CURRENT LITERATURE

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

Chemotherapy

Bhate, R. D., Gupta, C. M., Chattopadhyay, S. P. and Singh, I. P. Experience with multidrug therapy in paucibacillary leprosy. Indian J. Lepr. **58** (1986) 244–250.

Eighty paucibacillary leprosy cases were randomly put on two different multidrug regimens for 6 months followed by dapsone monotherapy. Regimen I was according to WHO (1982) recommendations consisting of dapsone and six once-a-month rifampin. In regimen II in addition to above two constituents, clofazimine was added 100 mg on alternate days. Dapsone thereafter was continued in both the regimens up to 1 year. The efficacy, acceptability and side effects of multidrug regimens were observed for a period of 1 year. Histopathological assessment was done on completion of multidrug therapy in all cases. A comparative evaluation of the effect of two multidrug regimens in paucibacillary leprosy patients is reported. Addition of clofazimine over WHO (1982) recommended regimen appears to have no added benefit. The duration of WHO (1982) recommended regimens was found to be inadequate in many cases.—Authors' Abstract

Dhople, A. M. and Green, K. J. An *in vitro* system using adenosine triphosphate and ^{3}H-thymidine to determine drug sensitivity of *Mycobacterium leprae.* IRCS Med. Sci. **14** (1986) 807–808.

We recently reported on the feasibility of using intracellular adenosine triphosphate (ATP) and ^{3}H-thymidine uptake as indicators of metabolic status and viability of *Mycobacterium leprae.* The present studies were, therefore, undertaken to evaluate drug sensitivity of *M. leprae in vitro* using these two indicators.

No significant inhibitory effect of dapsone on *M. leprae* was observed when the dapsone concentration in cultures was 10 ng/ml and less. When the dapsone level in the medium was 15 ng/ml, there was some inhibition—loss of ATP was 59% at 4 weeks compared to 28% in control cultures; loss of ATP was even higher at 8 weeks (74%), while in control cultures there was a gain in ATP levels over original. Similar results were also observed in the case of ^{3}H-thymidine uptake assays. From past experience, it was postulated that when ATP levels drop to such low levels, there is no possibility for these *M. leprae* to recover. At the end of 8 weeks, *M. leprae* from these cultures (with 15 ng/ml dapsone) were harvested and inoculated into foot pads of mice by standard procedure; 12 months later no multiplication of *M. leprae* in mouse foot pads was observed, thus suggesting that these *M. leprae* had lost their viability when incubated for 8 weeks in the presence of 15 ng/ml dapsone. Under identical conditions, *M. leprae* from control cultures multiplied at normal rates in the foot pads of mice. When the dapsone concentration in cultures was 20 ng/ml and more, a definite inhibition (killing effect) was observed.

Thus, it seems that the method employed here has merits in *in vitro* screening of potential antileprosy compounds, either singly or in combinations. The method further demonstrates the feasibility of using intracellular ATP levels and ^{3}H-thymidine uptake as indicators of metabolic activity of *M. leprae* and also shows the parallelism between metabolic activity and viability of *M. leprae* as determined by the mouse foot pad method.—(*From* the article)

Karp, W. B., Subramanyam, S. B. and Robertson, A. F. Binding of dapsone and its analogues to human serum albumin. J. Pharm. Sci. **74** (1985) 690–691.

The binding of dapsone, 4,4′-sulfonylbis(aniline) (1), and its diacetylated derivative, 4,4‴-sulfonylbis(acetanilide) (2), to human serum albumin is reported. To assess the ability of these compounds to displace 4′-[(4-aminophenyl)sulfonyl]acetanilide (3) from albumin, a dialysis rate technique was used. Competition for the bilirubin binding site on albumin was measured with the peroxidase assay. Compounds 1 and 2 strongly displaced both 3 and bilirubin from human serum albumin. The association constants for 1 and 2 with respect to bilirubin binding were 1.29×10^3 and 1.15×10^4 M^{-1}, respectively. These results suggest that the binding site for 3 and the bilirubin binding site are similar with respect to 1 and 2 and that the binding of dapsone and its derivatives probably does not involve the amino function.—Authors' Abstract

Uplekar, M. W. and Antia, N. H. Dapsone dependent nodular pannicultis. Indian J. Lepr. **58** (1986) 286–290.

A patient recorded to be suffering from tuberculoid leprosy since 1973 and on regular dapsone monotherapy for about 9 years developed asymmetrical, erythematous, subcutaneous, nodular swellings restricted chiefly to the extensor aspects of lower limbs 2 months after discontinuation of dapsone therapy. During the course of dapsone treatment, the patient had developed similar swellings twice previously, each time when he stopped the drug for about a month. The swellings disappeared on commencement of dapsone treatment. This has been reconfirmed under our supervision. The biopsy of one of the lesions revealed panniculitis with vasculitis. The original diagnosis of leprosy was probably invalid.—Authors' Abstract

Clinical Sciences

Bhatia, V. N., Sirumban, P., Padma, M. N. and Harikrishnan, S. Retrospective analysis of smear examinations from multiple sites in multibacillary leprosy. Indian J. Lepr. **58** (1986) 257–262.

A retrospective analysis was done on results of smears from six sites in untreated and treated multibacillary leprosy cases. The examination of three sites was found adequate to detect all multibacillary cases.—Authors' Abstract

Chattopadhyay, S. P. and Gupta, C. M. Immunotherapy in leprosy: a new approach. Indian J. Lepr. **58** (1986) 233–239.

An immunotherapeutic agent prepared from a patient's own affected skin was tried in 30 leprosy cases; 53.6% cases of lepromatous and borderline lepromatous group showed lepromin conversion from lepromin negativity to positivity after 12 weeks of immunotherapy. The clinical and bacteriological improvement was also good; 88.1% cases of borderline tuberculoid also showed fair to good clinical recovery following 12 weeks of immunotherapy.—Authors' Abstract

Dhopavkar, P. V., Lele, V. R., Kher, A. V., Kherdekar, M. and Grover, S. Amyloid goiter—a case report. Indian J. Lepr. **58** (1986) 295–298.

A rare case of amyloid goiter in association with leprosy is reported.—Authors' Abstract

Doehring, E., Reider, F., Dittrich, M. and Coddi, Z. Ultrasonographic findings in the livers of patients with lepromatous leprosy. J. Clin. Ultrasound **14** (1986) 179–183.

Abdominal ultrasonography, including assessment of the liver, spleen, pancreas, great abdominal vessels, and kidneys, was carried out in 7 patients with lepromatous leprosy, 6 patients with tuberculoid leprosy, and 32 healthy Congolese controls. Abnormal ultrasound findings were predominantly detected in the livers of patients with lepromatous leprosy and included an inhomogeneous echo texture of the hepatic parenchyma in all cases. Furthermore, six patients revealed echo-dense, partly irregular areas up to 1.5 cm × 3 cm in size

distributed throughout the liver. These were associated with shadowing and were considered to contain calcium. No abnormal findings were encountered in controls or in patients with tuberculoid leprosy except for one patient with tuberculoid leprosy who had a rounded caudal liver edge. The sizes and volumes of liver, spleen, and kidneys were not different in the three groups.—Authors' Abstract

Duncan, M. E. Leprosy in pregnancy. Postgrad. Doctor—Middle East **9** (1986) 384–392.

The article summarizes the adverse effects of leprosy in pregnancy on birthweight of the baby and on clinical complications in the mother, and gives practical advice on health care of leprosy patients during pregnancy and of their babies.—C. A. Brown (Trop. Dis. Bull.)

George, J., Balakrishnan, S., Bhatia, V. N., Anandan, D. and Harikrishnan, S. Thyroglobulin autoantibodies in leprosy. Indian J. Lepr. **58** (1986) 191–195.

Two hundred five sera from lepromatous leprosy patients were tested for the presence of thyroglobulin autoantibodies using tanned red cell hemagglutination technique. Six out of 182 sera from LL patients and 5 out of 23 sera from LL patients with ENL gave a positive reaction for thyroglobulin autoantibodies.—Authors' Abstract

Gupta, C. M. and Tutakne, M. A. An evaluation of palmar flexion creases and dermatoglyphics in leprosy. Indian J. Lepr. **58** (1986) 263–275.

The palmar flexion creases and dermatoglyphics of 150 male leprosy patients (100 paucibacillary and 50 multibacillary leprosy) were compared with 50 matched controls. Among palmar dermatoglyphics a significantly high frequency ($p < 0.001$) of palmar pattern in thenar/1st interdigital area was noticed on left palm of multibacillary leprosy patients. A slight increase in frequency of distal axial triradii (t′ and t″) was also seen on palms of leprosy patients. No difference in values of atd angle and C-line types were observed between patients and controls. Among palmar flexion creases a significantly high frequency of single radial base crease (SRBC) and lower frequency of double radial base crease (DRBC) was noticed on palms of leprosy patients as compared to controls ($p < 0.001$). The difference mainly exists on the left palm. A significantly high frequency of simian crease was also observed on palms of multibacillary leprosy patients ($p < 0.001$) and paucibacillary leprosy patients ($p < 0.05$) as compared to controls.—Authors' Abstract

Gupta, S. C., Singh, P. A., Bajaj, A. K., Budhraja, B. K. and Tripathi, A. Antispermatozoal antibodies in leprosy with special reference to their morphological patterns. Indian J. Lepr. **58** (1986) 196–201.

Sixty-two male patients with polar leprosy—38 lepromatous and 24 tuberculoid types—were investigated for the presence of antispermatozoal antibodies with special reference to their morphological patterns. Antibodies were detected by three different immunological techniques. Sperm agglutination was found to be the most sensitive. The incidence of antibodies was higher in patients with lepromatous leprosy and was directly proportional to the duration of the disease in both types of leprosy. Morphologically, head-to-head type of agglutination was observed in 50% of the patients, mixed in 41.7% and tail-to-tail type in 8.3%. There was no correlation between the number of ENL attacks and the incidence of antibodies. In polar tuberculoid leprosy patients the histological findings of testicular biopsy indicated cell-mediated tissue damage occurring in a noninfective form.—Authors' Abstract

House, P. H. Ocular leprosy: a case report and discussion of the pathology. Aust. N.Z. J. Ophthalmol. **14** (1986) 59–63.

Although leprosy is considered rare in Australia outside the Northern Aboriginal population, its presence within Indian and Southeast Asian migrant groups must be considered. A case of ocular leprosy is presented in which the definitive diagnosis was delayed because lid changes had been diagnosed as senile ectropion. The pathology of ocular leprosy is discussed, together with

relevant therapeutic considerations.—Author's Abstract

Jain, A. P., Gupta, O. P., Jajoo, U. N. and Kumar, K. Study of autoantibodies in lepromatous leprosy in rural Central India. Indian Med. Gaz. **119** (1985) 236–237.

Autoantibodies, i.e., rheumatoid factor in 40% (20/50), antinuclear factor in 10% (5/50), antithyroid (microsomal and mitochondrial) in 8% (4/50), and antisperm antibodies in 18% (18/50) were present in 30 LL, and 20 ENL patients. None of these had any clinical expression, i.e., arthritis, SLE, thyroid, sterility and gynecomastia, respectively. Variation in clinical picture had no effect on these autoantibodies.—(*From* Excerpta Medica)

Jain, G. L., Pasricha, J. S. and Guha, S. K. An attempt to reduce the loss of pain and touch sensations in leprosy patients. Indian J. Lepr. **58** (1986) 225–232.

An attempt was made to improve the perception of pain and touch sensations at the leprosy lesions. The loss of pain and touch sensations in a lesion was graded using Pain/Touch-Sensation-Testing-and-Grading devices. Application of a solution containing 1 mg of histamine per ml of DMSO at the affected area decreased the grades of the loss of pain sensation in 11 (31.4%) patients and of touch sensation in 8 (22.8%) patients, out of the 35 patients tested, indicating an improvement in the perception at the lesion. This effect, however, did not persist even for 5 minutes. A higher concentration (2 mg/ml) of histamine produced reduction in the sensory loss in a larger percentage (47% for pain and 35.3% for touch) of patients, although the duration of this effect was still not prolonged.—Authors' Abstract

Miller, J. A., Dowd, P. M. and Rode, J. Lepromatous leprosy. J. R. Soc. Med. **78** Suppl. 11 (1985) 46–47.

Subcutaneous nodules are a very uncommon feature in untreated lepromatous leprosy, and we report their occurrence in such a patient.—Authors' Abstract

Ottinger, M. L. and Black, J. R. Hansen's disease with pedal involvement; a case report. J. Am. Podiatric Med. Assoc. **76** (1986) 161–163.

Although Hansen's disease is a major world health problem, its incidence within the United States is very small. Clinically, this disease is seen within a wide range of presentations. It is not uncommon for the disease to be present for some years before the definitive diagnosis is made. In the initial course of Hansen's disease, biopsy may not produce a conclusive diagnosis. Finally, a skin rash, motor neuropathy, sensory neuropathy, and digital ulceration are latent symptoms of this pathologic process.—Authors' Summary

Patil, S. A., Sinha, S., Ramu, G. and Sengupta, U. Studies on serum proteins in leprosy by polyacrylamide gel electrophoresis (PAGE)—I. Indian J. Lepr. **58** (1986) 202–207.

Serum protein pattern was studied in the leprosy spectrum, their contacts, and in normal individuals by employing polyacrylamide gel electrophoresis. Sera from 80% of untreated BL/LL, 70% of untreated TT/BT patients and 67% of contacts have shown dysproteinemia either for 232 kD or for 175 kD or for both these proteins together. A tendency of these proteins to return to normal levels was observed after treatment. But both these proteins come back to normal levels only after subsidence of the disease.—Authors' Abstract

Pavithran, K. Amniotic band syndrome. Indian J. Lepr. **58** (1986) 291–294.

A case of amniotic band syndrome is reported in a 12-year-old boy. He had multiple deformities of the limbs which resembled leprosy. Recently, he developed foot drop due to pressure on the right common peroneal nerve by the constriction ring of the leg.—Author's Abstract

Rohatgi, J., Shorey, P., Lamba, P. A. and Sehgal, V. N. Uveal changes in leprosy. Indian J. Lepr. **58** (1986) 208–215.

A total of 424 leprosy patients were screened for uveal involvement. Uveal changes were found in 11.2% of these patients. Uveal involvement was more com-

mon in patients with greater duration of the disease and in patients on irregular dapsone therapy. Of the patients with uveal involvement 82% were lepromatous, 16% were borderline, and 2% were tuberculoid leprosy. Uveal changes in form of active iridocyclitis (21.3%), healed iridocyclitis (58.5%), iris atrophy (10.6%), iris pearls (7.4%), small irregular pupil (46.7%), and pupil refractory to dilatation (56.3%) were seen. Posterior segment involvement was rare. Lepromatous and borderline leprosy patients with no clinical evidence of uveal involvement had decreased power of accommodation as compared to normal subjects.—Authors' Abstract

Shah, A. Evaluation of nerve function deficit, its improvement by nerve decompression or corticosteroid therapy. Indian J. Lepr. **58** (1986) 216–224.

An attempt has been made to use the protocol for evaluation of nerve damage prepared at a "Workshop on Neuritis" held at Schiefflein Leprosy Research and Training Centre, Karigiri, in 3 cases of ulnar nerve decompression, 3 cases of median nerve decompression and 3 cases of ulnar neuritis on corticosteroid therapy. The nerve function deficit did show an improvement. The problems and suggestions are presented with a view to encourage the use of said protocol to enable comparison of data among different centers and different therapy.—Author's Abstract

Singh, M. Anterior leprotic retinitis of trantas: a case report. Med. J. Malaysia **40** (1985) 139–141.

A case of rare retinal lesion occurring in a young girl suffering from lepromatous leprosy is described. Fundus lesions in leprosy are extremely rare but do occur in some cases without causing any threat to vision. Their response to antileprotic treatment is not clearly known.—Author's Summary

Immuno-Pathology

Antia, N. H. and Mukherjee, R. Nerve tissue culture: a useful model for the study of nerve damage in Hansen's disease. Ind. Star **45** (1985) 8–9.

Attempts have been made to develop animal models to study the mechanisms responsible for the pathogenesis of neuritis in leprosy. Experiments carried out in these models have yielded limited understanding. Techniques have now become available for culturing highly organized nerve tissues or various cell types as pure populations. The Schwann cell, neurons and fibroblasts can now be cultured independently or in various combinations. In these culture models, therefore, the very early events of nerve damage can be visualized and studied.—(*From* Excerpta Medica)

Ashworth, M., Sinha, S., Patil, S. A., Ramu, G. and Sengupta, U. The detection of subclinical leprosy using a monoclonal based radioimmunoassay. Lepr. Rev. **57** (1986) 237–242.

The monoclonal antibody based competition radioimmunoassay test was used to examine sera from 100 healthy household contacts of known leprosy patients. Only 6 out of 100 contacts had detectable specific antibodies. It remains conjectural that this small fraction of contact subjects may be at much higher risk of developing disease than those without antibodies. Contacts who are antibody positive and lepromin negative (as were 4 of the 6), would best qualify for being offered chemoprophylaxis.—Authors' Summary

Engers, H. D., Houba, V., Bennedsen, J., *et al.* Results of a World Health Organization-sponsored workshop to characterize antigens recognized by mycobacterium-specific monoclonal antibodies. Infect. Immun. **51** (1986) 718–720.

An international workshop organized and sponsored by the Immunology of Tuberculosis (IMMTUB) component of the World Health Organization (WHO) Vaccine Development Programme to characterize the specificity and reaction patterns of murine

monoclonal antibodies (Mabs) raised against various mycobacteria was held in Geneva, 3 to 5 June 1985. A total of 31 Mabs (28 ascites and 3 culture supernatants) generated in nine different laboratories using several mycobacterial antigens for immunization (*Mycobacterium tuberculosis,* virulent and avirulent strains, *M. bovis* BCG, and *M. leprae*) were submitted in early 1985 to the IMMTUB Mab bank located in the WHO Immunology Research and Training Center (WHO/I.R.T.C.) in the Department of Pathology, University of Geneva. The samples were coded, aliquoted, and distributed to 12 laboratories for independent analysis by a variety of methods. In addition to the Mabs, several laboratories submitted various mycobacterial antigen preparations for testing. These antigens were derived by three different approaches: preparations eluted from Mab immunoadsorbant columns, an antigen preparation (nonpurified) obtained by recombinant DNA techniques and expressed in *Escherichia coli,* and a synthetic peptide prepared by conventional solid-state peptide synthesis methods. Subject to available reserves, samples of the IMMTUB mycobacterium-specific Mabs and the recombinant DNA library will be made available to qualified investigators by the contributing scientists, upon receipt of a short (one page) description summarizing the experiments to be conducted.—(*From* Excerpta Medica)

Gill, H. K. and Godal, T. Deficiency of cell mediated immunity in leprosy. Prog. Allergy **37** (1986) 377–390

Leprosy manifests itself in a range of clinical forms which have been systematically gathered into a spectrum. At one end of this spectrum is the high-resistant, paucibacillary tuberculoid leprosy while at the other end is the low-resistant, multibacillary lepromatous leprosy. Lepromatous leprosy (LL) is a systemic form of the disease in which the patient does not possess the ability to mount a cell mediated immune response against *Mycobacterium leprae.* This immunological deficiency is specifically related to *M. leprae* antigens. LL individuals exhibit an unimpaired immune response to such closely related antigens as BCG and PPD.

Recent studies have shown that LL lymphocytes fail to produce IL-2 in response to *M. leprae* and that their proliferative response to this antigen may be restored by IL-2. This, and the evidence that LL macrophages function normally, would appear to indicate that the immunological deficiency in lepromatous leprosy is due to a defect in the T-cell population. Data have been presented which show that such a deficiency may be brought about by suppressor T cells which have been induced by antigens specific to *M. leprae.*—Authors' Summary

Gumaräes Proença, N., Zaitz, C., Duarte, I. and Kliemann, T. A. E. [Behavior of the intradermal reaction with lepromin in patients with sarcoidosis.] Med. Cutan. Iber. Lat. Am. **13** (1985) 471–474. (in Portuguese)

The lepromin (Mitsuda) skin test was performed in 14 patients with sarcoidosis and in 40 controls. In the sarcoidosis patients results were negative in 85.8% and positive in 14.2%. In the control group, on the contrary, results were negative in 15.0% and positive in 85.0%. Thus it became evident that patients with sarcoidosis produce an impaired reaction to the characteristic granuloma formation of the Mitsuda test. This seems to be a paradoxical response, since sarcoidosis is an essentially granulomatous disease.—Authors' English Summary

Kaplan, G. and Cohn, Z. A. The immunobiology of leprosy. Int. Rev. Exp. Pathol. **28** (1986) 45–78.

Leprosy, a chronic infectious disease of man, is caused by the obligate intracellular bacterium *Mycobacterium leprae.* Infection with *M. leprae* affects the peripheral nerves and the dermis, causing an accumulation of macrophages and other immune cells at the infected sites. Host resistance to the bacterium determines the extent of local inflammatory reactions and its resulting damage to the affected tissues. In lepromatous disease little if any cellular immunity develops. Bacterial multiplication is uncontrolled and *M. leprae* disseminate throughout most of the dermis. In tuberculoid

disease, marked cellular immunity is observed and bacterial growth and dissemination are controlled.

The depression of cellular immunity in lepromatous patients is not fully understood. Since *M. leprae* cannot be grown *in vitro,* and a suitable animal model has not yet been developed, the study of host immunity to the pathogen is limited primarily to investigations of the cutaneous lesions of patients and to *in vitro* responses of the peripheral blood leukocytes to *M. leprae.* While the blood monocytes of leprosy patients appear to be activated normally by lymphokines, T-cell proliferation and production of lymphokines in response to *M. leprae* are impaired in lepromatous patients. Attempts to restore responsiveness in cells from these patients have been unsuccessful in our hands. The addition of exogenous IL-2 to leukocyte cultures does not appear to restore responsiveness to *M. leprae* in cells from nonresponsive patients. Rather, some enhancement, often not antigen specific, is observed in cells from patients with a pre-existing response. Similarly, depletion of monocytes does not restore responsiveness to *M. leprae* in nonresponder patients, but a nonspecific enhancement of proliferation is observed in monocyte-free cultures from patients who do respond to *M. leprae.* Thus, the defect in lepromatous nonresponder patients does not result from a simple lack of IL-2 production or suppression by monocytes and/or their products. Possibly, there is a low level or lack of *M. leprae*-responsive T cells in the circulation of these patients.

Attempts to overcome the defect in immunity of patients with lepromatous leprosy by immunoprophylaxis and immunotherapy are being investigated. This approach has become of major importance since the development of widespread drug resistance to dapsone as well as to the other chemotherapeutic agents used to control leprosy.—Authors' Summary

Kaplan, G., Witmer, M. D., Nath, I., Steinman, R. M., Laal, S., Prasad, H. K., Sarno, E. N., Elvers, U. and Cohn, Z. A. Influence of delayed immune reactions on human epidermal keratinocytes. Proc. Natl. Acad. Sci. U.S.A. **83** (1986) 3469–3473.

The epidermal changes that occur in human cutaneous immune responses have been investigated in the tuberculin reaction and in the lesions of tuberculoid and lepromatous leprosy and cutaneous leishmaniasis. In each situation, there was a dermal accumulation of monocytes and T cells, and the epidermis exhibited thickening. In the tuberculin response, the thickness of the epidermis sometimes doubled in 48–72 hr, and this was attributed to increases in both size and number of keratinocytes. In addition, the phenotype of the keratinocytes changed from Ia− to Ia+. Similar changes in keratinocyte Ia-antigen expression occurred in the epidermis overlying untreated tuberculoid leprosy and cutaneous leishmaniasis lesions, but not in lepromatous leprosy. We suggest that one or more epidermal growth factors may be generated in the course of a delayed immune reaction in the dermis.—Authors' Abstract

Kumar, S., Moudgil, K. D., Band, A. H., Naraynan, P. R., Gupta, S. K., Sharma, A. K. and Talwar, G. P. A dot enzyme immunoassay for detection of IgM antibodies against phenolic glycolipid-I in sera from leprosy patients. Indian J. Lepr. **58** (1986) 185–190.

A visual dipstick dot enzyme immunoassay (EIA) for the diagnosis of leprosy is described. The assay is based on detection of IgM antibodies against phenolic glycolipid (PGL-I) in sera from leprosy patients. The antigen (PGL-I or synthetic disaccharide of PGL-I) was dotted on a nitrocellulose pad stuck on a plastic strip (dipstick). Sera were used at a dilution of 1:200. Peroxidase coupled mouse anti-human IgM monoclonal antibodies were used as the conjugate. A positive test gave a blue dot against a white background. The test was highly specific for leprosy, and was quite sensitive for detection of bacilliferous (BL/LL) leprosy. The antigen dotted and preblocked dipsticks stored at room temperature up to 4 months of observation period, were usable in the assay.—Authors' Abstract

Modlin, R. L., Mehra, V., Wong, L., Fujimiya, Y., Chang, W.-C., Horwitz, D. A., Bloom, B. R., Rea, T. H. and Pattengale, P. K. Suppressor T lymphocytes from lepromatous leprosy skin lesions. J. Immunol. **137** (1986) 2831–2834.

The immune response in leprosy forms a spectrum with lepromatous leprosy patients exhibiting specific unresponsiveness to antigens of *Mycobacterium leprae*. This unresponsiveness is thought to be related to the prevalence of T8-positive lymphocyte in these lepromatous lesions. To analyze the immunoregulatory function of these T8 cells, we developed simple procedures to extract lymphocytes from skin biopsy specimens of patients with leprosy. These lymphocytes were sorted for T8 and T4 positive cells, and cell lines were established by expansion with interleukin-2 (IL-2) and irradiated feeder cells. All T8 positive lines tested were positive for IL-2 receptors and HLA-DR determinants. These lines were additionally assayed for lepromin-induced suppression of the normal peripheral blood lymphocyte ConA proliferative response. Thirteen of 32 lines from 6 lepromatous patients showed significant suppressor activity; whereas 9 lines from 6 tuberculoid patients and 1 line from normal peripheral blood failed to show suppression ($p < 0.001$). Taken together, the finding of *M. leprae*-triggered suppressor cells within lepromatous skin lesions may in part explain the *M. leprae* unresponsiveness of lepromatous leprosy patients.—Authors' Abstract

Sehgal, V. N., Gautam, R. K., Koranne, R. V. and Beohar, P. C. The histopathology of type I (lepra) and type II (ENL) reactions in leprosy. Indian J. Lepr. **58** (1986) 240–243.

The histopathological features in type I (lepra) reaction comprised a loose and disorganized granuloma in the upper and middermis, dermal edema and variable cellular contents, namely, epithelioid cells, lymphocytes, giant cells, and macrophages. ENL reactions were characterized by predominant involvement of subcutaneous vessels, vasculitis, and polymorphonuclear infiltration in and around the blood vessels.—Authors' Abstract

Microbiology

Dhople, A. M. Influence of prior periodic acid oxidation on the acid-fastness of *Mycobacterium leprae*. IRCS Med. Sci. **13** (1985) 1259–1260.

It has been shown that in a given mycobacterial culture, not all the cells are stained by the routine Ziehl-Neelsen procedure. Since *Mycobacterium leprae* is not yet cultured *in vitro* its enumeration has to be done by microscopic examination of stained smears. *M. marinum* was grown *in vitro*. The bacterial content in a given aliquot was low if counted by microscopic examination compared to that counted by colony counting. But both counts agreed if the bacterial smear was treated with periodic acid prior to staining with the Ziehl-Neelsen technique. It was demonstrated that young cells in the cultures are not stained without prior exposure to periodate. *M. leprae* from previously infected armadillos and mouse foot pads also behaved similarly. The counts were higher if smears were pretreated with periodate. More interestingly, *M. leprae* from patients given antileprosy treatment also showed the same phenomenon. It is suggested that these chromophobic *M. leprae* from treated patients are not stained by the routine Ziehl-Neelsen technique and presumably survive in their metabolically inactive chromophobic forms and reactivate the disease when conditions are ripe for reactivation.—(*From* Excerpta Medica)

Hunter, S. W., Gaylord, H. and Brennan, P. J. Structure and antigenicity of the phosphorylated lipopolysaccharide antigens from the leprosy and tubercle bacilli. J. Biol. Chem. **261** (1986) 12345–12351.

A family of major arabinose- and mannose-containing phosphorylated lipopolysaccharides was isolated from *Mycobacterium leprae* and *M. tuberculosis*. The only antigenic member of the family, lipoarabinomannan (LAM)-B, was purified by anion exchange and gel filtration chromatography in detergent and recovered in large quantities (15 mg/g of bacteria). It yielded a broad diffuse band on polyacrylamide gel electrophoresis but appeared homogeneous by this criterion and gel filtration. Besides arabinose and mannose, it contained glycerol and a polyol phosphate and was acylated by lactate, succinate, palmitate, and 10-methyloctadecanoate. The phosphate was released by alkalinolysis and identified by thin-layer chromatography and gas chromatography-mass spectrometry as myoinositol 1-phosphate. Thus, the group-specific "arabinomannan" of the genus *Mycobacterium* in the native state is acylated, contains the substituents of phosphatidylinositol, and is apparently membrane associated. LAM-B is one of the dominant immunogens of the leprosy bacillus reacting readily with antibodies from lepromatous leprosy patients and monoclonal antibodies in plate and nitrocellulose enzyme-linked immunosorbent assay and on electrophoretic immunoblots. It is immunologically cross-reactive with a like product from *M. tuberculosis*. LAM-B is clearly the pervasive "glycoprotein" antigen of the leprosy bacillus and may be the long sought lipoteichoic acid-like polymer of *Mycobacterium* with a role in cell wall physiology, macrophage recognition, and perhaps an involvement in cross-protective immunity.—Authors' Abstract

Lee, Y. N. and Colston, M. J. The measurement of adenylate energy charge (AEC) in mycobacteria, including *Mycobacterium leprae*. FEMS Microbiol. Lett. **35** (1986) 279–281.

The adenylate energy charge (AEC) (ratio of the mole fraction of ATP plus half the mole fraction of ADP in the total adenine nucleotide pool) of *Mycobacterium leprae, M. lepraemurium* and the cultivable *M. smegmatis* were determined following incubation in a variety of culture conditions. The AEC values for *M. smegmatis* were similar to those reported for other cultivable bacteria. The AEC values for *M. leprae* and *M. lepraemurium* purified from host tissue were lower than those of *in vitro*-grown organisms. The possible use of the AEC in *in vitro* studies with *M. leprae* is discussed.—Authors' Summary

Portaels, F., Asselineau, C., Baess, I., Daffé, M., Dobson, G., Draper, P., Gregory, D., Hall, R. M., Imaeda, T., Jenkins, P. A., Lanéelle, M. A., Larsson, L., Magnusson, M. Minnikin, D. E., Pattyn, S. R., Wieten, G. and Wheeler, P. R. A cooperative taxonomic study of mycobacteria isolated from armadillos infected with *Mycobacterium leprae*. J. Gen. Microbiol. **132** (1986) 2693–2707.

Seventeen strains of mycobacteria, recovered from six armadillos experimentally infected with *Mycobacterium leprae,* were examined in ten different laboratories. This collaborative study included use of conventional bacteriological tests, lipid analyses, determination of mycobactins and peptidoglycans, characterization by Py-MS, and immunological, metabolic, pathological and DNA studies. These armadillo-derived mycobacteria (ADM) formed five homogeneous groups (numbered ADM 1 to 5) on the basis of phenetic analyses. However, DNA studies revealed only four homogeneous groups since group ADM 1 and one of the two strains in group ADM 3 showed a high level of DNA relatedness. The phenetic and DNA studies confirmed that the ADM strains differed from all other known mycobacteria. Cultural, biochemical, metabolic and pathogenic properties as well as DNA–DNA hybridizations clearly differentiated these ADM from *M. leprae*.—Authors' Abstract

Experimental Infections

Fukunishi, Y. [Lysosomes and leprosy bacilli.] Jpn. J. Lepr. **54** (1985) 75–81.

The author examined by electron microscopy leprosy lesions from a hunman case of lepromatous leprosy, from nude mice inoculated with *Mycobacterium leprae* from human tissue, from armadillos inoculated with *M. leprae* of human origin, and from mangabey and rhesus monkeys inoculated with organisms from a naturally infected mangabey monkey. "It was considered that the multiplication by transverse fission of leprosy bacilli was done inside the phagolysosomes of each lepra cell of human and experimental lepromata."—C. A. Brown (Trop. Dis. Bull.)

Ha, D. K. K., Lawton, J. W. M. and Gardner, I. D. The effect of *in vivo* modulation of macrophage activities on *Mycobacterium lepraemurium* infection. J. Comp. Pathol. **96** (1986) 565–573.

During the early stage of *Mycobacterium lepraemurium* (*Mlm*) infection in mice, the mononuclear phagocyte system (MPS) was activated nonspecifically as demonstrated by enhanced listericidal activity. Such listericidal activity could be further increased by *Corynebacterium parvum* treatment, indicating that *Mlm* was not a good MPS stimulant. *C. parvum* treatment conferred only marginal protection upon mice during *Mlm* infection, as shown by the slight but significant prolongation of survival time and decreased bacillary load. In contrast, mice could not control splenic *Listeria* growth in the later stage of infection regardless of *C. parvum* treatment. Adoptive transfer of *Listeria*-immune spleen lymphocytes, however, did significantly suppress splenic *Listeria* growth. The significance of these findings is discussed.—Authors' Summary

Prabhakaran, K., Harris, E. B. and Hastings, R. C. Dapsone-resistance in *Mycobacterium leprae*: a genotypic change. ICRS Med. Sci. **14** (1986) 829.

Mycobacterium leprae (10^4) inoculated in the mouse foot pads multiplied to over 10^6 in about 6 months. Growth of the bacilli in the presence of 0.01% dapsone indicates a high degree of resistance to the drug. This property was retained through three passages of the bacteria in untreated mice. During the three passages lasting nearly 2 years, *M. leprae* would have gone through several generations. The fact that the bacteria did not revert to DDS sensitivity (when grown in the absence of the drug) suggests that the resistance is not a phenotypic adaptation induced by DDS. On the contrary, sulfone resistance seems to be a heritable genetic change in *M. leprae* caused by mutation or by selection of resistant mutants.—(*From* the article)

Walsh, G. P., Meyers, W. M. and Binford, C. H. Naturally acquired leprosy in the nine-banded armadillo: a decade of experience 1975–1985. J. Leukocyte Biol. **40** (1986) 645–656.

A decade has passed since our first report of naturally acquired leprosy in the nine-banded armadillo. Our studies and those of others during this period confirm the identification of the etiologic agent as *Mycobacterium leprae*. Confirmation is based on the results of histopathologic examination and microbiologic evaluations that included attempts to culture the organism, fluorescent antibody studies, mycolic acid analysis, and DNA determinations demonstrating complete relatedness between the natural agent and *M. leprae*. Surveys involving large numbers of animals demonstrate a significant prevalence of the disease in armadillos captured in Louisiana and Texas. The discovery of naturally acquired leprosy in a chimpanzee in 1977 and a sooty mangabey monkey in 1979 reinforce the concept of leprosy as a zoonosis. Extensive contact with armadillos has been implicated by other observers in seven patients with leprosy in Texas. We believe the prevalence of leprosy in wild armadillos requires that they be con-

sidered a source of infection in patients from geographic areas where leprosy and armadillos co-exist.—Authors' Abstract

Wang, H., Liu, J., Huang, H., Zhang, Y. and Lu, W. [Preliminary observation on experimental infection with *Mycobacterium leprae* in hedgehogs (*Erinaceus europaeus* Linné). Chung Kuo I Hsueh Ko Hsueh Yuan Hsueh Pao **7** (1985) 380–383. (in Chinese)

Hedgehogs were infected with *Mycobacterium leprae* in suspension (10^7/ml) by intravenous inoculation. The groups of hibernant and nonhibernant hedgehogs were observed comparatively for 11 to 37 months (average 26.5 and 27.2 months). All of the former group showed positive lesions histopathologically or had countable acid-fast bacilli in the slides of tissue homogenates. Similar lesions were found in only approximately one-third of the latter group. However, typical leprous lesions did not occur in any of the experimental animals of either group. The significance of the results is discussed and analyzed.—Authors' English Abstract

Epidemiology and Prevention

Boerrigter, G. and Ponnighaus, J. M. Ten years' leprosy control work in Malawi (Central Africa)—I. Methods and outcome after treatment. Lepr. Rev. **57** (1986) 199–219.

This paper describes the organizational structure of the LEPRA Control Project in Malawi (Central Africa) as it has evolved since 1973. It is meant to serve as a background to a series of publications which will describe the pattern of endemicity in Malawi, relapse rates, pattern of disabilities and duration of treatment required to achieve slit-skin smear negativity. In addition to describing the methods employed by the LEPRA Control Project, some data are presented on the outcome after treatment in a cohort of patients. It would seem that probably only 3–5% of this cohort did not benefit appreciably from the antileprosy treatment offered.—Authors' Summary

Freerksen, E. [The Paraguay project; a short report on a leprosy/tuberculosis eradication program.] Fortschr. Med. **104** (1986) 105–107. (in German)

The successfully completed leprosy eradication project in Malta and the leprosy/tuberculosis control program in Paraguay, which is being conducted at present, serve as models. For the first time it could be shown that with a combination of antimycobacterial working agents, which originate from tuberculosis research, it is possible to eliminate both mycobacterial diseases in a population by a joint action. The simple application of the medication and the form of the outpatient treatment are of especial importance in the developing countries.—Author's English Summary

Guha, P. K., Pandey, S. S., Singh, G. and Kaur, P. Family studies of leprosy cases. Indian Med. Gaz. **119** (1985) 148–149.

First degree relatives among the intrafamilial contacts of 400 leprosy patients were examined to detect any evidence of the disease in them. The type of leprosy detected in contacts in relation to that in the index cases was analyzed in order to evaluate the role of a genetic factor in determining the type of leprosy one suffers from. Observations in this study, however, do not tend to indicate the existence of a genetic diathesis.—(*From* Excerpta Medica)

Job, C. K., Harris, E. B., Allen, J. L. and Hastings, R. C. Thorns in armadillo ears and noses and their role in the transmission of leprosy. Arch. Pathol. Lab. Med. **110** (1986) 1025–1028.

Both ears from 494 wild nine-banded armadillos (*Dasypus novemcinctus*) and nose specimens from 224 animals were collected and histopathologically studied. Lepromatous granulomas were present in the ear specimens of 10 of 494 animals. There were thorns in the ears of 22.5% of animals, and in 36.6% of the nose specimens. In one armadillo, there was evidence to suggest that

Mycobacterium leprae entered the tissue through the thorn pricks. In the normal habitat of the armadillo in Louisiana there are thorny bushes consisting mostly of the green briar and the southern dewberry. Thorn pricks as a means of transmission of leprosy in the wild armadillos is suggested.—Authors' Abstract

Kim, S. J., Choi, I. H. and Kim, J. D. The HLA antigens and leprosy in Korea. Yonsei Med. J. **26** (1985) 154–158.

To investigate the genetic factors in Koreans with leprosy, 157 unrelated leprosy patients have been typed for HLA antigens and compared with 162 healthy controls. The patient group consisted of 124 with lepromatous leprosy and 33 with tuberculoid leprosy. HLA-A11 was found to be increased in lepromatous leprosy ($p = 0.0005$). HLA-Aw33 was found to be increased in both lepromatous leprosy ($p = 0.0002$) and tuberculoid leprosy ($p = 0.005$). HLA-Cw5 was found to be decreased in lepromatous leprosy ($p = 0.009$). Frequencies of HLA-B antigens did not differ significantly between the leprosy patients and the healthy controls.—Authors' Abstract

Neelan, P. N., Sirumban, P. and Sivaprasad, N. Limited duration acedapsone prophylaxis in leprosy. Indian J. Lepr. **58** (1986) 251–256.

A randomized controlled chemoprophylaxis trial was carried out in Madras city using 560 disease-free household child contacts of 264 multibacillary cases as study subjects. In the study, 13 cases were diagnosed among 280 contacts who received three injections of acedapsone at 10-week intervals as against 30 cases among 280 contacts who had the same number of placebo injections during the follow-up period of 225 weeks. The difference in the incidences in the two groups was statistically significant. (χ^2 6.45; $p < 0.02$). The protection due to the limited duration of acedapsone prophylaxis was 56.7%. There were no cases of multibacillary leprosy in either group. The efficacy of prophylaxis was significant in male children over 9 years of age and female children in the age group 1–8 years. The other prognostic factors like the infectivity status of the index cases in the household and the duration of exposure to them could have possibly influenced the effectiveness of prophylaxis in preventing progression from infection to clinical disease among the subjects studied. Their effects could not be assessed in this study.—Authors' Abstract

Ponnighaus, J. M. and Boerrigter, G. Ten years' leprosy control work in Malawi (Central Africa)—II. Patterns of endemicity since 1973. Lepr. Rev. **57** (1986) 221–236.

The British Leprosy Relief Association (LEPRA) has been operating a vertically structured mobile leprosy control service in five districts in the Southern Region of Malaŵi (Central Africa) since 1966. This service was extended from 1973 onward to the whole country with the exception of the two most southern districts.

Data concerning the patterns of endemicity of leprosy from 1974 to 1983 have been extracted from the available records of the LEPRA Control Project. It would seem that there are at least two distinctly different endemic situations in Malaŵi: districts on the Central African Plateau are characterized by a low level of endemicity of leprosy, while the districts in the Rift Valley along the shore of Lake Malaŵi have a level of endemicity that is five times higher. In both areas a parallel decline in detection rates has been observed since 1978–1979.

The authors consider this decline in detection rates to be a reflection of a genuine decline in incidence rates. This decline is accompanied by a trend toward higher lepromatous rates and shifts in the relative age distribution of new leprosy patients toward older age groups.

Projections are given concerning the expected numbers of new leprosy patients to be detected annually from 1984 to 1988.—Authors' Summary

Saylan, T. and Aytekin, A. H. Mass screening in leprosy endemic areas of Turkey: preliminary report. Lepr. Rev. **57** (1986) 243–249.

A multipurpose program was devised to enable a research team of university personnel to cooperate with local health units with the following objects: a) to find registered leprosy patients in selected areas and

provide them with curative and rehabilitative services; b) to locate people known to have been in close contact with registered patients and keep them under surveillance; c) to screen the whole population in selected areas in order to find new cases; d) to undertake periodic examination of suspected cases; and e) to provide on-the-spot training for health personnel in the control of leprosy. Because of a known high prevalence, the province of Van, situated in the eastern part of Turkey on the Iranian border, was chosen, and in the first year of this project, two regions of this province were covered, with the examination of over 15,000 people. This report describes the methodology and preliminary findings and discusses some of the difficulties in the interpretation of results with regard to the total population coverage achieved by such a screening program in 1984, and the previously recorded, official census figures. It is planned to continue and expand this research and to analyze results in a future publication.—Authors' Summary

Smith, W. C. S. and Parkhe, S. M. Disability assessment as a measure of progress in leprosy control. Lepr. Rev. **57** (1986) 251–259.

Leprosy control programs using methods of secondary prevention are in widespread use throughout the world. It is suggested that the most relevant method of assessing progress is by measuring the prevalence and incidence of disability. Disability assessment (WHO criteria) has been carried out in a control program in India at base line and now 4 years later. There has been little change in the prevalence of leprosy over this period but the prevalence of disability has fallen from 91.4 to 62.3 per 100,000. It is likely that this fall is the result of the control program, since there is no evidence of a secular trend of reduced disability in leprosy in India. The mechanism of this fall is thought to be the loss of disabled patients through death and migration; these are not being replaced by new disabled patients.—Authors' Summary

Rehabilitation

Birke, J. A. and Sims, D. S. Plantar sensory threshold in the ulcerative foot. Lepr. Rev. **57** (1986) 261–267.

Pressure threshold measurements were made using a set of three Semmes-Weinstein monofilaments on 132 plantar ulcer sites in 72 leprosy patients and 45 plantar ulcer sites in 28 diabetic patients. The most common sites of ulceration were the great toe and first metatarsal head. No patient could feel monofilaments smaller than 6.10 (75 g). The next smallest filament, 5.07 (10 g), was identified as the level of protective sensation in leprosy patients who customarily use footwear. Similar results were shown in a small group of diabetic patients.—Authors' Summary

Bourrel, P. [Realistic organization of surgery in countries with endemic leprosy.] Med. Afr. Noire **32** (1985) 377–384. (in French)

In 1969 the author wrote that the very limited number of specialized surgery centers and the budgetary difficulties involved with hospitalization mean that the great majority of leprosy cases must be treated as outpatients by general surgeons. It was argued that a surgeon capable of a few neurolyses and only four simple surgical repairs can prevent or treat almost 90% of all leprosy patients. The same author added in 1978 that the further development of complexity in treatment techniques and their reservation to rare specialized centers will produce the same dubious segregation accorded earlier to leprosy patients themselves. This remains true today and whenever conditions permit all efforts must be made to ensure that patients benefit from existing surgery facilities. Treating leprosy cases does not involve that one accepts a certain pathogenetic theory based on a worst case approach; instead one does all one can with whatever is available. And if one finds that a patient benefits from simple effective surgery why refuse this in the name of prin-

ciple, habit or hygiene when this approach could prevent many mutilations and injury.—(*From* Excerpta Medica)

Jagirdar, P. C. The usefulness of acupuncture in leprosy. Am. J. Acupuncture **14** (1986) 155–158.

The damage to motor nerves in leprosy causes imbalance at various joints and these postural alterations result in various deformities. Active exercises which can prevent disuse atrophy of muscles are not possible when the muscles are completely paralyzed. Needleless electroacupuncture produces electric impulses similar to nerve impulses. Electroacupuncture done at the correct acupuncture points can give active exercises to the paralyzed muscles and thus prevent disuse atrophy of the paralyzed muscles. Electroacupuncture can serve as the most effective physical therapy to prevent and treat early deformities such as claw hand, foot drop, trophic ulcer, etc. Acupuncture can give relief from the neuritic pain in leprosy.—Author's Abstract

Other Mycobacterial Diseases and Related Entities

Arden Jones, M. P. and Coates, A. R. M. A novel semi-automated technique for measuring inhibition of intracellular mycobacterial growth by macrophage activating factor. J. Immunol. Methods **83** (1985) 273–281.

We have developed a rapid, *in vitro* method for measuring T lymphocyte-derived macrophage activating factor (MAF) which inhibits the proliferation of *Mycobacterium microti* within macrophages. This MAF may be important in the control of mycobacterial disease *in vivo*. Because MAFs are a heterogeneous group of factors with different activities there is a need for assays which are relevant to specific macrophage effector functions. The existing assays for MAFs which are relevant to killing or inhibition of replication of mycobacteria within macrophages are currently too cumbersome or time consuming for the large-scale screening required for their detection and purification. In our assay, monolayers of mouse macrophages were infected with the murine pathogen *M. microti* and were cultured for 6 days with MAF or control medium. The intracellular bacteria were stained with auramine-O and were quantified using epifluorescence microscopy with television image analysis. Total assay time was one-seventh that of viable count methods, and image analysis estimates of bacterial loads are quicker and more objective than visual counts.—(*From* Excerpta Medica)

Collins, C. H. and Uttley, H. C. *In-vitro* susceptibility of mycobacteria to ciprofloxacin. J. Antimicrob. Chemother. **16** (1985) 575–580.

Two hundred seventy-six strains of mycobacteria were tested for susceptibility to ciprofloxacin (Bay 0 9867), a 4-quinolone antimicrobial agent. Most strains of *Mycobacterium tuberculosis, M. fortuitum, M. kansasii, M. marinum,* and *M. xenopi* were sensitive to minimum inhibitory concentrations (MICs) of 0.78–1.56 mg/l, equivalent to resistance ratios of 1 or 2. Most strains of *M. avium-intracellulare* and *M. chelonei* required MICs of 12.5 mg/l or more, giving resistance ratios of 8.—(*From* Excerpta Medica)

David, H. L., Clavel-Seres, S. and Clement, F. [Incorporation of ^{32}P into the phospholipids of *Mycobacterium leprae.*] Ann. Microbiol. (Paris) **136** (1985) 303–310. (in French)

Chromatographic experiments showed that the fastest moving phospholipid of *Mycobacterium leprae* could be identified as phosphatidylethanolamine. The results show that the phospholipids are synthesized by preformed enzymes; biosynthesis stops once the bacteria are suspended in an artificial or laboratory environment and are no longer capable of ensuring a protein synthesis.—English Summary from Excerpta Medica

Douvas, G. S., Looker, D. L., Vatter, A. E. and Crowle, A. J. Gamma interferon activates human macrophages to become tumoricidal and leishmanicidal but enhances replication of macrophage-associated mycobacteria. Infect. Immun. **50** (1985) 1–8.

Recombinant human gamma interferon (rIFN-γ) was examined for its ability to activate human peripheral blood monocyte-derived macrophages to kill tumor cells and to affect the replication of two phylogenetically distinct intracellular pathogens. *Mycobacterium tuberculosis* and *Leishmania donovani*. Macrophages preincubated overnight with doses of rIFN-γ from 5 to 500 U/ml killed [^{3}H]thymidine-labeled mouse L929 tumor targets, as measured by the release of [^{3}H]thymidine into the supernatant after 48 hr. Counts of macrophages initially infected with leishmania promastigotes showed that rIFN-γ-pretreated macrophages could both inhibit the replication of and kill the resulting intramacrophage amastigotes over a 7-day period. However, rIFN-γ pretreatment of macrophages actually enhanced mycobacterial replication over a 5- to 7-day period, as assessed by a) counting acid-fast bacilli or b) lysing macrophages to release bacteria and determining the numbers of viable units. Mycobacterial growth was not affected by rIFN-γ in the absence of macrophages. rIFN-γ pretreatment had opposite effects on the uptake of mycobacteria and leishmania. As many as 80% fewer activated macrophages ingested mycobacteria compared with controls; whereas 50% more activated macrophages were infected with leishmania. These results suggest that rIFN-γ may interfere with the immune destruction of intracellular tubercle bacilli and that the mechanisms of immunity against mycobacteria and leishmania may differ.—(*From* Excerpta Medica)

Grüneberg, R. N., Emmerson, A. M. and Cremer, A. W. F. Rifampicin for non-tuberculous infections? Chemotherapy **31** (1985) 2324–2328.

Large populations of rifampin-sensitive strains of *Mycobacterium tuberculosis* have been exposed *in vitro* to changing concentrations of rifampin (RMP) in line with changes in the blood level of the drug observed during treatment, and to much lower concentrations. Experiments in which the organism was exposed to either 7 or 14 days of cyclically-changing rifampin concentrations have resulted in the elimination of the *M. tuberculosis* test strains without the emergence of RMP resistance. The significance of these laboratory findings is discussed in relation to the debate as to whether rifampin should be used in short courses for the treatment of nontuberculous infections or whether it should be withheld for fear of inadvertently generating rifampin-resistant strains of tubercle bacilli. It is argued that the evidence for withholding rifampin from use in short courses against nontuberculous infections is slight.—Authors' Abstract

Jagadha, V., Andavolu, R. H. and Huang, C. T. Granulomatous inflammation in the acquired immune deficiency syndrome. Am. J. Clin. Pathol. **84** (1985) 598–602.

Granulomas were found in 16 biopsied specimens from eight patients with the acquired immune deficiency syndrome (AIDS), a disease characterized by a profound suppression of the T-cell arm of immunity. The pathogens were *Mycobacterium avium-intracellulare* (1), *M. tuberculosis* (3), *Histoplasma capsulatum* (2), *Candida albicans* (1), and unidentified in one patient. The sites of granuloma formation included the lung in 2, the pleura in 1, the liver in 3, the bone marrow in 6, the skin in 1, and the lymph node in 3 cases. The granulomas were epithelioid in nature, with aggregates of epithelioid histiocytes and macrophages. They were by and large small and loosely formed, with minimal or absent lymphocytic cuffing. Although it is a well-recognized concept that T-cell and macrophage interaction plays an important role in the granulomatous inflammatory response, granulomas have been produced in experimental animals independent of cell-mediated immune mechanisms. Granuloma formation in AIDS patients may well represent a clinical example of such a phenomenon.—(*From* Excerpta Medica)

Labidi, A., David, H. L. and Roulland-Dussoix, D. Cloning and expression of my-

cobacterial plasmid DNA in *Escherichia coli.* FEMS Microbiol. Lett. **30** (1985) 221–225.

The authors examined a small, 5.0-kb plasmid from *Mycobacterium fortuitum,* designated pAL5000. A restriction map of this plasmid was established. The complete sequence of pAL5000 was cloned in three different sites (BamHI, EcoRI, and EcoRV) of pBR322, and in the three possible orientations relative to the vector-derived sequence. The pBR322:pAL5000 hybrids were used to transform the *Escherichia coli* mutant (minA, minB). Two of the hybrid plasmids (pAL15 and pAL51) coded for the same protein in the minicells, indicating that a sequence of mycobacterial plasmid DNA may function as a promotor recognized by the transcription-translation apparatus of *E. coli.*—(*From* Excerpta Medica)

Labidi, A., David, H. L. and Roulland-Dussoix, D. Restriction endonuclease mapping and cloning of *Mycobacterium fortuitum var. fortuitum* plasmid pAL5000. Ann. Inst. Pasteur Microbiol. **136** (1985) 209–215.

A restriction map of *Mycobacterium fortuitum var. fortuitum* plasmid pAL5000 was established. The unique sites for ApaI, BamHI, BglII, BstEII, ClaI, EcoRI, EcoRV, HpaI, KpnI and NarI were located on the 5.0-kb plasmid. The plasmid had no sites for AhaIII, BclI, HindIII, PstI, SphI and XbaI. pAL5000 was cloned into pBr322 and propagated in *Escherichia coli.* Three hybrid pAL5000-pBR322 plasmids carrying the complete pAL5000 sequence were constructed by joining the plasmids at their BamHI, EcoRI or EcoRV sites. We also cloned into these plasmids a 1489-bp DNA fragment conferring resistance to kanamycin and originating from the streptococcal plasmid pJHl. The construction of these plasmids will facilitate the analysis and manipulation of pAL5000, and may allow the development of a vector system for genetic analysis in mycobacteria.—(*From* Excerpta Medica)

Mustafa, A. S. and Godal, T. BCG-induced suppressor T cells optimal conditions for *in vitro* induction and mode of action. Clin. Exp. Immunol. **62** (1985) 474–481.

In vitro activation with BCG of T cells from healthy individuals vaccinated with BCG lead to the induction of suppressor cells that suppressed the proliferation of fresh T cells in response to specific antigen. Kinetics of their induction revealed that they became radioresistant by day 8 and persisted up to 18 days of the culture period. Optimal antigen and monocyte concentrations as assessed by proliferation during the induction phase also resulted in maximum suppression. The strongest suppressor activity was observed when suppressor cells were added at an early time of fresh cell activation. IL-1 production from adherent cells in response to BCG was not affected, but IL-2 production by T cells was considerably reduced in the presence of suppressor cells. IL-1 containing supernatants and affinity purified IL-1 exogenously added to the culture system did not affect suppression. Whereas, recombinant IL-2 partially abrogated suppression in a dose-dependent manner. Further experiments suggested that suppressor cells might have inhibited BCG-induced IL-2 receptor expression on fresh T cells.—(*From* Excerpta Medica)

Pan, Y., Liu, S. and Li, W. [Laboratory studies on antituberculous activity of cyclopentylrifampicin. II. A long-acting antituberculotic cyclopentylrifampicin.] Chin. J. Antibiot. **10** (1985) 305–311. (in Chinese)

Cyclopentylrifampin (R-77-3, DL-473) has a longer and greater activity than rifampin and R-76-1 against tuberculosis in mice. R-77-3 has an unusually longer serum half-life (32.8 hr) and it was about 5 times and 2 times longer than rifampin (8.1 hr) and R-76-1 (17.1 hr). It has also been demonstrated that the concentrations of R-77-3 in plasma, the inhibitory and bacteriocidal activities of plasma in mice on *Mycobacterium tuberculosis* H37Rv can be continued about 5 days and 2–3 days, respectively. The longer activity of R-77-3 againt tuberculosis *in vivo* was confirmed by the pre-administration experiments. Only oral administration (10 mg/kg) once before infection by *M. tuberculosis* H37Rv was protective, the period of protection can be continued for 5 days, and it was longer than rifampin and R-76-1.—(*From* Excerpta Medica)

Toida, I., Yamamoto, S., Takuma, S., Suzuki, T. and Hirata, M. Lack of tuberculin activity of synthetic peptides. Infect. Immun. **50** (1985) 614–619.

We synthesized an octapeptide, H-Asp-Gly-Gly-Ser-Glu-Ser-Gly-Gly-OH, and a hexadecapeptide, H-Asp-Gly-Gly-Ser-Glu-Ser-Glu-Gly-Lys-Asn-Gly-Ser-Gln-Met-Arg-Leu-OH, which corresponded to amino acids 61 to 68 and 61 to 76, respectively, of the amino acid sequence of a crystalline protein reported to be tuberculin active. Authenticity and purity of the synthesized peptides were confirmed by high-pressure liquid chromatography, amino acid analysis, mass spectrometry, and protein sequencer analysis. Tuberculin activity of the synthesized peptides was examined in guinea pigs sensitized with *Mycobacterium tuberculosis* or *M. bovis* BCG and in tuberculin-positive healthy humans. Neither the octa- nor the hexadecapeptide was as active as tuberculin skin-test antigen.—Authors' Abstract

van Eden, W., Holoshitz, J., Nevo, Z., *et al.* Arthritis induced by a T-lymphocyte clone that responds to *Mycobacterium tuberculosis* and to cartilage proteoglycans. Proc. Natl. Acad. Sci. U.S.A. **82** (1985) 5117–5120.

Adjuvant arthritis characterized by chronic inflammation of the joints of rats is induced by immunization to *Mycobacterium tuberculosis.* To learn how autoimmune arthritis may be caused by a microbial antigen, we isolated a T-lymphocyte clone specific for *M. tuberculosis* antigens that was strongly arthritogenic. We now report that the clone recognized, in addition to *M. tuberculosis* antigens, antigens present in human synovial fluid, medium of chondrocyte cultures, and proteoglycans purified from cartilage. These observations indicate that the target antigen for the arthritogenic clone resides in the proteoglycan component of cartilage. Since this arthritogenic clone shows specificity for both a *M. tuberculosis* antigen and a cartilage constituent, we conclude that disease is probably caused by antigenic cross-reactivity. Thus, an autoimmune disease may be triggered by structural mimicry between antigens in the environment and self-antigens in the individual.—(*From* Excerpta Medica)

Wallace, R. J., Jr., Swenson, J. M., Silcox, V. A. and Bullen, M. G. Treatment of nonpulmonary infections due to *Mycobacterium fortuitum* and *Mycobacterium chelonei* on the basis of *in vitro* susceptibilities. J. Infect. Dis. **152** (1985) 500–514.

One hundred twenty-three patients with nonpulmonary infections due to *Mycobacterium fortuitum* or *M. chelonei* were treated by wound debridement and with chemotherapy on the basis of *in vitro* susceptibilites of the organism. Of 76 patients with infections caused by *M. fortuitum,* 13 required no therapy or were adequately treated with surgery alone. Patients with active localized disease received single-drug therapy (usually with a sulfonamide) for a mean period of 10.6 weeks for cellulitis and 7 months for osteomyelitis. Patients with extensive disease received amikacin or amikacin plus cefoxitin (mean, 4 weeks) followed by a sulfonamide (mean, 6 months). The 47 patients with infections caused by *M. chelonei* received no therapy or were treated with surgery alone (6); with amikacin (10), erythromycin (6), doxycycline (3), or cefoxitin (1); or with amikacin plus cefoxitin followed by cefoxitin alone for a total of 10–12 weeks (20); or other multiple-drug regimens (1). Surgery was performed on 74 (60%) patients. Schlichter tests or serum drug levels were determined for 81 (66%) patients. Response to therapy was excellent; 68 (90%) infections with *M. fortuitum* and 34 (72%) with *M. chelonei* were successfully treated. Cultures became negative within 6 weeks of chemotherapy, except for sternal osteomyelitis for which cultures were not negative until up to 14 weeks. Follow up for a mean period of 12 months following therapy was possible in 80% of cases. Relapses were rare except in patients with disseminated disease, and drug resistance developed in only one patient. These studies demonstrate the value of routine susceptibility testing of these mycobacterial species and the benefit of chemotherapy on the basis of *in vitro* susceptibilities.—(*From* Excerpta Medica)

Wallis, R. S., Fujiwara, H. and Ellner, J. J. Direct stimulation of monocyte release of interleukin 1 by mycobacterial protein

antigens. J. Immunol. **136** (1986) 193–196.

We examined stimulation of monocyte (MN) release of interleukin-1 (IL-1) by soluble microbial products. MN from tuberculin skin test nonreactive donors incubated with PPD (100 μg/ml) released IL-1 activity of 80.5 ± 33.9 U/ml (mean ± SD, N = 6), similar to that induced by optimal concentrations of LPS (76.4 U/ml). OKT3-reactive cells were not required for this process. PPD-stimulated IL-1 release by MN did not appear to be due to endotoxin contamination, as a) PPD contained 0.01% endotoxin, b) MN incubated in LPS (0.1 μg/ml) produced 19.5 ± 13.9 U/ml, significantly less than PPD ($p = 0.03$), and c) addition of polymyxin B (12.5 μg/ml) abrogated IL-1 production in response to LPS (0.1 μg/ml) but had no significant effect on PPD induction of IL-1. Antigen 5, a partially purified cytoplasmic antigen of *Mycobacterium tuberculosis,* had similar IL-1-inducing effects. Arabinogalactan (a mycobacterial polysaccharide), streptolysin O, and tetanus toxoid did not. Thus, mycobacterial protein antigens directly stimulate MN to release IL-1. This property may be central to the response of the naive host to mycobacterial infection and may play a pathophysiologic role in tuberculosis.—(*From* Excerpta Medica)

Yabu, K., Kaneda, S. and Ochiai, T. Relationship between β-lactamase activity and resistance to β-lactam antiobiotics in *Mycobacterium smegmatis.* Microbiol. Immunol. **29** (1985) 803–809.

Penicillin-susceptible mutants and β-lactamase-negative mutants were isolated from *Mycobacterium smegmatis* after nitrosoguanidine mutagenesis. Both the mutants were found to be susceptible to low levels of penicillin and cephalosporins by twofold dilution testing. Clavulinic acid reduced the minimal inhibitory concentrations of β-lactamase-labile β-lactams for the penicillin-susceptible mutants and the parent strain, but had no effect on the susceptibility of the β-lactamase-negative mutants. Comparison of the β-lactamase activities found in these mutants and the parent strain indicated that there was a rough correlation between the β-lactamase level in these organisms and their susceptibility to β-lactams.—(*From* Excerpta Medica)

Young, D. B. and Lamb, J. R. T lymphocytes respond to solid-phase antigen: a novel approach to the molecular analysis of cellular immunity. Immunology **59** (1986) 167–171.

Using cloned human T lymphocytes reactive with a 24 amino acid peptide (p20) of the carboxyl terminus of the HA-1 molecule of influenza hemagglutinin (HA), we have investigated the ability of solid-phase antigen to induce antigen-specific T-cell proliferation. The activation by nitrocellulose-bound virus and p20 was accessory-cell dependent and was not caused by immobilized antigen directly cross-linking the specific receptors. Furthermore, we report that separation of complex antigen mixtures such as influenza virus and HA by polyacrylamide gel electrophoresis under denaturing conditions (SDS-PAGE) followed by transfer to a nitrocellulose membrane can be used to allow direct screening of individual polypeptides in T-cell proliferation assays. With this immunoblotting procedure the antigenic site recognized by HA-reactive T cells was confirmed to reside in the HA-1 molecule of influenza virus of only the appropriate subtype. The general application of this approach is discussed in the case of infections and autoimmune diseases in which the immune response is predominantly T-cell mediated and where antibody studies may fail to identify key antigenic determinants involved in the activation of T cells.—Authors' Summary