb since some MoAb reacted only in this assay and not in the enzyme immunoassay. The most specific clones were found in the fusions with spleen cells of mice immunized with intact rather than sonicated bacteria. One MoAb (F29-29) reacted only with *M. tuberculosis* H37Rv; one (F41-3) only with *M. leprae* and another (F29-45) reacted with *M. tuberculosis* and *M. gastrii.* Several MoAb only reacted with three mycobacterial species: *M. tuberculosis, M. kansasii* and *M. gastrii.* Others showed unique patterns of reactivity by enzyme immuno- and immunofluorescence assay. The potential use of the MoAb for the identification of mycobacteria and mycobacterial antigens is discussed.—Authors' Summary

Mehta, L. N. and Antia, N. H. Ultrastructure of sciatic nerve of armadillo infected with *Mycobacterium leprae.* Indian J. Lepr. **56** (1984) 540–554.

Ultrastructural observation of sciatic nerves from 8 armadillos were made. Six animals had intravenous inoculation of *Mycobacterium leprae,* one had foot pad inoculation while one had natural leprosy. The available nerves were biopsied at various time sequences, ranging from 5 weeks to 24 months. Semithin sections did not reveal any neuropathy.

Ultrastructurally the perineurium was thick and endoneurial collagen was increased. Initially demyelination of nonmyelinated fibers was seen in all nerves irrespective of the mode of infection. This was followed by demyelination of small myelinated fibers. Active remyelination was predominant after 17 months. Schwann cell activity was increased and various stages of division were seen. Bacilli were extracellular, intra-axonal, in endothelium and in perineurium. Significant observations were on blood vessels. These observations are discussed.—Authors' Abstract

Modlin, R. L., Bakke, A. C., Vaccaro, S. E., Horwitz, D. A., Taylor, C. R. and Rea, T. H. Tissue and blood T-lymphocyte subpopulations in erythema nodosum leprosum. Arch. Dermatol. **121** (1985) 206–209.

To study T lymphocytes in erythema nodosum leprosum (ENL), monoclonal antibodies were used to identify T-lymphocyte subpopulations in the blood and skin lesions of patients with ENL and patients with nonreactional lepromatous leprosy. The blood of nonreactional lepromatous patients had a lymphopenia and a proportionate reduction in pan T cells, helper-inducer, and suppressor-cytotoxic subsets, but a normal helper-suppressor ratio, as compared with controls. Patients with ENL did not differ significantly from the controls. In skin lesions, an admixture of helper and suppressor phenotypes among foamy histiocytes was found. The ENL tissue had more numerous cells of the helper-inducer phenotype and fewer of the suppressor-cytotoxic phenotype, as compared with nonreaction lepromatous tissues. In 22 patients with simultaneous examination of tissue and blood T-cell subsets, there was no correlation between tissue and blood helper-suppressor ratios, indicating that some sort of selection process brings lymphocytes into tissues from peripheral blood.—Authors' Abstract

Modlin, R. L., Rowden, G., Taylor, C. R. and Rea, T. H. Comparison of S-100 and OKT6 antisera in human skin. J. Invest. Dermatol. **83** (1984) 206–209.

The monoclonal antibody OKT6 and antisera against S-100 protein have both been advocated as immunologic markers of Langerhans' cells in the skin. S-100 antiserum has an advantage in its ability to stain Langerhans' cells in paraffin tissues. In order to evaluate whether these antibodies stain equivalent numbers of Langerhans' cells in skin, we compared the staining patterns of S-100 antiserum and OKT6 antibody on biopsy specimens from 40 patients with leprosy using immunoperoxidase techniques. Utilizing OKT6 antibody, greater numbers of positive Langerhans' cells were found in the epidermis in tuberculoid leprosy, reversal reaction, and erythema nodosum leprosum than in lepromatous leprosy. However, these differences were not observed with the S-100 antiserum and, overall, fewer cells were found as compared with the OKT6 antibody. In the dermis both antibodies stained "dendritic cells" that were found encircling granulomas in tuberculoid leprosy and reversal reaction. Staining in lepromatous leprosy granulomas, in contrast to the epidermal staining pattern, revealed rare OKT6+ cells, while S-100 cells were numerous and were more diffusely distributed throughout the granuloma. Our results indicate that antiserum to S-100 protein and OKT6 antibody stain morphologically similar cells (dendritic cells), but do not provide comparable results concerning distribution and frequency of these cells.—Authors' Abstract

Narayanan, R. B., Bhutani, L. K., Sharma, A. K. and Nath, I. Fibronectin in leprosy lesions: Observations using monoclonal antibodies to human fibronectin. Indian J. Lepr. **56** (1984) 532–539.

Croystat sections of dermal lesions from 33 untreated patients with leprosy were studied by indirect immunofluorescence using monoclonal antibodies to human fibronectin. Macrophages in borderline (BL) and lepromatous (LL) leprosy showed intense staining with antifibronectin antibodies. In the tuberculoid lesions cells of the mononuclear phagocyte series in an epithelioid cell granuloma stained for fibronectin. The lymphocytes surrounding these granulomas also showed the presence of fibronectin. These results suggest that the granuloma of leprosy consists of macrophages expressing fibronectin, and the lymphocytes in the lesion also appear to express this protein. In 6 borderline cases, the subepidermal collagen band showed intense staining with antifibronectin antibodies. In addition, the distribution of Ia-like antigens and fibronectin was studied on the plastic adherent cells obtained from peripheral blood of 11 untreated patients with leprosy. Results indicate that higher percentages of adherent cells from lepromatous patients express fibronectin in comparison to adherent cells from tuberculoid patients or controls. However, no difference was observed in the expression of the Ia-like antigens by the adherent cells from these patients.—Authors' Abstract

Nath, I., Jayaraman, J., Sathish, M., Bhutani, L. K. and Sharma, A. K. Inhibition of interleukin-2 production by adherent cell factors from lepromatous leprosy patients. Clin. Exp. Immunol. **58** (1984) 531–538.

Twenty-four hour supernatants (MoF) were obtained from monocyte rich 2 hour adherent cells of 19 leprosy patients and four healthy contacts. MoF from borderline and lepromatous patients produced 52–61% inhibition of human interleukin-2 (IL2) production by a PHA conditioned T cell line (Jurkat). Nonadherent cell supernatants and MoF from tuberculoid and healthy individuals had little effect on IL2 production. The suppression effected by MoF was in the first 12 hours of initiation of PHA stimulated Jurkat cell cultures. Suppressive MoF did not interfere with 1) IL2 release, 2) IL2 utilization by ConA-induced T cell blasts or 3) constitutive proliferation of Jurkat cells. Such MoF were released spontaneously from adherent cells of bacilliferous leprosy patients but required *in vitro* antigen triggering in long term treated lepromatous patients. It is possible that the unresponsiveness associated with lepromatous leprosy is related to the inhibition of IL2 production by suppressive factors, thereby preventing the further expansion of antigen reactive T cells.—Authors' Summary

Nath, I., Sathish, M., Jayaraman, T., Bhutani, L. K. and Sharma, A. K. Evidence for the presence of *M. leprae* reactive T lymphocytes in patients with lepromatous leprosy. Clin. Exp. Immunol. **58** (1984) 522–530.

Evidence for the presence of *Mycobacterium leprae*-reactive T cells in many lepromatous leprosy (LL) patients was obtained using *in vitro* antigen-induced lymphoproliferative responses. 1) Co-cultures of T enriched cells from LL patients when combined with 2 hour adherent cells (AC) from HLA-D compatible tuberculoid leprosy individuals showed significant levels of ^{3}H-thymidine incorporation in the presence of soluble and integral *M. leprae* antigens. 2) More interestingly, autologous T cell + AC co-cultures also showed significant improvement in antigen-induced lymphoproliferation in nine of 16 lepromatous patients. Insignificant improvement was observed in similar co-cultures of tuberculoid leprosy patients. 3) Addition of exogenous, purified human interleukin-2 (IL2) to antigen stimulated PBMC from some lepromatous patients showed the best improvement in terms of overall ^{3}H-thymidine incorporation, indicating that lepromatous patients possess T cells which can differentiate to an IL2 responsive state. Significantly, the level of proliferation varied within the group. A proportion of clinically similar lepromatous patients failed to show improvement by any of the above methods.—Authors' Summary

Padilla, M. A. and Sanchez, J. L. Fluorescence of *Mycobacterium leprae.* Bol. Assoc. Med. PR **76** (1984) 207–209.

The identification and quantitation of acid-fast bacilli in tissue sections is a tedious task for the pathologist since they may be difficult to identify in conventionally stained smears. This is a study comparing the efficacy and sensitivity of a method of fluorescent dyes for the detection of acid-fast bacilli with the conventional Fite–Faraco stain in known cases of leprosy. In general terms, the fluorescent technique was found to be more sensitive than the acid-fast stain particularly in those cases characterized by low bacterial index, such as indeterminate and tuberculoid leprosy.—Authors' Abstract

Rastogi, N. and David, H. L. Phagocytosis of *Mycobacterium leprae* and *M. avium* by armadillo lung fibroblasts and kidney epithelial cells. Acta Leprol. (Genève) **2** (1984) 267–276.

In vitro cell cultures of lung fibroblasts and kidney epithelial cells were established from a freshly killed armadillo and were inoculated with *Mycobacterium leprae.* Lung fibroblasts were also inoculated with *M. avium.* Phagocytosis was allowed for 6 hours at an input of about 50 bacilli/cell, and the ultrastructure was then studied at 24 and 48 hours, and 4, 7, and 10 days. Following observations were made: 1) Armadillo cells could be maintained for nearly 3 months. 2) Both lung fibroblasts and kidney epithelial cells phagocytized *M. leprae* at a high rate. 3) Lung fibroblasts also phagocytized *M. avium* but with a rate much slower than *M. leprae.* 4) Both in *M. leprae* and in *M. avium* an electron-translucent halo surrounded the ingested bacteria within hours of phagocytosis, and this halo appeared to lessen the diffusion of lysosomal enzymes towards the bacterium. 5) *M. leprae* was often phagocytized in clumps of 3–5 bacilli, which separated inside the cell resulting in small individual phagosomes with a single bacterium. 6) In our experimental conditions, there was no evidence of multiplication for both *M. leprae* and *M. avium.* 7) The phagocytized bacilli slowly degraded with longer incubation periods; however, the undigested cell bodies remained inside the phagosomes.—Authors' Summary

Ridel, P. R., Jhol, J. S. and Krahenbuhl, J. L. Cell-mediated immunity in mice treated with *Mycobacterium leprae* or with macrophages harboring *M. leprae.* Ann. Immunol. (Paris) **135D** (1984) 39–50.

Following treatment of BALB/c or C3H/HeN mice in the hind foot pads with irradiated *Mycobacterium leprae,* a marked enhancement of natural killer (NK) activity was observed in cells from the draining popliteal lymph node or from the spleen. NK activity was further enhanced when the treatment consisted of killed *M. leprae* which had been incorporated into mouse peritoneal macrophages. This effect was noted as early as 2 weeks after treatment and persisted for at least 9 weeks. Lymphoblastic

transformation in response to suboptimal doses of the T-cell mitogen concanavalin A or to *M. leprae* antigen was assayed in parallel in cells from the draining popliteal lymph node and from the spleen. In contrast to NK assays, treatment with *M. leprae* alone moderately altered the response to mitogen. However, there was a prominent enhancement of the T-cell response when treatment consisted of *M. leprae*-laden macrophages.—Authors' Summary

Ridley, M. J., Oates, C., Waters, M. F. R. and Ridley, D. S. Lysozyme as a measure of cellular dynamics in the lesions of leprosy. Br. J. Exp. Pathol. **66** (1985) 109–122.

The levels and distribution of lysozyme-positive cells and exudate were studied in leprosy lesions through the spectrum, in untreated and treated patients, in relapse and in reactions. Altogether 124 skin biopsies were examined by the immunoperoxidase technique. Monocytes, neutrophil-polymorphs and mast cells were the most conspicuous cells seen. Lysozyme proved to be a useful means of indexing renewal of these cells in the lesions. Peak numbers of monocytes were seen in lesions of active lepromatous leprosy (LL) and of tuberculoid leprosy (TT), at poles of opposite immunological performance. In TT the stimulus for recruitment was delayed hypersensitivity (DH). A decline in DH from TT towards the middle of the spectrum, mid-borderline, was accompanied by a fall in monocyte level. Furthermore, reacting lesions due to enhanced DH also had increased numbers of monocytes. On the other hand reactions associated with immunological deterioration were similar to active lepromatous leprosy (LL) and monocyte influx was raised in response to the stimulus of free multiplication of bacilli in both cases. In TT delayed hypersensitivity acted also to promote the rapid transformation of monocytes to epithelioid and giant cells, all of which were strongly positive for lysozyme. This was in contrast to much lower levels in histologically similar macrophage-epithelioid cells of BT granulomas. Lysozyme synthesis was not seen in macrophages after ingestion of *Mycobacterium leprae*. Early foamy change was made conspicuous by lysozyme deposited in phagocytic vacuoles, but old foam cells in regressing lepromas were negative. Lysozyme bound to dead extracellular *M. leprae* but not to viable or intracellular organisms. Dead bacilli or immune complexes appeared to be the stimulus for neutrophil-polymorph recruitment, mainly in reactions.—Authors' Summary

Schwerer, B., Meeker, H. C., Sersen, G. and Levis, W. R. IgM antibodies against phenolic glycolipid I from *Mycobacterium leprae* in leprosy sera: Relationship to bacterial index and erythema nodosum leprosum. Acta Leprol. (Genève) **2** (1984) 395–402.

Serum IgM antibodies against *Mycobacterium leprae*-derived phenolic glycolipid I (PG) were determined in 121 leprosy patients, in contacts, and controls by an enzyme-linked immunosorbent assay technique. Anti-PG IgM levels correlated with disease classification, increasing from the tuberculoid towards the lepromatous pole of the disease spectrum. There was a linear correlation between serum IgM PG-antibody levels and the bacillary index (BI), a measure of bacterial load. Elevated anti-PG IgM in bacillary-negative patients was usually indicative of active disease, undetected by BI. We conclude that anti-PG IgM levels are valuable for monitoring the degree of disease activity. Serum anti-PG IgM levels were significantly lower in patients with erythema nodosum leprosum (ENL) as compared to those without ENL, suggesting that IgM PG-antibodies are also involved in the pathogenesis of ENL.—Authors' Summary

Sharp, A. K. and Banerjee, D. K. Macrophage activity in *Mycobacterium leprae* infection. Acta Leprol. (Genève) **2** (1984) 259–266.

The outcome of a *Mycobacterium leprae* infection is likely to depend upon the balance between the invading organism and the host's immune response. Macrophages are known to play a major role in this response and because *M. leprae* is an intracellular parasite, being found commonly in the macrophages of infected hosts, we have attempted to examine the macrophage/*M. leprae* relationship. Our model has been the athymic nude mouse which has been shown

to be susceptible to lepromatous infection but whose macrophages when cultured *in vitro* actually kill phagocytosed *M. leprae.* We have shown that *in vitro* this killing effect is probably mediated, at least to some extent, by macrophage-generated hydrogen peroxide. Further, we have examined macrophages from nude and normal mice at various stages of *M. leprae* infection in terms of their ability to produce hydrogen peroxide and superoxide. It would appear from our results that activation of macrophages to produce these two bactericidal metabolites increases with increasing bacterial load. However, it would seem that T-cell-mediated mechanisms are also required for effective control of infection as the hyperactive macrophages seen in the nude mouse are unable to control *M. leprae* growth in contrast to the limited infection seen in normal mice.—Authors' Summary

Sharp, A. K., Colston, M. J. and Banerjee, D. K. Susceptibility of *Mycobacterium leprae* to bactericidal activity of mouse peritoneal macrophages and to hydrogen peroxide. J. Med. Microbiol. **19** (1985) 77–84.

Macrophages from athymic nude mice were infected *in vitro* with *Mycobacterium leprae* to study the intracellular fate of this organism. Using the proportional bactericidal test, we have shown that the viability of *M. leprae* declines rapidly within these macrophages, although results of clearance experiments demonstrate that live and killed organisms are cleared at comparable rates. We have also shown that *M. leprae* are susceptible to the bactericidal effects of hydrogen peroxide and we suggest that hydrogen peroxide generated by macrophages is responsible for the killing of intracellular *M. leprae.*—Authors' Summary

Yano, I., Tomiyasu, I., Kitabatake, S. and Kaneda, K. Granuloma forming activity of mycolic acid-containing glycolipids in *Nocardia* and related taxa. Acta Leprol. (Genève) **2** (1984) 341–349.

It was concluded that glycolipids containing shorter chain mycolic acids, less acyl numbers or monosaccharide like glucose, can also form granuloma in mice without protein antigen and therefore seem likely to possess adjuvant activity. Furthermore, it gives a useful model to elucidate the mechanism of macrophage activation, other types of immunological responses and infection of acid-fast bacteria.—(*From* the Article)

Microbiology

Asselineau, C., Daffé, M., David, H. L., Laneelle, M. A. and Rastogi, N. Lipids as taxonomic markers for bacteria derived from leprosy infections. Acta Leprol. (Genève) **2** (1984) 121–127.

Lipid analysis allows the specific detection of *Mycobacterium leprae* among various other bacteria isolated from leprosy lesions. In this report mycolates and glycolipid compositions were used for such a discrimination.

Comparative studies of the lipid composition of tissue fragments from different organs of experimentally infected armadillos, and of cultivable strains isolated from these tissues showed that the last ones did not multiply extensively in the tissues of the animals.—Authors' Summary

Athwal, R. S., Deo, S. S. and Imaeda, T. Deoxyribonucleic acid relatedness among *Mycobacterium leprae, Mycobacterium lepraemurium,* and selected bacteria by dot blot and spectrophotometric deoxyribonucleic acid hybridization assays. Int. J. Syst. Bacteriol. **34** (1984) 371–375.

Deoxyribonucleic acid relatedness between *Mycobacterium leprae* or *M. lepraemurium* and other selected bacteria was studied by both dot blot and spectrophotometric deoxyribonucleic acid hybridization assays. The results obtained by the two methods were similar, except for the relatedness values between *M. leprae* and two corynebacterial strains. Among the mycobacterial species examined, acid-fast organisms isolated from armadillos and a man-

gabey monkey with leprosy-like disease showed 100% relatedness with *M. leprae* grown experimentally in armadillos, suggesting their common origin. In this study we demonstrate the usefulness of the dot blot hybridization technique as a screening method for mycobacterial taxonomy.—Authors' Summary

Barksdale, L. and Kim, K.-S. *Proprionibacterium, Corynebacterium, Mycobacterium* and lepra bacilli. Acta Leprol. (Genève) **2** (1984) 153–174.

Evidence is presented which suggests that certain key markers of lepra bacilli reside collectively in *Proprionibacterium acnes, Corynebacterium tuberculostearicum* and *Mycobacterium leprae.* The unrestricted replication of *M. leprae* depends most probably upon the presence of an immune-deficiency-inducing viral agent or possibly on the combined effects of the organisms considered.—Authors' Summary

Benedetti, E. L., Dunia, I., Ludosky, M. A., Nguyen Van Man, Dang Duc Trach, Rastogi, N. and David, H. L. Freeze-etching and freeze-fracture structural features of cell envelopes in mycobacteria and leprosy derived corynebacteria. Acta Leprol. (Genève) **2** (1984) 236–248.

The structural properties of the cell wall and cell membrane of several mycobacteria and of leprosy-derived corynebacteria (LDC) are investigated by freeze etching and freeze fracture. In all cases the freeze fracture split the cell wall in two asymmetric halves. The cell wall fracture faces of the mycobacteria are characterized by a filamentous network which vary with respect to the amount and complexity among microorganisms of the same species and even more of different species.

In LDC the structure organization of the cell wall and cell membrane differs from that of mycobacteria. The most striking difference is the presence on the fracture faces of the LDC cell wall of different classes of particulated entities of yet unknown nature.

In the mycobacteria and LDC the periseptal annuli likely provide a potential frame for cell envelope and cell membrane assembly.—Authors' Summary

Brennan, P. J. New-found glycolipid antigens of mycobacteria. In: *Microbiology—1984.* Leive, L. and Schlessinger, D., eds. Washington, D.C.: American Society for Microbiology, 1984, pp. 366–375.

Mycobacteria are highly distinctive in that they choose glycolipids of rather exotic composition as the means of expressing their outermost immunologically identifiable anatomy. While the chemical unraveling of these has been a productive exercise, their application to meaningful serological assays has been fraught with difficulty due to their inherent lipophilicity. As seen in this present review, ELISA is a choice system for exploring the antigenicity of lipids and holds great promise for the specific diagnosis of mycobacterioses. The challenge now is to probe further the role of these substances in the cellular immunological aberrations which characterized many mycobacterial infections. Their peripheral location and abundance on the bacterial cell wall could augur a role in the onset of suppressor cell activity in leprosy and *Mycobacterium kansasii* infections, as well as in the passive protection of the bacillus within the macrophage environment. These must be the major topics of future research.—Author's Conclusions

Brown, S., Lanéelle, M.-A., Asselineau, J. and Barksdale, L. Description of *Corynebacterium tuberculostearicum* sp. nov., a leprosy-derived corynebacterium. Ann. Microbiol. (Paris) **135B** (1984) 251–267.

Leprosy-derived corynebacteria (LDC) have been extensively studied over the past decade. A composite of their biological properties (cell morphology, staining reactions, cellular inclusions and guanine-plus-cytosine content of their deoxyribonucleic acid; 16 strains studied) and their chemical structures (peptidoglycan type, major cell wall polysaccharide, major glycolipid as well as characteristic mycolic acids) appears to define them as members of the genus *Corynebacterium.* In relation to other corynebacteria found in humans, including "JK corynebacteria," they seem to be distinct. They are here named *Corynebacterium tuberculostearicum* sp. nov. because they produce a 10-methyloctadecanoic (tuberculo-

stearic) acid (8 strains studied). This and some of their other attributes are considered in relation to properties of leprosy bacilli and *Mycobacterium leprae.*—Authors' Summary

Clark-Curtiss, J. E., Jacobs, W. R., Docherty, M. A., Ritchie, L. R. and Curtiss, R., III. Molecular analysis of DNA and construction of genomic libraries of *Mycobacterium leprae.* J. Bacteriol. **161** (1985) 1093–1102.

Molecular analysis of DNA from *Mycobacterium leprae,* "*M. lufu,*" and *M. vaccae* has demonstrated that the G+C (guanine plus cytosine) contents of the DNAs are 56%, 61%, and 65%, respectively, and that the genome sizes are 2.2×10^9, 3.1×10^9, and 3.1×10^9 daltons, respectively. Because of the significant differences in both G+C content and genome size among *M. leprae,* "*M. lufu,*" and *M. vaccae* DNAs, these species are not related, although hybridization experiments under nonstringent conditions, with two separate cloned *M. leprae* DNA inserts as probes, indicate that there are some conserved sequences among the DNAs. The G+C content of *Dasypus novemcinctus* (armadillo, the animal of choice for cultivating *M. leprae*) DNA was determined to be 36%. Genomic libraries potentially representing more than 99.99% of each genome were prepared by cloning into the cosmid vector, pHC79, in *Escherichia coli* K-12. A genomic library representing approximately 95% of the genome of *M. vaccae* was prepared in pBR322. *M. leprae* DNA was subcloned from the pHC79::*M. leprae* library into an expression vector, pYA626. This vector is a 3.8-kilobase derivative of pBR322 in which the promoter region of the *asd* (aspartate semialdehyde dehydrogenase) gene from *Streptococcus mutans* has been inserted in place of the *Eco*R1-to-*Pst*1 fragment of pBR322. Several (44% of those tested) pYA626::*M. leprae* recombinants and one pBR322::*M. vaccae* recombinant synthesized new polypeptides in minicells of *E. coli,* indicating that mycobacterial DNA can be expressed in *E. coli* K-12, although expression is probably dependent upon use of nonmycobacterial promoters recognized by the *E. coli* transcription-translation apparatus.—Authors' Abstract

David, H. L., Lévy-Frébault, V., Dauguet, C. and Grimont, F. DNA from *Mycobacterium leprae.* Acta Leprol. (Genève) **2** (1984) 129–136.

The cell walls of mycobacteria resisting all enzymatic and chemical methods for solubilization, good quality DNA require that they are converted into spheroplasts before extraction. *Mycobacterium leprae* cannot grow in laboratory media, spheroplasts cannot be induced, and therefore the bacteria must be ruptured using physical methods. In these investigations *M. leprae* were disrupted by sonication. The DNA isolated from sonicated *M. smegmatis* and sonicated purified DNA isolated from *M. smegmatis* spheroplasts were used as controls. Additional controls were DNA from *Escherichia coli* K-12 and DNA from armadillo liver. The DNA base compositions determined from melting curves were: armadillo liver, 37.99%; *E. coli* K-12, 51.61%; *M. smegmatis* spheroplasts, 66.48%; purified sonicated DNA from *M. smegmatis,* 65.00%; and *M. leprae,* 66.96%.—Authors' Summary

David, H. L., Sérès-Clavel, S., Clément, F. and Rastogi, N. Further observations on the mycobacteriophage D_{29}—mycobacterial interactions. Acta Leprol. (Genève) **2** (1984) 359–367.

Our investigations showed that the initiation of D_{29} DNA synthesis started in *Mycobacterium tuberculosis* after a 20-minute lag, while it started in *M. smegmatis* 2–4 minutes after infection. These findings were possibly determined by the regulation of enzymes and cofactors supplied to D_{29} by the host bacteria, and may explain partially the delayed growth of these phages in the tubercle bacilli. Further investigations are necessary to clarify the suspected regulatory mechanisms, which may be significant in the understanding of the regulation of cell division in the mycobacteria. But because our investigations did not progress sufficiently we think it is not pertinent to discuss this matter further except to indicate that our data suggested that *M. leprae* contained the enzymes and cofactors necessary for the initiation of D_{29} DNA synthesis.

Secondary to the main purpose of these

investigations, we observed that in *M. tuberculosis* as in *M. smegmatis*, D_{29} infection caused important changes in the fine structure of the bacteria. The most interesting changes were an asymmetric-symmetric transition in cytoplasmic membranes profiles, the deposition of Thiéry staining material in the inner layer of the membranes, and the appearance of multilayered membranes during infection. These findings were of interest because in *M. leprae* Imaeda and Convit showed multilayered membranes, and Silva and Macedo showed symmetric membranes in which the two layers stained in the cytochemical method of Thiéry. According to our own observations, it is possible that those features reputed to be characteristic of *M. leprae* instead may be signs of cell damage.—(*From* the Article)

Draper, P. Host-grown *Mycobacterium leprae*: A credible microorganism. Acta Leprol. (Genève) **2** (1984) 99–112.

In this paper recent advances in knowledge of the following aspects of *Mycobacterium leprae* were reviewed: animal sources, ultrastructure, lipid composition, wall structure, molecular biology, metabolic capabilities and measurement of viability.

It is proposed that isolates from infected tissues are consistent, that is, they share chemical and biochemical features although originating from many parts of the world; are unique, that is, they have properties possessed by no other microorganism; and are biologically credible, meaning that their behavior and properties are consistent with and sufficient for their postulated role as an intracellular pathogen.

It is now clear that host-grown *M. leprae* is phenotypically quite distinct from organisms so far cultivated *in vitro* from infected tissues. The technique of DNA hybridization provides a method, unaffected by phenotypic changes brought about by transfer from a host to a medium, to compare host-grown *M. leprae* with cultivable microorganisms supposed to be genotypically identical.—Author's Summary

Matsuo, Y. Further studies of the use of cycloheximide for cultivating *Mycobacterium lepraemurium* in cell culture. Microbiol. Immunol. **28** (1984) 1093–1098.

The advantage of using cycloheximide for cultivating *Mycobacterium lepraemurium* in cell culture was further demonstrated. Continuous multiplication of the bacillus in successive subcultures was obtained in MFP, HEp-2 and Vero cells maintained in culture medium containing 0.1 μg of cycloheximide per ml. Growth characteristics were comparable to those observed in the cultures of A31 cells previously reported. The procedure was simple and convenient. Comparable results, however, have not been obtained in cultures of other established cell lines, HeLa 229, L, MDCK, and Neuro-2a.—Author's Abstract

Minnikin, D. E., Dobson, G. and Draper, P. Characterization of *Mycobacterium leprae* by lipid analysis. Acta Leprol. (Genève) **2** (1984) 113–120.

The lipid composition of the leprosy bacillus, harvested from experimentally infected nine-banded armadillos, strongly supports its status as a distinct species of the genus *Mycobacterium*. Phthiocerol dimycocerosate waxes and glycosylated phenophthiocerol dimycocerosates are distinct from those characterized from a number of other mycobacteria. The polar lipids of a single isolate lack diacylated forms of phosphatidylinositol di- and pentamannosides, lipids usually found in most mycobacteria. A simple mycolic acid pattern composed of α-mycolates and ketomycolates is characteristic of most preparations of *Mycobacterium leprae*.—Authors' Summary

Papa, F. P., Rauzier, J. Y. and David, H. L. Occurrence of antigen BCG 60 in leprosy derived corynebacteria and other coryneforms. Acta Leprol. (Genève) **2** (1984) 351–358.

Previous studies have shown that anti-BCG 60 monoclonal antibody could recognize major antigen A 7 of *Mycobacterium leprae*. In the present investigation we attempted to search the presence of the same antigen from the strains of leprosy derived corynebacteria (LDC+) which were isolated from leprosy lesions and were supposed to be the cultivable form of the leprosy bacillus. A comparative study was equally performed on seven strains of *Corynebacteria*

which were isolated in France and had no relation with leprosy patients (LDC−). Utilizing the ELISA technique with an anti-BCG 60 monoclonal antibody, it was found that all three LDC+ strains contained an antigenic determinant common to *M. leprae*; however no LDC+ specificity was found as this antigenic determinant was also revealed on two LDC− strains.—Authors' Summary

Picard, B., Frehel, C. and Rastogi, N. Cytochemical characterization of mycobacterial outer surfaces. Acta Leprol. (Genève) **2** (1984) 227–235.

A cytochemical study of mycobacterial outer surfaces was carried out on both pathogenic (*Mycobacterium leprae, M. avium*) and nonpathogenic (*M. aurum*) strains. Different cytochemical markers were used: ruthenium red, concanavalin A, wheat germ agglutinin, colloidal iron and cationized ferritin. The cytochemical staining pattern varied according to the species studied. The relationship between outer surface properties of mycobacteria and their capacity of adhesion to or ingestion by bone marrow macrophages was also considered.—Authors' Summary

Portaels, F., Daffé, M., Lanéelle, M. A. and Asselineau, C. Étude de la composition lipidique de mycobactéries cultivables isolées de foies de tatous infectés par *Mycobacterium leprae.* [Study of the lipidic composition of cultivable mycobacteria isolated from livers of armadillos infected by *Mycobacterium leprae.*] Ann. Microbiol. (Paris) **135A** (1984) 457–465. (in French)

Four bacterial strains isolated from two livers of armadillos experimentally infected with *Mycobacterium leprae* were studied. Lipids obtained after saponification and methylation and complex lipids obtained by solvent extraction were examined. The presence of mycolates showed that the four strains belonged to the genus *Mycobacterium,* but the mycolate patterns, identical for the four strains, were different from those of all strains studied so far. Three of these strains contained phthioceranic acids, which were not found in the fourth one. Only the last strain contained mycosides of the C type, while the three others contained a new type of glycolipid. Their content in mycolates and in glycolipids demonstrated a clear-cut difference between these strains, on the one hand, and *M. leprae* on the other.—Authors' English Summary

Ryter, A., Frehel, C., Rastogi, N. and David, H. L. Macrophage interaction with mycobacteria including *M. leprae.* Acta Leprol. (Genève) **2** (1984) 211—226.

Resistance properties of pathogenic mycobacteria to macrophage bactericidal activity seems to be due mostly to the composition and constitution of their cell walls. In the case of *Mycobacterium tuberculosis,* sulfatides and polyglutamic acid could be implicated in the phenomenon of fusion inhibition between phagosomes and lysosomes. *M. leprae* and *M. lepraemurium,* which do not seem to inhibit fusions are protected by a thick electron-transparent zone (ETZ) that seems to be composed of mycosides. This layer would inhibit lysosomal enzyme diffusion inside phagosomes.

As ETZ does not exist in mycobacteria before their phagocytosis, we have tried to see when and how it is formed inside macrophages. We have compared ETZ formation in *M. leprae* and *M. avium,* which both contain mycosides. These two species were allowed to be phagocytized by mouse bone-marrow derived macrophage and samples were taken for electron microscopy during the first hours of phagocytosis and also during several weeks of incubation. In *M. avium* ETZ appeared within 1–2 hours after phagocytosis. It seems to be formed by a sort of swelling of the thin electron-transparent layer of the bacterial cell wall. This swelling occurs only in regions where the external polysaccharide layer of *M. avium* starts to disappear. After 1–3 hours, this layer was completely absent and all bacteria were enveloped in a thick ETZ.

In *M. leprae,* the ETZ is also formed within 1 hour after ingestion. However, the presence in some bacteria of a very thin, dense layer located at the original place of the outer dense layer of the cell wall does not fit well with the idea of ETZ swelling. In addition, the appearance of a thick, dense layer located between the ETZ and the phagosome membrane is not yet understood.

The ETZ also formed rapidly in macrophages infected with heat-killed cells of *M. avium* or *M. leprae*. This shows that its formation does not require the active participation of the bacterium.

As already proposed, ETZ seems to lessen considerably the diffusion of lysosomal enzymes towards the bacterium in both species. In *M. leprae* it seems especially efficient because despite acid phosphatase activity found in many phagosomes, neither the number of bacteria per macrophage nor their state of degradation changed during 3½ months of macrophage culture. In the case of *M. avium,* which multiplies inside macrophage phagosomes, the number of partially degraded bacteria increased with time as if the degradation rate was slower than the growth rate.—Authors' Summary

Silva, M. T., Macedo, P. M., Portaels, F. and Pattyn, S. R. Correlation viability/morphology in *Mycobacterium leprae.* Acta Leprol. (Genève) **2** (1984) 281–291.

The present study regards the correlation between the percent of viable *Mycobacterium leprae* (as determined by the mouse foot pad technique) and the quantitative ultrastructural analysis of *M. leprae* cells in 6 armadillo's samples and 1 nude mouse foot pad. The quantitative ultrastructural study of 3 LL patients and 1 *M. leprae*-infected nude mouse was correlated to the morphological index. The results show that most *M. leprae* cells with continuous undeformed cell walls, continuous symmetric membranes, ribosomes and fibrillar nucleoids are viable bacilli. Some cells with the above ultrastructural pattern may be dead bacilli that did not yet enter the macrophage-induced degradative process that results in the disposal of the bacteria. Our results also show that degenerating *M. leprae* cells largely predominate in most samples studied. This means that, even in the absence of antileprosy treatment, dead *M. leprae* cells accumulate in the host's tissues. This point has to be taken into account in the calculation of the generation time of *M. leprae in vivo,* the dynamics of the leprosy bacillus in susceptible hosts being influenced by the simultaneous occurrence of growth, death and degradation.

Since known facts in regard to the physiology of bacterial membranes make it difficult to accept the PAS-symmetric membrane of viable *M. leprae* as the membrane of growing bacilli, a search of *M. leprae* cells with asymmetric membranes was undertaken in appropriate samples from nude mice. Several *M. leprae* cells with normal ultrastructure and Thiéry-asymmetric membranes were found in the foot pads of one mouse. Although this observation must be confirmed in another sample, it suggests that *M. leprae* would not be an exception to the general concept that the membranes of all growing Gram-positive bacteria have PAS-positive components located only in the outer layer. The *M. leprae* cells that have normal ultrastructure and symmetric membranes and that are viable would represent some sort of resting cells, that is, living but not growing bacteria.

All of the 11,263 individual bacillary profiles scored in the ultrastructural study included in the present study exhibited the micromorphological characteristics of acid-fast bacteria.—Authors' Summary

Skinsnes, O. K., Chang, P. H. C. and Kuba, B. A. Liberated intracellular pathogen–leprosy model. Acta Leprol. (Genève) **2** (1984) 195–210.

Thirty-three mycobacterial strains, 30 by culture and 3 directly from tissues, isolated from lepromatous leprosy and leprosy infected armadillos, were compared by numerical taxonomy and by antibodies from lepromatous patients. An additional 17 strains of the MAIS complex were similarly compared and all strains were compared by rabbit antibodies induced by tissue bacilli from armadillos from culture HZ-15 and by members of the MAIS complex. The results are discussed in terms of the identification of *Mycobacterium leprae* against a background of prior long-held hypotheses as to the characteristics of this bacillus.—Authors' Summary

Tsukiyama, F., Katoh, M., Nomura, T. and Matsuo, Y. Use of fluorescent staining method for determining the viability of *Mycobacterium lepraemurium.* Hiroshima J. Med. Sci. **34** (1985) 161–163.

The cell suspension of *Mycobacterium lepraemurium* was exposed to heating of

40°C to 70°C for various lengths of time. The percent green-fluorescent cell by the modified fluorescein diacetate (FDA)-ethidium bromide (EB) staining method was calculated and compared with the infectivity to mice. The reduction in the percentage was associated significantly with the loss of the infectivity.—Authors' Abstract

Wieten, G., Boon, J. J., Groothuis, D. G., Portaels, F. and Minnikin, D. E. Rapid detection of mycobacterial contamination in batches of whole cells of purified *Mycobacterium leprae* by pyrolysis gas chromatography. FEMS Microbiol. Lett. **25** (1984) 289–293.

Preparations of *Mycobacterium leprae*, purified from host animal tissues, were screened by pyrolysis high-resolution gas-chromatographic analysis (Py-GC) for the presence of (myco)bacterial contaminants, which produce wax-ester mycolates. Contamination levels as low as 5% of the total number of acid-fast bacilli could be detected, based on the presence or absence of the marker compounds: a series of isomeric alkenes. The on-line pyrolysis capillary GC method provides an easy procedure for rapid quality control of preparations of purified *M. leprae*.—Authors' Summary

Experimental Infections

Aoyagi, T., Wada, T., Kojima, F., Nagai, M., Yamazaki, T., Koseki, Y. and Umezaza, H. Systemic changes in hydrolytic enzyme activities in mice affected with murine leprosy. Biochem. Int. **10** (1985) 105–113.

In order to investigate the behavior of hydrolytic enzymes in chronic infections, the activities of 17 hydrolytic enzymes were tested in limb muscles, heart muscle, spleen, liver, and the kidney of lepromatous mice infected with *Mycobacterium lepraemurium* and their controls. Typical increases in those enzymatic activities were seen in spleen and liver, where pathological changes were the most pronounced, especially at the 11th week after the inoculation of the bacilli. At the 16th week, the enzymatic changes became less remarkable, probably because of the decreased viability of tissues in these organs. The enzymatic changes observed could not be explained as due to bacterial enzymes. These findings are compatible with the notion that the increases in hydrolytic enzyme activities are related to tissue damage caused by murine leprosy.—Authors' Summary

Bach, M. A. and Hoffenbach, A. Antimycobacteria antibodies in *M. lepraemurium* murine infection. Acta Leprol. (Genève) **2** (1984) 403–411.

Two groups of BALB/c mice were inoculated with either 10^7 or 10^5 *Mycobacterium lepraemurium* (MLM) subcutaneously into the left hind foot pad. Mice receiving 10^7 MLM were followed throughout infection for granuloma size, and antibody production against sonicated MLM, whole MLM, and whole *M. triviale*, using a radioimmunoassay. All mice were sacrificed at 37 weeks post-infection and acid-fast bacilli were enumerated in both foot pads and in the spleen.

Noticeable individual variations were observed in the pattern of progression of the granuloma, and in the resistance to the infection, as assessed by measurements of bacilli local growth and dissemination.

Antibody formation against MLM sonicate was detected as early as 6 weeks post-inoculation, a time when granulomas started to develop. Antibody production increased further when the infection progressed, against MLM (sonicate or whole bacilli), as well as against whole *M. triviale*. No correlation could be found between antibody activity and local bacilli growth or bacilli dissemination.

Mice receiving 10^5 MLM s.c. were followed for granuloma size and antibody production against sonicated MLM or other sonicated mycobacteria (pool of 6 different species). Antibody production could be detected against MLM and other mycobacteria as soon as 4 and 8 weeks after infection, respectively, i.e., several weeks prior to the

appearance of granulomas, which occurred at 12 weeks of infection.—Authors' Abstract

Brett, S. J. Macrophage activation in mice infected with *Mycobacterium lepraemurium.* Acta Leprol. (Genève) **2** (1984) 379–386.

Enhanced levels of systemic macrophage activation were observed in mice subcutaneously or intravenously infected with *Mycobacterium lepraemurium,* as assessed by nitroblue tetrazolium reduction, hydrogen peroxide production and tumor cell cytostasis. High levels of systemic macrophage activation did not correlate with the ability to limit bacterial growth as the most susceptible strain (BALB/c) exhibited the highest levels of macrophage activation.—Author's Abstract

Brett, S. J. T-cell responsiveness in *Mycobacterium lepraemurium* infections in a "resistant" (CBA) and a "susceptible" (BALB/c) mouse strain. Cell. Immunol. **89** (1984) 132–143.

Antigen-specific and mitogen-nonspecific T lymphocyte proliferation and lymphokine release (interleukin 2 and macrophage activation factor) were studied in BALB/c and CBA mice infected intravenously with 10^8 *Mycobacterium lepraemurium* (MLM) organisms. The responsiveness of spleen cells from infected animals to ConA and specific MLM antigen declined as the infection progressed. The decreased responsiveness appeared earlier and was more profound in the relatively susceptible BALB/c strain than in the relatively resistant CBA strain. Nylon-wool-purified, T-cell-enriched spleen cells from both strains, however, responded to both *M. lepraemurium* antigen and ConA until the later stages of infection (17 weeks post-infection). The relevance of nonspecific immunodepression mediated by nylon-wool-adherent spleen cells to the progressive nature of this infection is discussed.—Author's Abstract

Gelber, R. H., Humphres, R. C. and Fieldsteel, A. H. A comparative study of four rodent systems to monitor initial therapy of lepromatous leprosy: In search of a more sensitive system to assess bacterial viability. Acta Leprol. (Genève) **2** (1984) 319–325.

The testing of new antileprosy regimens is handicapped by the lack of a highly susceptible animal that can detect small numbers of viable *Mycobacterium leprae* in the presence of large numbers of dead *M. leprae,* as occurs in patient tissue following chemotherapy. Shepard, *et al.* demonstrated previously that the standard 5×10^3 inoculum of *M. leprae* from skin biopsies of previously untreated lepromatous patients taken as little as 3 days after the initiation of rifampin 600 mg daily or a single 1500 mg dose regularly do not multiply in the feet of mice. Similarly, Kohsaka, *et al.* presented the finding that bacilli obtained from skin biopsies of human lepromatous leprosy patients treated for only 2 days with 450 mg rifampin daily did not multiply in nude mice. However, all the therapeutic regimens used thus far, including those utilizing rifampin, are not sterilizing and despite years of treatment patients continue to harbor drug-sensitive, viable bacilli, "persisters." In order to define optimal chemotherapy of lepromatous leprosy, which might safely permit discontinuation of therapy, a more sensitive monitoring system to assess the relative initial killing of *M. leprae* by candidate regimens would be useful.

Fieldsteel has shown that when as few as 5 viable *M. leprae* were injected with 10^7 heat-killed bacilli in foot pads of the neonatally thymectomized Lewis rat (NTLR), multiplication could be demonstrated. Since previous clinical trials utilizing mouse inoculation had been limited to 5×10^3 to 10^4 bacilli inoculated, it was reasoned that the NTLR and presumably the congenitally athymic rat (nude) would be more sensitive than the normal mouse in detecting a small percentage of viable bacilli and hence would serve as a more useful monitor of human chemotherapy trials than the mouse. The present study was initiated to test this hypothesis and compare in clinical trial two putative bactericidal regimens, which include rifampin and dapsone.—(*From* the Article)

Hoffenbach, A., Lagrange, P. H. and Bach, M. A. Influence of dose and route of inoculation and of mouse strain on the pro-

duction of interleukin 2 in mice infected with *Mycobacterium lepraemurium*. Acta Leprol. (Genève) **2** (1984) 413–420.

In order to evaluate the influence of route and dose of inoculation on interleukin 2 (IL2) production, C57BL/6 mice were infected either intravenously (I.V.) or subcutaneously (S.C.) with 10^5 or 10^8 *Mycobacterium lepraemurium*.

The role of genetic factors on the production of IL2 during *M. lepraemurium* infection was investigated in 7 inbred mouse strains (C57BL/6, DBA/2, F1 (C57BL/6 × DBA/2), DBA/1, BALB/c, CBA and A/J) after I.V. infection with 10^7 *M. lepraemurium*. At different times after *M. lepraemurium* inoculation, the number of AFB within the spleens of infected mice was counted and the ability of ConA-activated spleen cells to produce IL2 was studied. In S.C. inoculated C57BL/6 mice the increase in foot pad thickness was measured during the progression of infection.

After 1 month of infection, heavily infected C57BL/6 mice (10^8 bacilli) showed an early and strong deficiency of IL2 production, regardless of the route of inoculation, whereas mice infected with a lower dose (10^5 bacilli) did not. In S.C. infected mice the decrease of IL2 production was observed when the foot pad enlargement reached to the plateau phase. The data obtained from the enumeration of AFB within the spleens of infected mice allowed to rank the infected mouse strains into 2 separate groups according to the pattern of the *Bcg* gene expression. An IL2 deficiency was only observed in C57BL/6, DBA/1, (C57BL/6 × DBA/2)F1 and DBA/2 infected mouse strains. No evident correlation could be shown between splenic IL2 activity upon ConA stimulation and the number of AFB recovered from the spleens of these 7 inbred mouse strains.—Authors' Summary

Izaki, S., Hibino, T., Isozaki, Y., Hsu, P. S., Izaki, M. and Matsuo, O. Plasminogen activator and plasminogen activator inhibitor associated with granulomatous inflammation: A study with murine leprosy. Thromb. Haemost. **52** (1984) 243–249.

Plasminogen activator that is associated with the development of hypersensitivity granulomas (gPA) was partially purified from a saline soluble fraction of murine lepromas elicited in "resistant" mice, C57BL/6N. The gPA was shown to consist of two subspecies (23,000 and 48,000 in molecular weight) with essentially identical enzymologic properties. The gPA was found to be a relatively heat stable weakly alkaline serine proteinase with trypsin-like characteristics in the specificity for synthetic substrates and proteinase inhibitors. It showed a high affinity for H-D-Ile-Pro-Arg-pNA (Km = 1.4×10^{-4} M), H-D-Val-Leu-Lys-pNA (Km = 5.2×10^{-4} M), and L-pyroGlu-Gly-Arg-pNA (Km = 9.3×10^{-4} M). The gPA did not demonstrate antigenic cross reaction with urokinase-type or tissue-type plasminogen activator.

Two distinct enzymatic regulators of the gPA were also demonstrated in the saline soluble fraction of the hypersensitivity granulomas. The gPA and its regulation are assumed to be correlated with macrophage activation in the hypersensitivity granulomas.—Authors' Summary

Job, C. K., Sanchez, R. M. and Hastings, R. C. Manifestation of experimental leprosy in the armadillo. Am. J. Trop. Med. Hyg. **34** (1985) 151–161.

Three experiments, using different routes and doses of infection, were conducted using 42 armadillos. Thirty-six of them developed generalized disease. There is no significant sex or age difference in susceptibility. Route and dose of infection make very little difference in the disease prevalence except that the intravenous administration of a large dose reduces the period of development of generalized disease. It is quite possible that in armadillos the resistance to the disease is partly genetic. Although a majority of the armadillos developed lepromatous disease, borderline leproma is fairly common. In skin nodules large colonies of extracellular bacilli are demonstrated. Bacilli are also demonstrated in liver parenchymal cells.—Authors' Abstract

Levy, L. and Aizer, F. A dual effect of tilorone on multiplication of *Mycobacterium leprae* in mice. Agents Actions **15** (1984) 398–402.

Tilorone, a synthetic inducer of interfer-

on found earlier to inhibit multiplication of *Mycobacterium leprae* in the foot pad of the mouse while it enhanced infections of mice by *M. lepraemurium* and *M. marinum,* has been shown to exert a dual effect on *M. leprae* infection of the mouse. When administered continuously, incorporated into the mouse diet in a concentration of 0.015 g per 100 g diet, the drug was usually immunosuppressive, permitting enhanced multiplication of the organisms. When administered in a threefold larger concentration beginning during the lag phase or early during logarithmic multiplication, tilorone was antimicrobial; however, when administered in the larger concentration beginning after logarithmic multiplication had been well established, the drug was immunosuppressive. The antimicrobial action of tilorone against *M. leprae* appears to be a direct action that is weak and slow in onset. The mechanism of the immunosuppressive action remains to be elucidated.—Authors' Abstract

Løvik, M., Haugen, O. A. and Closs, O. Delayed-type hypersensitivity after immunization with ultrasonicated *Mycobacterium lepraemurium* in C3H and C57BL mice. Scand. J. Immunol. **30** (1984) 227–235.

Subcutaneous immunization with ultrasonicated *Mycobacterium lepraemurium* (MLMSon) in incomplete Freund's adjuvant (IFA) induced long-lasting skin reactivity with the kinetics of a tuberculin-type delayed hypersensitivity (DTH) reaction in both C3H and C57BL mice. The reactivity generally was stronger in C57BL than in C3H mice, and with increasing doses of MLMSon test antigen the local reaction increased more in C57BL than in C3H mice. Pretreatment of C3H mice with cyclophosphamide before immunization caused a shift in the dose-response curve so that the local reaction increased more with increasing doses of test antigen. Histological examination of the reaction elicited by MLMSon in immunized mice revealed a predominantly mononuclear cell infiltrate, and local reactivity could be transferred by immune cells but not by immune serum. The local reaction elicited by MLMSon exerted an adjuvant effect on the induction of DTH to sheep erythrocytes. Thus, MLMSon in IFA given subcutaneously induced stable DTH that conformed to the criteria for tuberculin-type DTH.—Authors' Abstract

Martin, L. N., Gormus, B. J., Wolf, R. H., Gerone, P. J., Meyers, W. M., Walsh, G. P., Binford, C. H., Hadfield, T. L. and Schlagel, C. J. Depression of lymphocyte responses to mitogens in mangabeys with disseminated experimental leprosy. Cell. Immunol. **90** (1985) 115–130.

Mononuclear cells from mangabey monkeys with disseminated experimental leprosy had increasingly severe depression of blastogenic responses to phytohemagglutinin, concanavalin A, and pokeweed mitogen as the disease progressed. Blastogenic responses were not depressed in cells from mangabeys with more localized disease. Blastogenic responses of cells from normal mangabeys appeared to vary with a circannual rhythm. The demonstration of significant negative correlations between the blastogenic responses to mitogens and the percentages of OKT8+ cells suggested that the mangabey OKT8+ subset may contain cells with suppressor function. The depressed responses to mitogens by cells from monkeys with disseminated experimental leprosy were associated with relatively high percentages of OKT8+ cells. Polyclonal immunoglobulin plaque-forming cell responses to pokeweed mitogen were depressed in cells from experimentally infected mangabeys. The results indicated that defects in immune regulation may occur in experimental leprosy in mangabeys, similar in some respects to the defects that have been reported in human leprosy.—Authors' Abstract

Martin, L. N., Gormus, B. J., Wolf, R. H., Walsh, G. P., Meyers, W. M. and Binford, C. H. Experimental leprosy in nonhuman primates. Adv. Vet. Sci. Comp. Med. **28** (1984) 201–236.

The clinical, histopathologic, bacteriologic, and immunologic features of experimental leprosy in mangabeys closely resemble human borderline lepromatous to lepromatous leprosy. So far, successful

transmission has been obtained with a small number of animals, and it is clear that we do not completely understand the reasons why the disease was not successfully transmitted in nonhuman primates long before. This review article was undertaken in order to reassess the earlier attempts at transmission in light of the more recent successful experimental infections. Certain rather simplistic differences in experimental procedures have been noted, but the final answers still lie buried in the complexities of the unique host–parasite interactions that are involved in this disease. We would expect that future studies on the requirements for transmission in nonhuman primates will open new pathways for the understanding of leprosy in humans.—(*From* the Article)

Meyers, W. M., Binford, C. H., Walsh, G. P., Wolf, R. H., Gormus, B. J., Martin, L. N. and Gerone, P. J. Animal models of leprosy. In: *Microbiology—1984.* Leive, L. and Schlessinger, D., eds. Washington, D.C.: American Society for Microbiology, 1984, pp. 307–311.

Disseminated multibacillary leprosy in unaltered subjects has been reported in three species of armadillos, chimpanzees, mangabey monkeys, rhesus monkeys, African green monkeys, nude mice, and nude rats. The chimpanzee has not been shown to be regularly susceptible and thus requires much more study to establish its potential usefulness. Armadillos, by virtue of their accessibility, have great potential for experimentation. Nude mice and nude rats have the advantage of being available to appropriately equipped laboratories, but are relatively short lived. Infections in athymic nude mice are overwhelming and, like those in the armadillo, are much more severe than in most leprosy patients. Moreover, these rodents, although not artificially altered, have an established genetic immunological deficiency, in contrast to the intact immune system of humans susceptible to leprosy. The mangabey, rhesus, and African green monkeys, even though still in an early stage of experimentation, appear to offer the best hope as ideal models of multibacillary leprosy.—Authors' Summary and Conclusions

Palande, D. D. Correction of intrinsic-minus hands associated with reversal of the transverse metacarpal arch. J. Bone Joint Surg. **65-A** (1983) 514–521.

In a hand with paralysis of the intrinsic muscles, the clawing of the fingers is associated with deformity of the distal transverse metacarpal arch. The arch is often flattened in the open hand position and is sometimes reversed. Sixteen hands were operated on with a new tendon transfer procedure aimed at simultaneous correction of the deformities of the arch and of the fingers. Adequate restoration of the arch was attained in all 16 hands, while the deformity and disability were corrected in 85% of the fingers.—Author's Abstract

Ravisse, P., Rastogi, N., David, H. L. and Guelpa-Lauras, C. C. Experimental leprosy in the armadillo and nude mice: Comparative histobacteriology and ultrastructure. Acta Leprol. (Genève) **2** (1984) 327–339.

In the last 14 years, the armadillo has proved an ideal animal model for studying experimental leprosy and mass production of *Mycobacterium leprae.* However, recently a number of groups working with the nude mouse have claimed its ability as a better experimental model so far as leprosy research and production of leprosy bacilli is concerned. We therefore decided to compare experimental *M. leprae* infection of both the armadillo and the nude mouse. We compared the degree of infection as well as the physiological and morphological state of proliferating bacteria by histobacteriological and electron microscopic studies.

Histobacteriological studies of the infected nude mice showed that the highest number of acid-fast bacilli (AFB) was present in the leproma formed at the site of inoculation, the foot pad, but often the morphological index (MI) was low. A bacillary diffusion was observed, however the internal organs did not show extensive lesions and contained few AFB with a still lower MI. Similarly the AFB present in the nodes were few in number, formed smaller globi than those formed in the foot pad, and were essentially non-solid staining bacilli with a lower MI. In the armadillo, on the other hand, lesions were extensive and apparent in the internal organs, a much higher AFB count was found and the MI was higher than in nude mice.

Ultrastructural studies showed that a much higher proportion of *M. leprae* cells inside armadillo tissues existed as intact bacteria than in nude mice. Essentially damaged bacteria were found in the nodes from nude mice. The number of *M. leprae* bacilli in lung, liver, spleen and kidney from all the nude mice was too little to be studied under the electron microscope. Intact bacilli were observed only in mouse foot pads, which however contained two distinct populations of *M. leprae*; the intact and the damaged bacilli, which were arranged together in separate globi.—Authors' Abstract

Sankaran, K., Hoffeld, J. T., Chaparas, S. D. and Oppenheim, J. J. Genetic resistance of mice to persistent infection with *Mycobacterium lepraemurium in vitro*: Association with macrophage bactericidal responsiveness to lymphokines and dissociation from production of hydrogen peroxide by macrophages. J. Immunopharmacol. **6** (1984) 1277–1289.

Peritoneal and splenic adherent macrophages (SAC) from *Mycobacterium lepraemurium* susceptible (C3H/HeJ) and resistant (C57BL/6J) mice were studied for their abilities to generate H_2O_2 *in vitro*. Unexpectedly, SAC from the susceptible C3H/HeJ strain produced more H_2O_2 than those of the resistant C57BL/6J. *In vivo* sensitization with *M. bovis* (BCG), or *C. parvum* increased production of H_2O_2 by SAC from both strains, whereas *in vivo* sensitization with *M. lepraemurium* enhanced H_2O_2 production only in the C3H/HeJ susceptible strains. *In vitro* addition of a crude lymphokine enhanced H_2O_2 production by C3H/HeJ SAC more than by C57BL/6J SAC. *In vitro* addition of *M. lepraemurium* caused an inhibition of H_2O_2 production by SAC from both strains but the inhibition was greater for the resistant C57BL/6J strain. *M. lepraemurium* phagocytosed *in vitro* by untreated peritoneal macrophages of both mouse strains were morphologically altered to the same extent. However, the addition of lymphokine dramatically increased the degree of bacterial lysis in only the C57BL/6J strain. These results, support the view that H_2O_2 plays a limited, if any, role in the protection of the host from *M. lepraemurium* and may even contribute to susceptibility by inhibiting the host's immune response.—Authors' Abstract

Wolf, R. H., Gormus, B. J., Martin, L. N., Baskin, G. B., Walsh, G. P., Meyers, W. M. and Binford, C. H. Experimental leprosy in three species of monkeys. Science **227** (1985) 529–531.

Eleven mangabey monkeys inoculated with *Mycobacterium leprae* developed lepromatous-type leprosy. Nine of the mangabeys were inoculated with *M. leprae* isolated from a mangabey with naturally acquired lepromatous leprosy. Immune function was depressed in some of these animals after dissemination of the disease. Two mangabeys developed lepromatous leprosy after inoculation with human *M. leprae* passaged in an armadillo. Three rhesus and three African green monkeys inoculated with mangabey-derived *M. leprae* also developed lepromatous leprosy. Mangabeys may be the first reported nonhuman primate model for the study of leprosy. Rhesus and African green monkeys may also prove to be reproducibly susceptible to the disease.—Authors' Abstract

Epidemiology and Prevention

Alvarez Mesa, M., Perez Batista, C., Baez Muñiz, G., Gutierrez de la Solana Dumas, J. and Rodriguez Garcia, R. Importancia del chequeo de convivientes en la incidencia de lepra de 1977–1980 en nueve áreas de salud de Ciudad de la Habana. [Importance of checking up individuals living together for leprosy incidence during 1977–1980 at nine health areas of Havana City.] Rev. Cub. Med. Trop. **35** (1983) 327–337. (in Spanish)

The prevalence of leprosy corresponding to nine health areas of Havana City was studied. From the study of 139 patients, 25 families with more than 1 patient per family

were detected and related to this disease incidence in those health areas during 1977–1980. Based on periodical checking up of first and second degree relatives living together, we succeeded in ascertaining the infection source in a high percentage of cases.—(*From* Authors' English Summary)

Bahmer, F. A. Gegenwärtiger Stand der Lepra in der Bundesrepublik Deutschland. [Current status of leprosy in West Germany.] Hautarzt **35** (1984) 402–407. (in German)

Data on the prevalence of leprosy in the Federal Repulic of Germany obtained by questionnaire from numerous departments of dermatology were compared to those from the German Federal Health Office to which leprosy cases have to be reported. Between 1962 and 1980, the Federal Health Office cases numbered 86, whereas 106 were reported to us by questionnaire. Of the latter, 85 patients were male, 21 female. A sharp increase in cases was seen in the late 1970s, mainly due to the growing number of refugees from Southeast Asia and to foreign workers and their family members .from southern Europe. Patients from Africa and Latin America were only infrequently observed. A total of 16 patients were Germans. The lepromatous type of leprosy predominated, followed by the tuberculoid type, and the borderline cases are the least frequent. Although no secondary cases have been reported so far with certainty in this country and the risk of transmission seems small in a highly developed country such as the Federal Republic of Germany, a certain amount of awareness should be maintained in view of the important role early diagnosis plays in the fate of the patient.—Author's English Summary

Basset, A. Aperçu sur l'épidémiologie de la lèpre en France au cours de ces cinquante dernières années. [Remarks on the epidemiology of leprosy in France in the past 50 years.] Bull. Mem. Acad. R. Med. Belg. **139** (1984) 156–164. (in French)

In metropolitan France there are about 1000 leprosy patients. Most of them come from overseas countries. Their origin varied with the evolution of the relations between France and the countries where leprosy is endemic. This period has spanned the therapeutic revolution brought by the sulfones.—(*From* Author's English Summary)

Chakinis, C., Scollard, D., Nelson, K., Smith, T., Ryan, S., Brown, A., Umland, E., Vithayasai, V. and Schauf, V. Antibodies to *Mycobacterium leprae* detected by enzyme linked (ELISA) and radio (RIA) immunoassay: Comparison among families with leprosy patients. Acta Leprol. (Genève) **2** (1984) 387–394.

Serodiagnosis of leprosy and other mycobacterial diseases has been complicated by the extensive crossreactions which occur between mycobacterial species. Use of a unique *Mycobacterium leprae* antigen, PG-1 offers enhanced specificity in serodiagnosis of *M. leprae* infection. In this field survey of families with treated leprosy patients, the sensitivity of detecting antibody to PG-1 by RIA was similar to that reported for treated patients by others in an IgM ELISA. However, combining results of RIA for PG-1 antibody with a sensitive ELISA for IgG antibodies to *M. leprae* proteins improved the reliability of diagnosing leprosy in these families. To determine the general applicability of our preliminary results with these assays, further studies are needed in well-characterized, untreated leprosy patients.—(*From* the Article)

Dave, D. S. and Agrawal, S. K. Prevalence of leprosy in children of leprosy parents. Indian J. Lepr. **56** (1984) 615–621.

A cross-sectional clinical study was done in slums and in the adjoining village of Raipur town. All of the children in 100 families in which at least one patient of proved leprosy was present were examined. Children of 100 nonleprous families served as controls. In leprous families the prevalence was 14.2 times higher in comparison to children in the control group. Also prevalence was higher in children of those families in which the number of patients was more than one, or there was lepromatous leprosy. In children the common types of lesion were tuberculoid, indeterminate, borderline and pure neural type, in that order, while no case of lepromatous leprosy was seen.—Authors' Abstract

de Lange, G., Wright, P., van Eede, P., van Leeuwen, F., Hoang, L. T. and Nguyen, Hong Thi Diem. Association between leprosy and immunoglobulin allotypes: Gm-A2m and Km frequencies in Vietnamese. J. Immunogenet. **11** (1984) 173–180.

The relationship between immunoglobulin allotypes and leprosy was studied in 91 unrelated patients and 100 healthy controls from Vietnam. Twenty Vietnamese patients with tuberculosis were also typed for the Gm, A2m and Km allotypes. The results were compared with those from the healthy controls. No significant association was found for the allotypes G1m(z.a.x.f) G2m(n), G3m(g.b), A2m(1, 2) and Km(1, 3) between the two groups of patients and the controls. Heterogeneity in the distribution of G2m(n), G3m(b), A2m(2) and Km(3) was found when 60 polar-lepromatous (LL) patients and 27 borderline-tuberculoid (BT) patients were separated out of the 91 leprosy patients. In the LL patients there appeared to be a significantly higher frequency of G2m(n), G3m(b) and A2m(2) in comparison with the BT patients ($p < 0.05$). A significantly lower frequency of Km(3) was found in the LL patients in comparison with the healthy control group ($p < 0.05$). The frequencies of the Gm A2m haplotypes and of the occurrence of the Km(1) and Km(3) in the Vietnamese population were calculated on the basis of the results in the 100 samples of healthy controls. The main haplotype is $Gm^{af,n,b}$ (frequency 0.676), occurring with $A2m^1$ (0.200), as well as with $A2m^2$ (0.476).—Authors' Summary

de Vries, J. L. and Perry, B. H. Leprosy case detection rates by age, sex, and polar type under leprosy control conditions. Am. J. Epidemiol. **121** (1985) 403–413.

The knowledge of leprosy epidemiology is still extremely limited as to basic epidemiologic characteristics. Only the infectious agent and the reservoir of infection have been firmly established. It is all the more surprising that very few studies of analytical leprosy epidemiology are reported in the literature. In order to contribute to the analysis of these characteristics, data are presented on the age and sex distribution of types of leprosy from the Pogiri Leprosy Control Project, a large leprosy control project in Andhra Pradesh, India. This data base includes records on biannual examination of some 160,000 household contacts of nearly 48,000 leprosy cases observed from 5–9 years between 1962 and 1970. These data indicate a peak of leprosy prevalence and incidence in the age group 35–44 years. The sex differential in leprosy, observed in these data, appears more related to sex differences in social contact, as sex ratios of leprosy vary widely among different populations. Finally, the age distribution of tuberculoid leprosy shows a bimodal curve, with peaks at ages 10–14 and 35–44 years. The first peak appears related both to the occurrence of early and self-healing lesions in school children, and to the more frequent examination of school children. Additional observations are presented on type ratios of leprosy in single- and multiple-case households, and on percent of single lesions for tuberculoid cases detected over time.—Authors' Abstract

Ganapati, R., Revankar, C. R., Bandkar, K. R. and Dongre, V. V. Leprosy detection through non-survey techniques. Indian J. Lepr. **56** (1984) 622–625.

The most practical and cheap techniques other than mass surveys to detect leprosy in urban slums are still not known. The population inhabiting a large, somewhat isolated slum in North Bombay was exposed to intensive health education programs over a period of two years. Leprosy cases reporting as a result of these measures as well as those detected by trained workers casually or through contact examination were registered at two weekly clinics conducted within the slum. A total of 184 patients, out of whom 27 were smear positive, were identifiable by these means. The population of the slum was found to be 18,228. Total prevalence rate of leprosy after examining 14,723 subjects in the colony was revealed to be 24 per 1000 (smear positive cases: 2.2 per 1000) out of which a prevalence of 12.4 per 1000 (smear positive cases: 1.8 per 1000) had actually come to our knowledge even before instituting mass surveys. The results of this study indicate that in comparable urban situations it should be possible to identify 54% of total leprosy cases by tech-

niques other than surveys. More significantly, a striking feature of this study is that 82% of cases of true public health significance, namely, smear positive patients, could be found by these means.—Authors' Abstract

Guízar-Vázquez, J., Rostenberg, I., Núñez, C., Peña, F., Fuentes, A., González, V., Vázquez, C., Quintanar, E., Rodríguez, H., Macotela, E. and Armendares, S. Segregación de marcadores genéticos en la descendencia de pacientes con lepra. [Segregation of genetic markers in the descendants of leprosy patients.] Dermatol. Rev. Mex. **28** (1984) 13–33. (in Spanish)

The present paper analyzed: a) The form of segregation from parents with leprosy to their offspring, for the following genetic markers: β-2 glycoprotein-1 (Bg) Group specific component (Gc), Australia antigen, and blood systems; ABO, Rh (CDE/cde), MN and P; b) determination of serum concentrations of IgM and Gc in families with a *propositus* with leprosy, and in "normal families"; and c) correlation analysis between the serum concentration of IgM with serum concentration of Gc. The results suggest: a) A segregation not at random (parents-offspring), for the following antigens: D, E, M, P and βgN. b) The study corroborated the high levels of IgM described in leprosy patients; also, we describe high levels of IgM in the offspring of leprosy patients. c) We do not find any correlation between the sera concentration of IgM with the sera concentration of Gc; however we found that the "normal women" with genotype Gc 2-1, showed the higher levels of IgM.—Authors' English Abstract

Haile, R. W. C., Iselius, L., Fine, P. E. M. and Morton, N. E. Segregation and linkage analyses of 72 leprosy pedigrees. Hum. Hered. **35** (1985) 43–52.

Data on 72 families with multiple cases of leprosy were analyzed for a susceptibility gene linked to the HLA loci. We conducted segregation analysis with the program POINTER and identity of HLA types by descent analysis to determine the most likely mode of inheritance. We then conducted linkage analysis with the program LINKAS, first assuming linkage equilibrium and then allowing for linkage disequilibrium and etiological heterogeneity. Segregation results suggest a recessive mode of inheritance, especially for the tuberculoid forms of leprosy. The linkage results, limited to tuberculoid forms and assuming a recessive model, suggest a hypothesis of loose linkage with no unlinked locus. When an additive model is assumed, the best fit is obtained with a hypothesis of complete linkage ($\theta = 0.0$) with heterogeneity. We currently favor the additive model as the more plausible one.—Authors' Abstract

Jesudasan, K., Bradley, D., Smith, P. G. and Christian, M. Incidence rates of leprosy among household contacts of "primary cases." Indian J. Lepr. **56** (1984) 600–614.

The data consisted of information from 1564 "primary cases" of leprosy of all classifications and 9162 of their household contacts. Household contacts of indeterminate (Ind), borderline (BL) and lepromatous (LL), "primary cases" (PC) had an incidence rate (IR) of 5 per 1000 person years at risk (PYR). Household contacts of tuberculoid (TT) and borderline tuberculoid (BT) patients had an IR of 3.2 and 3.8 per 1000 PYR, respectively. Compared with an incidence rate of leprosy of 1.6 per 1000 PYR among individuals not exposed to leprosy in the same area, household contacts of nonlepromatous patients had a relative risk twice as high, and contacts of lepromatous and borderline lepromatous patients a relative risk of 3 times as high. The incidence rate was higher among household contacts of bacteriologically positive patients, among contacts closely related, and in households with multiple cases. The peak age-specific incidence rate among household contacts was between the ages 5–9 years. The significance of these findings are discussed.—Authors' Abstract

Jesudasan, K., Bradley, D., Smith, P. G. and Christian, M. The effect of intervals between surveys on the estimation of incidence rates of leprosy. Lepr. Rev. **55** (1984) 353–359.

This paper examines the effect that variation in the interval between successive cross-sectional surveys may have on esti-

mates of the incidence rates of leprosy. The results of the present study showed that when surveys of the contacts of leprosy patients were conducted in consecutive years (gap between surveys of one year) the estimated incidence rate of leprosy was 4.7 per 1000 person years of risk. When there was a gap of three years between surveys the estimated incidence rate of leprosy was only 1.9 per 1000 person years of risk. Thus when the between-survey interval increased from one to three years, the estimated incidence rate of leprosy was halved. Similar findings were obtained from the results of prevalence surveys in the general population. The implications of these findings in relation to survey work in leprosy and possible vaccine trials are discussed.—Authors' Summary

Ji, B., Tang, Q., Li, Y., Chen, J., Zhang, J., Dong, L., Wang, C., Ma, J. and Ye, D. The sensitivity and specificity of fluorescent leprosy antibody absorption (FLA-ABS) test for detecting subclinical infection with *Mycobacterium leprae.* Lepr. Rev. **55** (1984) 327–335.

From the examination of 854 sera from different sources by the fluorescent leprosy antibody absorption (FLA-ABS) test, the sensitivity and specificity of this test for leprosy has been confirmed. A positive FLA-ABS test in a non-leprosy individual should be considered as an indicator of subclinical leprosy infection. The subclinical infection rates of two endemic areas ranged from 11.4 to 16.3% and were at least 200 times higher than the cumulative prevalence rate of clinical infection. The combination of a positive FLA-ABS test with a negative Mitsuda reaction indicates that the individual has been infected with *Mycobacterium leprae,* but cell-mediated immunity has not been induced. Such individuals are at a greater risk of developing multibacillary leprosy and should be carefully followed up or some prophylactic measures should be considered. Since subclinical infection cannot be differentiated from very early leprosy by the FLA-ABS test alone, it is not a reliable diagnostic test of early leprosy.—Authors' Summary

Koffi, J. K., Yobouet, P., Danguy, E. and Guessennd, G. La lèpre en Côte d'Ivoire. [Leprosy in the Ivory Coast.] Med. Afr. Noire **30** (1983) 367–371. (in French)

The authors describe the epidemiology of leprosy in Ivory Coast, briefly for the period 1960–1969, in more detail for the next decade. There has been a gradual reduction in incidence from 1.24% in 1960 to <0.1% in 1980. The prevalence (new, old, and inactive cases) fell from a peak of 2.64% in 1963 to 1.35% in 1976, and has since declined only slightly. The great majority (over 95%) of new cases are in adults, and 8% are lepromatous. As a result of leprosy campaigns originating in 1956, an increasing number of patients were put "under observation but without treatment." This population reached a peak in 1975, and has declined since. The results are impressive; the authors warn against complacency, and suggest the hospitalization for several months of new lepromatous patients, detected by mobile antileprosy teams.—G. H. Rée (*From* Trop. Dis. Bull.)

Lombardi, C. Aspectos epidemiológicos da mortalidade entre doentes de hanseniáse no Estado de São Paulo, Brasil (1931–1980). [Epidemiological aspects of mortality among sufferers from Hansen's disease in São Paulo State (1931–1980).] Rev. Saúde Pública **18** (1984) 71–107. (in Portuguese)

The study comprehends the patients (n = 27,260) of Hansen's disease (leprosy) deceased in the State of São Paulo, Brazil, from 1931 to 1980. The author studies the time factor as related to some epidemiological characteristics connected with personal aspects such as patient's age and sex, final clinical form of the disease, place of death, time lag between the occurrence of disease and diagnosis, and time span between the occurrence of disease and death. The time factor as related to the specific mortality and lethality coefficients in Hansen's disease is analyzed by comparison with the proportional mortality data in the State of S. Paulo, with regard to the above mentioned period. Time-related trends in the profile of causes of death among the patients under consideration are also presented. Causes of death are classified according to all the sections of the Manual of the International Statistical Classification of Diseases, Injuries and

Causes of Death (ninth revision—1975) and also according to some of its categories and subcategories classically known as relevant to the pathology of Hansen's disease. In the sub-group containing patients who died from Hansen's disease, the distribution in time of some important personal characteristics such as age, final clinical form of disease, time span between the occurrence of disease and diagnosis, and time span since the beginning of disease until the patient's death are studied. The results found in this sub-group were compared to the ones found in relation to the universe of this study. The results obtained basically show: a) there is a tendency toward the improvement of public health patterns in the group studied, parallel to that observed for the State of S. Paulo as a whole, but this tendency is influenced by specific factors, such as sulfone therapy; b) the existence of two clearly distinct phases in the trends, of the specific mortality curves: i.e., before and after 1950; c) the low socioeconomic status of the group studied and its social stigmatization.—Author's English Summary

Pearson, M. Social factors and leprosy in Lamjung, west central Nepal: Implication for disease control. Ecol. Dis. **1** (1982) 229–236.

Such is the ability of leprosy to generate misconceptions and fears, that many patients are reluctant to be identified. Deformity and paralysis which may occur compound the stigma attached to this rare disease of slow insidious onset. Epidemiological studies of leprosy refer only to known disease and often to highly selected groups of the population. Cohorts are therefore incomplete, and variations in prevalence may reflect social attitudes and data reliability.

This paper describes the demographic and spatial distribution of leprosy in Lamjung, a district of west central Nepal. Variations in known leprosy prevalence between sexes, ethnic groups and areas are related to social and physical factors. An apparent paradox of low leprosy prevalence in an ethnic group with a high proportion of infectious leprosy is associated with adverse social attitudes and poor survey coverage. Although the data are too limited for an epidemiological analysis, variations associated with social and physical factors have crucial implications for disease control.—Author's Abstract

Pfau, R. Leprosy control in Sind. J. Pakistan Med. Assoc. **34** (1984) 255–258.

Eleven figures show the clinical and epidemiological features of leprosy in Sind from 1964 to 1979 and the leprosy services available in Sind and Karachi.—(*From* Trop. Dis. Bull.)

Smith, J. H., Long, E. G., Crouse, D. T., Christie, J. D., Folse, D. S., Imaeda, T. and Barksdale, L. Leprosy in wild armadillos (*Dasypus novemcinctus*) of the Texas Gulf Coast. Acta Leprol. (Genève) **2** (1984) 311–318.

Inocula from human lepromatous lesions into nine-banded armadillos results in localized and systemic proliferation of acid-fast bacilli which are non-culturable and replicate the histologic features of human lepromatous leprosy. The armadillo has become the major experimental model of leprosy in uncompromised hosts and a principal source of the acid-fast bacilli associated with leprosy. Wild armadillos from Louisiana have a histopathologically identical disease produced by non-culturable acid-fast bacilli. Counterculture mores in Texas have resulted in markedly increased armadillo–human contact over the last 15 years. The present study reports the results of a survey of 451 wild armadillos, principally from Texas Gulf Coastal countries, which were screened, studied anatomic pathologically, bacteriologically, electron microscopically and by DNA homology with *Mycobacterium leprae* from human sources.—Authors' Summary

van Eden, W., Gonzalez, N. M., de Vries, R. R. P., Convit, J. and van Rood, J. J. HLA-linked control of predisposition to lepromatous leprosy. J. Infect. Dis. **151** (1985) 9–14.

In a study of the relation between HLA and lepromatous leprosy, HLA haplotype segregation was analyzed in 28 families with multiple cases of different types of leprosy. The inheritance of HLA-DR2, HLA-DR3, and HLA-MT1, which had previously been shown to be associated with susceptibility

to leprosy or with a leprosy type, was analyzed separately. Segregation occurred in a significantly nonrandom fashion in both polar tuberculoid leprosy and lepromatous leprosy. This finding indicated HLA-encoded control of a predisposition to both of these forms of the disease. In both cases the segregation observed among healthy siblings was random. Thus, susceptibility to leprosy per se is probably not controlled by HLA-linked genes. HLA-DR3 was inherited preferentially by children with polar tuberculoid leprosy rather than lepromatous disease ($p = 0.02$), and HLA-MT1 was inherited preferentially by children with lepromatous leprosy ($p = 0.04$). The results confirmed the association of these genetic markers with leprosy type.—Authors' Abstract

Xu, K., *et al.* A preliminary approach on clustering of disease in leprosy families: Application of probability density function model of Poisson distribution. Chin. J. Clin. Dermatol. **13** (1984) 9–11. (in Chinese)

An analogy by a Poisson distribution model was performed with a probability density function of 47,662 leprosy families in Jiangsu province, in order to search clustering in the leprosy families. A significant clustering of cases in multi-case families with at least 2 leprosy patients was found. Besides the opportunity for infection with *Mycobacterium leprae,* individual genetic predisposition and other environmental factors should not be overlooked.—(*From* Authors' English Summary)

Rehabilitation

Brandsma, J. W. and Andersen, J. G. Primary defects of the hand with intrinsic paralysis. Lepr. Rev. **55** (1984) 403–406.

The primary defects of the hand that has suffered intrinsic paralysis are described. The extent to which these defects are corrected by standard tendon transfer operations is discussed.—Authors' Summary

Cottler-Fox, M., Edwardsson, C.-A., Hansen, S., Sedig, K., Engardt, M. and Britton, S. Hand function and leprosy. Ethiop. Med. J. **22** (1984) 161–164.

A study was made of objective and subjective hand function in 114 patients admitted on a random occasion in January 1982 to the All Africa Leprosy and Rehabilitation Training Center in Addis Ababa; 46% suffered from ulnar-median injuries and 44% from ulnar injuries. Other isolated or combined nerve injuries were much less frequent. Subjectively sensory and motor functional loss in combined ulnar-median injuries was most disabling and should accordingly receive most rehabilitative attention.—Authors' Abstract

Shah, A. Correction of ulnar claw hand by a loop of flexor digitorum superficialis motor for lumbrical replacement. J. Hand. Surg. **9-B** (1984) 131–133.

The current dynamic procedures for correction of ulnar claw hand are extensor carpi radialis longus many-tail graft, palmaris longus four finger many-tail graft, and extensor bypass operation. Each of these procedures takes a minimum of one and one-half hours and needs extensive postoperative re-education and training. These procedures employ a graft of plantaris tendon, fascia lata or extensor tendon, with the disadvantages of operating at another site and adhesion of grafts. The procedure described in this article obviates these disadvantages. The operation is simple to perform and has given good results even when performed at leprosy camps where no extensive re-education physiotherapy was available. This operation also tends to correct the reversal of transverse arch. The technique and observations are discussed in detail.—Author's Abstract

Srinivasan, H. Surgical decompression of the ulnar nerve. Indian J. Lepr. **56** (1984) 520–531.

There is sufficient evidence that surgical intervention is by and large beneficial and

can arrest progress of paralysis and permit recovery to a greater or lesser extent. But it requires considerable clinical judgment and technical skill, otherwise we will only be adding surgical insult to the pathological iniury. Timely and adequate surgery for preventing deformities therefore requires involvement and participation of the surgeon and his team right from the planning stage of treatment of early nerve paralysis.—(*From* the Article)

Other Mycobacterial Diseases and Related Entities

Averbakh, M. M., Romanova, R. Y., Insanov, A. B., Abramova, Z. P. and Eremeev, V. V. [Circulating immune complexes in the blood of patients with pulmonary tuberculosis.] Zh. Mikrobiol. Epidemiol. Immunobiol. **12** (1984) 91–94. (in Russian)

The level of circulating immune complexes (CIC) in the sera of patients with fibrocavernous tuberculosis and infiltrative tuberculosis has been found to be correlated with the dissociation and level of mycobacterial antigens contained in CIC. Successful chemotherapy results in the normalization of all the characteristics under study.—Authors' English Abstract

Backman, A., Pirilä, V., Förström, L., Heiskala, M., Numela, T., Tala, E. and Uotila, K. A new method for testing tuberculin skin reactivity—chamber test. Tubercle **65** (1984) 279–283.

The Mantoux test and a chamber tuberculin test applied to the surface of the skin in four concentrations were performed on 229 children and 516 adults. The results were recorded at 72 hours. There was a significant correlation between the two tests. The chamber tuberculin test is technically easy, painless and atraumatic. It gives an opportunity of using a full range of concentrations of tuberculin resulting in a quantitative measurement of sensitivity in one and the same test procedure.—Authors' Summary

Balestrino, E. A., Daniel, T. M., de Latini, M. D. S., Latini, O. A., Ma, Y. and Scocozza, J. B. Serodiagnosis of pulmonary tuberculosis in Argentina by enzyme-linked immunosorbent assay (ELISA) of IgG antibody to *Mycobacterium tuberculosis* antigen 5 and tuberculin purified protein derivative. Bull. WHO **62** (1984) 755–761.

IgG antibody to *Mycobacterium tuberculosis* antigen 5 and tuberculin purified protein derivative (PPD) was measured, by enzyme-linked immunosorbent assay (ELISA), in serum samples from 86 patients with active pulmonary tuberculosis and 91 nontuberculous control subjects from Santa Fé, Argentina. The geometric mean titer for the tuberculosis patients was 74.6 with antigen 5 and 99.5 with PPD. In 91 control subjects the geometric mean titers were 3.6 and 15.6, respectively. Titers were not related to tuberculin reactor status or prior BCG vaccination. At a serum dilution endpoint of 1:40, ELISA with antigen 5 had a sensitivity of 81.4% and a specificity of 93.4% for tuberculosis. At 1:40, ELISA with PPD showed a sensitivity of 82.6% and a specificity of 54.9% for tuberculosis. Applied at a serum dilution of 1:40 to a hypothetical model population with a tuberculosis prevalence of 2%, ELISA using antigen 5 would correctly classify 93.2% of persons and ELISA with PPD, 55.5%. At a dilution of 1:80, accuracy is increased to 99.3% with antigen 5 and 83.3% with PPD, but sensitivity decreases to 64% with antigen 5 and 72.1% with PPD. Thus, antigen 5 is more accurate than PPD for the diagnosis of tuberculosis using ELISA.—Authors' Abstract

Bhargava, D. K., Chawla, T. C., Tandon, B. N., Shriniwas, Kapur, B. M. L. and Tandon, H. D. Cell mediated immune response in intestinal tuberculosis. Indian J. Med. Res. **80** (1984) 264–269.

Cell-mediated immune response was studied in 22 patients of intestinal tuber-

culosis. Delayed hypersensitivity skin test with recall antigen PPD was positive in 45–95% of patients. Skin sensitization with DNCB was carried out in 20 patients and 20 controls. Nine (45%) patients could not be sensitized to DNCB though, all of them were tuberculin (PPD) positive. Among the responders, 9 developed 2+ reaction and 2 showed 3+ reaction. In contrast all the control subjects could be sensitized to DNCB and a majority of them had 4+ reactions. Leukocyte migration was positive in 17 of 22 patients. There was a good correlation between tuberculin response and leukocyte migration inhibition. These findings indicate that there is a subtle defect in cell-mediated response in patients with intestinal tuberculosis. No significant difference was observed in various parameters in patients with ulceroconstrictive and ulcerohypertrophic lesions.—Authors' Abstract

Casal, M. J., Rodriguez, F. C. and Benavente, M. C. *In vitro* susceptibility of *Mycobacterium fortuitum* and *Mycobacterium chelonei* to cefmetazole. Antimicrob. Agents Chemother. **27** (1985) 282–283.

The *in vitro* susceptibility of *Mycobacterium fortuitum* and *M. chelonei* to cefmetazole was studied by the agar dilution method. At a concentration of 16 μg/ml or lower, 44 isolates (96%) of *M. fortuitum* and 8 isolates (40%) of *M. chelonei* were inhibited.—Authors' Abstract

Cross, G. M., Guill, M. A. and Aton, J. K. Cutaneous *Mycobacterium szulgai* infection. Arch. Dermatol. **121** (1985) 247–249.

Multiple inflammatory skin lesions and osteomyelitis of the right ankle developed in a 51-year-old man who had been receiving prednisone therapy for several months. Cultures of both the skin and bone lesions yielded *Mycobacterium szulgai,* a scotochromogenic mycobacterium, which is an unusual human pathogen. The patient's condition responded to treatment with isoniazid, ethambutol hydrochloride, and rifampin.—Authors' Abstract

Frías Iniesta, J. F., Pozo Román, T., González Herrada, C., Hernanz Hermosa, J. M. and Bueno Marco, C. Lupus eritematoso discoide hiperqueratósico tratado con talidomida. [Hyperkeratotic discoid lupus erythematosus treated with thalidomide.] Actas Dermosifiliogr. **75** (1984) 373–379. (in Spanish)

We report a case of discoid lupus erythematosus, in a variant which is specially uncommon and resistant to treatment as the disseminated hyperkeratotic form is, and in which the use of thalidomide produced excellent results. We recommend use of this drug in patients with discoid lupus erythematosus unresponsive to conventional therapy.—Authors' English Summary

Grange, J. M., Kardjito, T., Beck, J. S., Ebeid, O., Köhler, W. and Prokop, O. Haptoglobin: An immunoregulatory role in tuberculosis? Tubercle **66** (1985) 41–47.

A significant correlation was found between levels of haptoglobin in sera from Indonesian patients with tuberculosis and the extent to which these sera suppressed mitogen-driven activation of normal lymphocytes *in vitro.* As in a previous study, we showed that there was an inverse relationship between haptoglobin levels and the circulating lymphocyte count in the same group of patients; it is now suggested that this protein has an immunoregulatory role in tuberculosis. Similar observations have been made by other workers on patients with cancer.

There was no difference in the distribution of the 3 allotypes of haptoglobin in patients and healthy subjects nor was there any association between such allotypes and the lymphocyte count, the dermal reactivity to tuberculin, or the degree of suppression.—Authors' Summary

Heifets, L. B., Iseman, M. D., Cook, J. L., Lindholm-Levy, P. J. and Drupa, I. Determination of *in vitro* susceptibility of *Mycobacterium tuberculosis* to cephalosporins by radiometric and conventional methods. Antimicrob. Agents Chemother. **27** (1985) 11–15.

Among eight cephalosporins and cephamycins tested in preliminary *in vitro* screening against *Mycobacterium tuberculosis,* the most promising for further study was found

to be ceforanide, followed by ceftizoxime, cephapirin, and cefotaxime. Moxalactam, cefoxitin, cefamandole, and cephalothin were found to be not active enough against *M. tuberculosis* to be considered for further *in vitro* studies. The antibacterial activity of various ceforanide concentrations was investigated by three methods: a) the dynamics of radiometric readings (growth index) in 7H12 broth; b) the number of CFU in the same medium; and c) the proportion method on 7H11 agar plates. There was a good correlation among the results obtained with these three methods. The MIC for most strains ranged from 6.0 to 25.0 μg/ml. The BACTEC radiometric method is a reliable, rapid, and convenient method for preliminary screening and determination of the level of antibacterial activity of drugs not commonly used against *M. tuberculosis.*—Authors' Summary

Jenkins, J. S., Powell, R. J., Allen, B. R., Littlewood, S. M., Maurice, P. D. L. and Smith, N. J. Thalidomide in severe orogenital ulceration. Lancet **2** (1984) 1424–1426.

Thalidomide was given to 15 patients with severe orogenital ulceration (OGU). Four patients underwent a double-blind controlled trial with glutethimide as placebo and 11 were treated openly. Treatment with thalidomide produced complete resolution of ulcers in 14 and significant improvement in the remaining patient. No peripheral neuropathies developed. Patients did not respond to glutethimide. Thalidomide is an effective treatment for severe OGU. Adequate contraceptive measures should be taken during treatment.—Authors' Summary

Kubin, M., Holub, M., Mohelská, H. and Schlegerova, D. Experimental infection with *Mycobacterium kansasii* in athymic nude mice. Exp. Pathol. **25** (1984) 233–244.

Subcutaneous infection with *Mycobacterium kansasii* did not cause mortality in athymic nu/nu and control nu/+ mice. It had a stimulatory activity comparable to obligatory mycobacterial pathogens: thymus-dependent lymphatic tissues areas were populated during the infection by lymphocytes also in nu/nu mice in which a low degree of migration inhibition factor production could be seen. In both models the chief site of mycobacterial trapping and phagocytosis was the hyperplastic interdigitating cell mesh in the thymus-dependent areas of lymph nodes. A detailed morphological description of the organ changes is given.—Authors' Summary

Lema, E. and Stanford, J. Skin-test sensitisation by tubercle bacilli and by other mycobacteria in Ethiopian school-children. Tubercle **65** (1984) 285–293.

Quadruple skin-testing with a range of 22 new tuberculins and PPD-RT23 was carried out on 665 school children without BCG scars and 666 with BCG scars, in and around the towns of Butajira and Hosana in Shoa district of Ethiopia. Marked differences in patterns of sensitization were distinguished between the five schools visited. In general, *Mycobacterium chitae, M. diernhoferi, M. kansasii* and *M. vaccae* were common sensitizing agents in all schools, *M. avium* subspecies *brunense, M. gilvum, M. rhodesiae* and *M. xenopi* were absent, and the remaining species investigated were variably present between the schools. Contact with *M. tuberculosis* and *M. leprae* appeared greatest in Hosana and the possibility of sensitization by *M. ulcerans* around the village of Enseno was discovered. The data also provided indirect evidence of the value of BCG in Shoa district. An interesting observation was the very variable enhancing effect that BCG vaccination had on sensitization to individual fast-growing species.—Authors' Summary

Liew, F. Y. Specific suppression of responses to *Leishmania tropica* by a cloned T-cell line. Nature **305** (1983) 630–632.

Spleen cells from BALB/c mice infected with *Leishmania major* 69 days previously (when they displayed a strong suppressor T-cell activity) were grown *in vitro* in the presence of irradiated syngeneic spleen cells, killed promastigotes of *L. major* and 20% T-cell growth factor (TCGF). After 2 months of continuous culture the growing T-cell blasts were cloned by limiting dilution. One vigorously growing clone LTC5 was chosen for further study: 10^7 LTC5 cells or the su-

pernatant fluid from the cultures were inoculated into BALB/c mice immediately before immunization with killed *L. tropica* promastigotes and this treatment was found to ablate delayed-type hypersensitivity to *L. major* antigen but did not do so when other antigens were used to immunize and test the mice. LTC5 cells were also found to suppress the *in vitro* growth of other T-cell lines. LTC5 cells inoculated subcutaneously with promastigotes of *L. major* into BALB/c mice enhanced considerably subsequent lesion development. The LTC5 cells possessed the Lyt 1^+2^- $I\text{–}J^-$ phenotype and were TCGF dependent. The cells had no helper function and were not cytotoxic to macrophages.—R. S. Bray (*From* Trop. Dis. Bull.)

Mackett, M., Yilma, T., Rose, J. K. and Moss, B. Vaccinia virus recombinants: Expression of VSV genes and protective immunization of mice and cattle. Science **227** (1985) 433–435.

Vesicular stomatitis virus (VSV) causes a contagious disease of horses, cattle, and pigs. When DNA copies of messenger RNA's for the G or N proteins of VSV were linked to a vaccinia virus promoter and inserted into the vaccinia genome, the recombinants retained infectivity and synthesized VSV polypeptides. After intradermal vaccination with live recombinant virus expressing the G protein, mice produced VSV-neutralizing antibodies and were protected against lethal encephalitis upon intravenous challenge with VSV. In cattle, the degree of protection against intradermalingually injected VSV was correlated with the level of neutralizing antibody produced following vaccination.—Authors' Abstract

Nozawa, R. T., Kato, H., Yokota, T. and Sugi, H. Susceptibility of intra- and extracellular *Mycobacterium avium-intracellulare* to cephem antibiotics. Antimicrob. Agents Chemother. **27** (1985) 132–134.

Intra- and extracellular susceptibility of 35 clinically isolated *Mycobacterium avium-intracellulare* strains to cefotaxime (CTX), ceftizoxime (CZX), and cefoperazone was studied. MICs for 50% of the isolates *in vitro* were 6.25 μg/ml for CTX and CZX and 25 μg/ml for cefoperazone. A strain susceptible to CTX (MIC, 0.78 μg/ml) and CZX (MIC, 1.56 μg/ml) infected human peripheral blood mononuclear cells in the presence of 20% autologous plasma. The mycobacteria replicated exclusively in monocytes under the above culture condition. Concentrations of CZX 1- to 16-fold higher than its *in vitro* MIC had little effect on intracellular replication of the strain. A concentration of CTX 16-fold higher than its *in vitro* MIC was bacteriostatic to the mycobacteria, but CTX of lower concentrations showed no effect on intracellular replication. Thus, ineffectiveness of the cephems on the therapy of *M. avium-intracellulare* infection was suggested.—Authors' Abstract

Smith, D., Reeser, P. and Musa, S. Does infection with environmental mycobacteria suppress the protective response to subsequent vaccination with BCG? Tubercle **66** (1985) 17–23.

Using a guinea pig model of experimental airborne tuberculosis, we were unable to find evidence to support the hypothesis that infection with environmental mycobacteria (*Mycobacterium simiae* or *M. avium-intracellulare*) interferes with the induction of a protective response in animals subsequently vaccinated with BCG.—Authors' Summary

Waldor, M. K., Sriram, S., Hardy, R., Herzenberg, L. A., Herzenberg, L. A., Lanier, L., Lim, M. and Steinman, L. Reversal of experimental allergic encephalomyelitis with monoclonal antibody to a T-cell subset marker. Science **227** (1985) 415–517.

Administration of a monoclonal antibody (GK1.5) that recognizes the L3T4 marker present on helper T cells prevented the development of experimental allergic encephalomyelitis (EAE) in mice. Furthermore, treatment with GK1.5 reversed EAE when the antibody was given to paralyzed animals. *In vivo* injection of GK1.5 selectively reduced the number of $L3T4^+$ cells in the spleen and the lymph nodes. These results suggest that manipulation of the human equivalent of the murine $L3T4^+$ T-cell subset with monoclonal antibodies may provide effective therapy for certain autoimmune diseases.—Authors' Abstract