# CURRENT LITERATURE

*It is intended that the current literature shall be dealt with in this department. It is a function of the Contributing Editors to provide abstracts of all articles published in their territories, but when necessary such material from other sources is used when procurable.*

*(Due to lack of space in* THE JOURNAL, *Vol. 27, No. 1, 1959, the following abstracts were held over for this issue. Asst. Ed.)*

LECHAT, M. L'ACTH dans le traitement de l'orchite lépromateuse. [ACTH in the treatment of lepromatous orchitis.] Ann. Endocrinol. **17** (1956) 270-275.

Six cases of orchitis occurring in lepromatous patients are recorded, 5 of whom had received cowpox vaccination. Treatment by ACTH was given in 10 mgm. dosage for 3-5 days, during which time the swelling and pain of the testes disappeared. The last patient received without accident a single dose of 60 mgm. In this preliminary note the author wonders if hormone treatment associated with sulfones would not permit, in certain cases, a more efficient chemotherapy of leprosy. —A. DUBOIS

LECHAT, M. F. Le traitement de la réaction lépreuse. Revue de la littérature et données actuelles. [Treatment of lepra reaction; review of literature and present information.] Maroc Méd. **36** (1957) 1039-1062.

In this review of 23 pages, with a bibliography containing 158 references, the author presents from the clinical point of view the various incidents called "reactions" which occur in leprosy patients. Also reviewed are the numerous treatments which have been recommended for "lepra reaction" and for the syndrome generally called "erythema nodosum leprosum." Of these treatments, those which seemed to him of interest are: Anthiomaline or vitamin K for mild lepra reactions, and Chlorpromazine, transfusions of plasma, and long-acting ACTH for severe cases. Cortisone and "cortisone-like" drugs may also be added. For ENL repository ACTH is recommended, and pregnenolone for the frustrate forms. For dermatitis he suggests, depending on the severity, the antihistamines, vitamin PP, or plasma transfusions. —M. VIETTE

[This extensive and valuable review has been reprinted, as Bulletin d'Information sur la Lepre No. 5, by the Père Damien Section of FOREAMI (Fonds Reine Elisabeth pour l'Assistance Médicale aux Indigènes du Congo Belge), 14 Square de Meeus, Brussels, to which requests for copies should be addressed.

[The following part of the author's introduction seems worth repeating because of the unusual point of view. Si l'on nous permet une comparaison triviale, nous dirons que l'évolution de la lèpre chez quelques milliers de malades fait penser au cheminement des automobiles dans le centre de Manhattan, vu par un étranger de la terrasse du Rockefeller Center, un vendredi a 5 heures de l'apres-midi. Les voitures suivent certaines grandes avenues dans un sens déterminé, comme les lépreux, qui sont d'une forme on de l'autre; elles progressent lentement, comme la lèpre; elles se dirigent toutes vers la banlieue, comme la lèpre qui, sans traitement, évolue le plus souvent de manière inéluctable. Le léprologue qui édifie une classification est semblable à l'étranger qui voudrait ainsi, du soixante-sixiéme étage, établir les lignes de force auxquelles obéit le flux des voitures et reconstituer les règles de la circulation, ce qui n'est pas une tâche impossible. Cependant, de cet observatoire, on aperçoit parfois des conduites isolées et aberrantes: une voiture qui remonte une avenue à contrecourant, une autre qui tourne court dans un virage, une autre encore qui s'emballe et accélère brusquement. En lèpre, on donne à toutes ces conduites anormales le nom de réaction.]

REZETTE, J. Essais de traitement des ulcères et des maux perforants lépreux par le déhydrocholate de sodium. [Trial treatment of ulcers and leprous perforating sores by sodium dehydrocholate.] Ann. Soc. belge Méd. Trop. **36** (1956) 581-587.

Sodium dehydrocholate (Dycholium) was found useful in the treatment of ulcers and malum perforans in leprosy patients. The dosage was 1 gm. (5 cc.) intravenous, daily or even twice daily for 8-20 days. Because of scarcity of the drug, the treatment was limited to 9 patients. All of them were cured or greatly improved, especially ulcers of less than one month duration. No toxic effects were seen except some bouts of fever.
—A. DUBOIS

RAMANUJAM, K. Chlorpromazine in the "painful" complications of leprosy. Leprosy Rev. **28** (1957) 60-65.

Chlorpromazine (Largactil) was tried out in 31 leprosy cases with painful complications—neuritis (21), joint pains (3), neuritis and joint pains (2), and generalized body pain (5). The drug was administered orally in 25 mgm. doses 2 or 3 times daily. To a group of 18 patients, a sodium salicylate mixture was also given (and also Phenergan [promethazine] at bedtime). A second group of 9 patients received, in addition to the Largactil, distilled water intravenously. For a third group [originally only 4?] the additional treatment was potassium antimony tartrate intravenously. Of the first group, 12 patients (two-thirds) had complete relief in 4-16 days, after which the others were transferred to the third group. Of the second group, 5 had complete relief in 5-9 days, and the others were also transferred to the third group. This group, which finally totalled 13, all obtained complete relief with the combined treatment. Largactil combined with antimony therefore appears to be the best treatment for these painful conditions, and it was found suitable for outpatient treatment.—[From abstract in *Trop. Dis. Bull.* **54** (1957) 833.]

[U.S.P.H.S.] Chemotherapy of tuberculosis, progress and promise. Publ. Hlth. Rep. **72** (1957) 412-420.

One of the points in this unsigned review, from the Division of Special Health Services, is that in the course of the investigations reviewed, "bacteriological change had emerged as the most sensitive index of the effectiveness of antimicrobial agents." [Whether or not this may be applicable to lepromatous leprosy, it hardly fits the picture of tuberculoid leprosy.] Much thought is being given to the possibilities of the prophylactic possibilities of isoniazid. A nationwide study has been started, one of the questions being whether or not the drug would prevent infection and disease in highly exposed contacts. Another is that of its effect on previously infected persons not in highly exposed situations. "This difficult and costly investigation may show that isoniazid has no prophylactic value, or that its value is offset by interference with natural immunity, or that it is effective only while it is being taken. It may show that it delays but does not prevent. On the other hand, if it is effective in any one of the areas under investigation . . . we will have gained an important public health weapon in the fight against tuberculosis." [This abstract is offered for the consideration of those who have responsibilities with respect to the control of leprosy in their countries, in connection with the moot question of whether or not contacts should be given prophylactic sulfone treatment.]
—H. W. W.

PODDAR, R. K. AND CHATTERJEE, K. R. Autoradiographic detection of sulphone in affected tissues of leprosy patients. Nature **180** (1957) 854-855 (correspondence).

In previous experiments [THE JOURNAL **25** (1957) 299] the authors had shown by assay of DDS labelled with $S^{35}$, that in leprosy patients the drug was preferentially concentrated in the affected tissues. They have now made autoradiographic studies to determine the exact site of localization of the drug in the affected tissues. Patients were given a single oral dose of $S^{35}$ (4 $\mu$c./kgm.), the initial specific activity of which was 1.0 $\mu$c./mgm. After 6-18 hours, biopsy specimens were taken and fixed in formalin. Five-

micron frozen sections on a slide were covered with autoradiographic stripping film and exposed for 180 days in dry, light- and air-tight chambers at 4°C., and so developed as to keep the background as low as possible. Positive autoradiographs are shown in 3 figures. One shows the drug in the transverse section of a blood vessel; the other two are of longitudinally- and transversely-cut sections of peripheral nerves. The density of the blackened photographic grains is more marked where infiltrating cells are predominant, and the space between adjacent fibrils is seen to be almost free of drug.—[From abstract in *Trop. Dis. Bull.* **55** (1958) 412.]

GOKHALE, S. K. AND GODBOLE, S. H. Serum lipolytic enzyme activity and serum lipid partition in leprosy and tuberculosis. Indian J. Med. Res. **45** (1957) 327-336.

The lipolytic enzyme activity of the blood is affected in certain pathologic conditions, supposedly because of an altered fat metabolism. The authors studied the lipolytic enzyme activity and its relation to lipid partition in the serums of 165 healthy subjects, 105 patients with leprosy, and 103 patients with tuberculosis. The serum constituents studied were phospholipids, total cholesterol, total fatty acids, total lipids, lipolytic esterase, and lipase with and without sodium tauroglycocholate activities. In patients with leprosy or tuberculosis the serum lipid constituents were higher than in normal subjects, but the lipolytic enzyme activity gave lower figures than normal. This difference was greater in patients with leprosy than in those with tuberculosis. This increase in the serum constituents and the decrease in the serum lipolytic enzyme activity indicate a disturbance in the fat metabolism in these two diseases.—[From Foreign Letters in *J. American Med. Assoc.* **165** (1957) 1472, supplied by Sr. Hilary Ross.]

SERIÉ, C. AND SCHALLER, K. F. L'électrophorese et la lèpre. [Electrophoresis in leprosy.] Bull. Soc. Path. exot. **50** (1957) 17-20.

Electrophoresis of the sera of 202 leprosy patients (81 lepromatous, 53 tuberculoid and 68 indeterminate) was carried out, each patient being examined twice with a 6-months interval. The patients were divided into four groups: (1) untreated, (2) BCG-vaccinated, (3) sulfone-treated, and (4) both vaccinated and sulfone-treated. In Group 1 the electrophoresis curve remained unchanged or became more abnormal after 6 months, in Groups 2 and 3 the curve improved in 61%, and in Group 4 it improved in 73%, particularly in the lepromatous cases (80%). The sulfone treatment and BCG vaccination therefore exert a favorable influence on the humoral state of the patient, most marked when they are combined. —M. VIETTE

WOZONIG, H. Die Knochenveränderungen bei Lepra. [Bone changes in leprosy.] Ztschr. Tropenmed. u. Parasitol. **7** (1956) 464-471.

Bone lesions in 50 leprosy patients in the Haile Selassie I Hospital, in Addis Ababa, have been studied by x-ray examination in order to evaluate their diagnostic significance. Bone lesions develop in almost every case in the course of the disease, usually beginning in the 4th to 6th year. They are usually confined to hands and feet. The terminal phalanges of fingers and toes, which are particularly involved, undergo slow atrophy and resorption. The process takes decades, and leads to gradual absorption, osteoporosis, decalcification, specific osteomyelitis, periostitis, cyst formation, arthropathies, laxations, spontaneous fractures, exostoses, synarthrosis, and pseudoarthrosis. These changes result from trophic disturbances secondary to neural involvement and are regarded as non-specific. Disturbances of blood supply and the development of anesthesia of the extremities lead to injuries and secondary infections. —E. KEIL

SERIAL, A. Algunas consideraciones sobre la técnica histologica de la forma tuberculoide de lepra. [Considerations on the histologic technique for the tuberculoid form of leprosy.] Leprología **2** (1957) 62-65.

The purpose of this article is to point out the importance of the fixation for the interpretation of histopathologic pictures in leprosy. In general, ordinary 10% formalin is not recommended for histopathology in dermatology. One long-experienced dermatolo-

gist is cited as recommending Helly's solution, or Zenker's fixative, ordinarily if not exclusively. Tuberculoid lesions fixed in formalin are liable to show marked vacuolization which may be confused with conditions to be seen in lepromatous leprosy lesions. [The present abstractor, who uses Zenker's fluid exclusively in regular work, heartily endorses the substance of this report.] —H. W. W.

GERNEZ-RIEUX, C. AND TACQUET, A. L'hémagglutination et l'hémolyse conditionée. Application au diagnostic de la tuberculose et de la lèpre. [Conditioned hemagglutination and hemolysis; application to the diagnosis of tuberculosis and leprosy.] Semaine Hôp. de Paris **32** (1956) 1129-1136.

This review covers the present concepts of conditioned hemagglutination and hemolysis reactions. In the diagnosis of tuberculosis the titers usually (and arbitrarily) adopted are 1/16 in adults and 1/8 in children. However, higher titers may be found in certain healthy persons, probably due to infections by paratuberculosis bacilli or even by nonacid-fast bacteria which have an antigenic relationship with the Koch bacillus. On the other hand, tuberculosis patients may have very low titers. Search for blocking antibodies or incomplete antibodies has been unsuccessful. In tuberculosis, it is not possible to distinguish infections by the human and bovine bacilli. The titers are little changed under the influence of treatment. On the contrary, in leprosy where the antibody titers are at times much higher than in tuberculosis, their diminution usually accompanies clinical improvement resulting from treatment. —M. VIETTE

KERBASTARD, P. Réaction d'hémagglutination dans la lèpre. [The hemagglutination reaction in leprosy.] Méd. Trop. **17** (1957) 251-263.

The hemagglutination test was performed in 165 recently-detected leprosy cases and in 20 healthy persons. In the latter the maximum titer was 1/8. It was higher than 1/8 in 13 out of 67 tuberculoid cases (19%), in 6 out of 12 indeterminate cases (50%), and in 64 out of 86 lepromatous cases (74%). Of another 64 lepromatous cases that had been treated for 2 years, only 47% gave titers above 1/8. The examination was repeated every 3 months for 12 to 18 months in 105 patients. Decrease of titer always corresponds to clinical improvement. Of the stationary titers, about one-half were observed in improved cases, the other half in cases which were clinically stationary or aggravated. Finally, an increase of the titer in tuberculoid and indeterminate cases had no relationship to an aggravation of clinical lesions, but in the lepromatous cases increase frequently accompanied clinical aggravation or the occurrence of frequent reactions.

—M. VIETTE

KENT, J. F., OTERO, A. G. AND HARRIGAN, R. E. Relative specificity of serologic tests for syphilis in *Mycobacterium leprae* infection. American J. Clin. Path. **27** (1957) 539-545.

Seven serologic tests and antigens (four slide flocculation tests, the VDRL, Kline, Rein-Bossak, and Mazzini; one tube flocculation test, the Kahn standard; two complement-fixation tests, the Kolmer and CF52) were performed at the Walter Reed Army Institute of Research, Washington, D. C., on serum specimens obtained from 34 native Cubans with leprosy, in 28 of whom the disease was of the lepromatous type, tuberculoid in 4, and neural in 2. None presented clinical or anamnestic evidence of treponematosis and all were serologically negative with the treponemal immobilization test. The frequency of reactions varied from 6 to 21 in tests with the lipoid antigens that are present in ordinary extracts of tissue, and from 7 to 17 in tests with cardio-lecithin-cholesterol antigens. The specificity of the cardiolipin antigens was correlated with their content of lecithin. —SR. HILARY ROSS

ROLLIER, R. AND PLANCHET, H. Essai d'interpretation des fausses séro-réactions dans la lèpre. [Interpretation of false serum reactions in leprosy.] Maroc Méd. **36** (1957) 1206-1212.

The serologic reactions for syphilis were studied in 515 leprosy patients, and with a certain number of them comparisons were made with the Nelson test, serum elec-

trophoresis, and the albumin-globulin ratio. In patients with benign forms (tuberculoid and indeterminate) the serology is comparable with that of the healthy population. Patients with the malign form often present dissociated serum reactions, and an abnormally high percentage of positive reactions, except with the Nelson test. Serum electrophoresis shows frequent disturbances in lepromatous cases, but they are not in accord with the modifications of the serologic reactions. The latter agree more with the variations of the albumin-globulin ratio and with the abundance of bacilli in the smears. Patients with bacillus-rich lesions and a markedly abnormal albumin-globulin ratio frequently show positive or dissociated serum reactions. These three characteristics subside at the same time that the leprosy infection subsides under the influence of treatment. It is concluded that the false serologic reactions for syphilis are real, ascribable to the presence of the Hansen bacillus or to its metabolic products. —M. VIETTE

ABE, M., MATSUO, K., TAKAHASHI, S., INABA, T. AND TACHIKAWA, N. The relation between serological reaction and lepromin reaction in leprosy. La Lepro **26** (1957) 297-304 (in Japanese; English abstract).

The lepromin reaction with the Dharmendra antigen [see note below], the leproagglutination test with the cardiolipin-lecithin antigen (Ogata), and the Middlebrook-Dubos hemagglutination reaction were tested in 571 cases of leprosy (436 lepromatous, 135 tuberculoid), and the relation of the intensities of the three reactions to the disease types and the interrelationships between the reactions were studied statistically. The (Dharmendra) lepromin reaction was negative in 391 (89.7%) of the lepromatous cases, and positive in 109 (80.7%) of the tuberculoid cases. End titers higher than 1:32 in the leproagglutination test were observed in 83.2% of the lepromatous cases, and less than 1:16 in 86.7% of the tuberculoid cases. The end titer of the Middlebrook-Dubos test in relation to the disease type was not so definite as with the previous two reactions, but the mean end titer was higher in the lepromatous cases. The lepromin reaction was in a negative correlation to both of the other tests, which two were in positive correlation. The lepromin reaction and leproagglutination are considered to be important means for determining the type of leprosy. [*Note:* The use of the term "lepromin reaction" is confusing and unjustified, for as shown by the heads of the tables in the text the reaction was the 48-hour one.]—[From the English abstract.]

COLLIER, W. A., BUENO DE MESQUITA, S. J. AND VAN ZANTEN, E. Kwantitatieve complementbinding en lepromineactie bij tuberculose. [Quantitative complement-fixation and lepromin reaction in tuberculosis.] Nederlandsch Tijdschr. v. Geneesk. **101** (1957) 2057-2063; *also, in English,* Doc. Med. Geograph et Trop. **9** (1957) 261-268.

In the leprosy-endemic environment of Surinam the quantitative complement-fixation test with the Essen tuberculosis antigen (Behring) was performed in 122 cases of tuberculosis and 219 lepromatous leprosy cases, and 52 healthy soldiers recently arrived from Europe were used as controls. Of the tuberculosis patients, 20% gave negative test, 34% were moderately positive, and 46% were strongly positive. The corresponding percentages for the leprosy cases were 14, 19 and 67. Of the 52 healthy controls 94% were negative, only 6% of the tests being positive. With the lepromin test 80% of the tuberculosis cases gave the early (Fernandez) reaction, and 39% of the leprosy cases did so. The positivity percentages for the Mitsuda reaction were 90 and 21, respectively. [The proportions of positive reactions are unusually high for active lepromatous cases.] The Mitsuda reaction was 3+ positive in 41% of the tuberculosis patients, and the impression was gained that tuberculosis infection markedly increases reactivity to lepromin. Further investigation is required to assess the value of the Mitsuda test in tuberculosis, in a leprosy-free environment. The connection between the results of the Mantoux and Fernandez tests is not clear. In tuberculous patients, however, there is a certain correlation between high antibody titers and strong Mantoux reactions (20 mm. and

over). An analogous correlation may be found between the Mantoux and Mitsuda reactions.—[From summaries in *Trop. Dis. Bull.* **54** (1957) 1423 and **55** (1958) 171.]

SATO, S., FUKUDA, M., KAMIKAWA, Y., MAJIMA, S., ABE, H. AND TAKEDA, M. Rescrutinizing of lepromin reaction, in particular, of Mitsuda's reaction. Sci. Rep. Res. Inst. Tohoku Univ. **7** (1957) 335-349.

The authors have studied the late reactions to lepromin in leprosy patients in 4 leprosaria, 2 in northern Honshu and 2 in southern Kyushu, mainly with respect to the increasing number of positive reactors among lepromatous cases. This finding has been reported in the past by certain South American writers (Mom, 1947; Fiol, 1948; Basombrio *et al.*, 1950; and Schujman, 1953), and also in Japan by certain of the present authors and others (Fukuda and Majima, 1945 and 1955; Saikawa, 1952; and Moriya, 1952, 1953, 1954. [See Sato and Fukuda, THE JOURNAL **26** (1958) 205-218.] The numbers of positive lepromatous cases had become so high that it was difficult to distinguish them on the basis of their reactivity from nonlepromatous cases. The numbers of cases in the present study were 492, 485, 235 and 265, totalling 1,477, of which 1,111 (75.2%) were lepromatous and 366 (24.8%) nonlepromatous. The cases were so classified for the purpose of this study, excluding the less well-understood borderline and indeterminate cases "in fear of committing blunders." Each patient was tested with a Hayashi-Mitsuda lepromin and Dharmendra's antigen. The early reactions with the two antigens showed good correlation with each other and with the types of the disease, so that reaction seems adequate for classification. With the Mitsuda antigen positive late reactions (taking 3 mm. as the lower limit of positivity) in the lepromatous cases were no less than 85.2% (14.8% over 6 mm.), and in the nonlepromatous cases 98.7%. Figures for the Dharmendra antigen are not shown on the same 3 mm. basis, but it appears that 53.6% of the lepromatous and 76.6% of the nonlepromatous cases gave late reactions 4 mm. and upward. On the basis of their findings 7 mm. would have to be used to separate positives and negatives. This increase in positive reactivity is ascribed chiefly to present-day chemotherapy. [The report contains no data on untreated cases tested as controls.] —H. W. W.

FUKUDA, M. Studies on the lepromin reaction. Part 4. Symptoms and clinical course of leprosy cases in which the lepromin reaction and the type do not agree. La Lepro **26** (1957) 305-310 (in Japanese; English abstract).

The early and late reactions to Mitsuda's antigen and the early reactions to Dharmendra's antigen were studied in 718 lepromatous and 181 nonlepromatous cases. Early reaction: Lepromatous cases, 5.3% positive with Mitsuda (mostly subsided cases) and 7.8% positive with Dharmendra (still showing residual skin lesions, and many bacteriologically positive). The late, Mitsuda reaction, on the basis of more than 3 mm., was positive in 88.3% of the lepromatous cases, more than half of which had skin lesions and positive smears. If, however, more than 7 mm. were to be used for judging positivity, only 15.2% of the lepromatous cases would have been read positive—and they mostly (but not entirely) subsided cases. The nonlepromatous cases would no longer be 100% positive; but the 13.3% negatives, it is said, coinncided with clinical observations.—[From the English abstract.]

ALONSO, A. M. AND AZULAY, R. D. Lepromin-reacões positivas e lepra lepromatosa. (Positive lepromin reactions and lepromatous leprosy.) Bol. Serv. Nac. Lepra (Rio de Janeiro) **15** (1956) 111-113.

Report of a lepromatous case which showed a 5 mm. nodule after a lepromin test made in July 1954, but was negative when tested 2 months later. After another 2 months, in November, and again in May 1955, he gave 5 mm. reactions, but was negative in the next two tests. It is pointed out that a simple clinical reading of the reaction to lepromin, without histologic control, may sometimes be misleading. —H. W. W.

Souza Campos, N. Les conditions qui déterminent la positivité de la réaction de Mitsuda. [Conditions which determine the positivity of the Mitsuda reaction.] Maroc Méd. **36** (1957) 1015-1019.

The lepromin or Mitsuda reaction is usually positive in leprosy patients with the benign form and negative in those with the malign form, positivity therefore being of good prognosis. It may also be positive in individuals who are apparently free from leprosy but living in contact with patients with the infective form and without contact with the Koch bacillus (the tuberculin reaction being negative). Such persons may remain leprosy-free, but should the disease appear it will be of good prognosis; in contrast, it is of bad prognosis in individuals who are Mitsuda negative. In persons without contact with leprosy patients, but in contact with the Koch bacillus and presenting a positive tuberculin reaction, the Mitsuda reaction is often positive. It is almost always positive in persons with tuberculosis. Finally, BCG vaccination is followed in a great number of cases by Mitsuda positivity. Certain authors believe that repeated injections of lepromin may convert negative reactors to positive, but this has not been sufficiently demonstrated. It can be said that the Mitsuda reaction, a sign of resistance to leprosy infection, is due to tuberculosis, or to leprosy (infection or disease), or to BCG vaccination. —M. Viette

Rotberg, A. Fator "N" de resistência à lepra e relações com a reatividade lepromínica e tuberculínica; valor duvidoso do BCG na imunização antileprosa. [The N factor in resistance to leprosy and its relations with reactivity to lepromin and tuberculin; doubtful value of BCG in antileprosy immunization.] Rev. brasileira Leprol. **25** (1957) 85-106.

This article deals at length with a subject with which the author treated briefly in a summary presented at the Madrid congress [The Journal **21** (1953) 590]. His "Factor N," held to be a basic and specific factor of resistance to leprosy infection, is said to be present in about 80% of the population, the other 20% being the "anergic margin." In the absence of this factor, tuberculosis infection would not induce lepromin positivity; hence lepromatous cases are typically lepromin negative, although they are tuberculin positive in considerable proportions. The author does not agree that tuberculization of a region leads to the disappearance of leprosy; on the contrary, active tuberculosis may aggravate latent or initial leprosy infections. Nor would BCG vaccination interfere with the anergic margin; it does not induce lepromin positivity in lepromatous cases, or in healthy people lacking the N factor. With that factor present, leprosy, tuberculosis, BCG vaccination, "and possibly other unknown factors," may produce lepromin positivity. BCG would become of importance in prophylaxis if it could be proved that it produces strong (2+ and 3+) lepromin reactivity in a significantly higher proportion of individuals than become lepromin positive spontaneously. —H. W. W.

Jopling, W. H. and Ridley, D. S. Lack of effect of cortisone on the negative lepromin test. Leprosy Rev. **28** (1957) 157.

The "conversion" of tuberculin negativity to positivity in sarcoidosis by adding cortisone to the tuberculin was not paralleled in lepromatous leprosy. Double-strength lepromin containing 1.25 mgm. of cortisone acetate in each 0.1 cc. was used in 6 lepromin-negative cases, but they failed to react to the mixture.—[From abstract in *Excerpta Medica* **12** (1958) 114.]

[Symposium] Indução da reactividade leprominica por meio de testagem repetida. [Induction of reactivity to lepromin by means of repeated testing.] Rev. brasileira Leprol. **25** (1957) 167-207.

The precipitating stimuli for this symposium, according to the editor, Nelson de Souza Campos, came from three things: (1) A review editorial in The Journal [**23** (1955) 310-315] in which it was noted that the positivization of originally lepromin

negative persons who were given BCG could not [in view of the repeated lepromin testing] properly be ascribed to the vaccination alone. (2) An article by Ignacio and associates on their experience with Culion children, which occasioned that editorial. (3) Reports of Bechelli and associates on "spontaneous" development of lepromin reactivity in persons who had been tested previously and found negative, which change Souza Campos suggested might be ascribable to the first test. Souza Campos sought widely among authorities in the field of leprosy, bacteriology, and immunity for opinions concerning the following two alleged points of view, both based on the supposedly settled thesis that Mitsuda positivity signifies resistance, or immunity, to leprosy infection: (1) Some workers hold that the simple inoculation of lepromin will convey immunity. [This is a claim which the reviewer has not heard made, although certain workers are known to be investigating the possibility.] (2) Most workers agree that to establish resistance or immunity, with development of lepromin positivity, requires that there be actual infection with *M. leprae*, or *M. tuberculosis*, or vaccination with BCG. He then asked which of these hypotheses could be supported on immunobiologic grounds. Although there were some who preferred not to contribute, 26 answers by 29 writers were received—and are published in the Portuguese language. The statements are of great variety with respect to length and content, and many do not conform to the questionnaire. Summarizing at the end, the editor wrote that the intention had been fully achieved; that while there was much emphasis on the need for further studies, a large number admitted, with or without qualifications, that simple inoculations or reinoculations of the Mitsuda antigen is capable of inducing positivity, while on the other hand there were some who denied that capability. The great majority have no doubt that BCG can cause conversion from negative to positive, and that it represents a valuable auxiliary in the prophylaxis of leprosy. —H. W. W.

Olmos Castro, M. and Arcuri, P. B. La reacción de Fernandez con lepromina proteica total (L.P.T.). [The Fernandez test with total protein lepromin (LPT).] Leprología **1** (1956) 143-156; *duplicated, ibid,* **2** (1957) 5-12.

This antigen is an extract of lepromas [now called "leprolin" by the authors] intended to elicit the early, or Fernandez reaction [see The Journal **26** (1958) 51-56]. Presumably healthy, noncontact persons gave a positivity rate of 3.6%, varying with age. Tuberculoid cases tested were 77.2% positive, lepromatous cases 0.5%. In healthy noncontacts previously sensitized with one or more intradermal injections of the Mitsuda-Hayashi lepromin, positivity was 75.2%. Six weeks after BCG vaccination normal persons were 46.8% positive when the vaccination was oral, and 81.2% when it was intradermal. In persons with progressive tuberculosis, Mantoux positive at 1:1,000, the positivity was 43.0%. These results show that LPT is a sufficiently sensitive antigen for eliciting various degrees of specific sensitization or cosensitization. It is yet to be determined how long it will retain its activity; the first lot made (September 1957) was still active 5 months later. A serious difficulty in the investigation of sensitization in leprosy and in the study of cosensitization is the use of integral or bacillary lepromins [regular lepromin or purified bacillus suspension] because of their high sensitizing capacity. Investigations made in man and the dog indicate that repeated injections of LPT cannot induce sensitivity of the tuberculin type. This indicates that LPT should be valuable for the study of cosensitization with BCG in pre- and postvaccination tests, and especially in testing for sensitivity in leprosy contacts since injection of the antigen can be repeated as many times as necessary without creating sensitivity.—[From the authors' abstract, supplied by G. Basombrio.]

Dozier, S. M., Fusillo, M. H. and Woodham, G. E. Demonstration of circulating antibodies to *M. tuberculosis* in humans. I. Technique. Theoretical considerations and development of a new concept of antibody assay as applied to tuberculosis. American Rev. Tuberc. & Pulmon. Dis. **75** (1957) 949-953.

The authors introduce a technique for the demonstration of circulating antibodies to *M. tuberculosis* in patients with active tuberculosis. This reaction is based on the theory that specific antibody globulin is extracted from serum by antigen-antibody reaction with a polyvalent tubercle-bacillus antigen. The adsorbed antibody is then assayed by a fall in titer of Coombs serum when human O Rh-positive cells sensitized with anti-D serum are used as the indicator system. The data indicate that the antibody globulin measured was related to the active disease state, but was unrelated to the results of the skin test. This method is regarded as probably applicable to bacterial or other diseases in which a particulate antigen can be prepared. —H. W. W.

Fusillo, M. H., Weiss, D. L. and Dozier, S. M. Demonstration of circulating antibodies to *M. tuberculosis* in humans. II. Clinical laboratory studies. The existence of specific circulating antibody in patients with active tuberculosis. American Rev. Tubere. & Pulmon. Dis. **75** (1957) 954-964.

This report deals with the experience with the new test (see above) on sera from considerable numbers of persons—normal, tuberculous and a few others. Of 74 normals, 3 gave "false positive" reactions (and they had recently had upper respiratory reactions, and when 2 were retested later their reactions were normal). Also negative were all of 7 arrested tuberculosis cases, whereas all but 2 of 80 active tuberculosis cases were positive (both of the 2 "apparently false negatives" were meningitis cases). The study confirms [it is said] the original observation that the circulating antibody tested is independent of the state of tissue hypersensitivity. The method is held to be readily adaptable to antigens made from other bacteria or fungi. [Nothing is said of tests for specificity with other mycobacteria, a point of interest in thinking of this test in connection with leprosy.] —H. W. W.

Seibert, F. B., Miller, E. E., Buseman, U., Seibert, M. V., Soto-Figueroa, E. and Fry, L. The significance of antibodies to tuberculoprotein and polysaccharide in resistance to tuberculosis. American Rev. Tubere. & Pulmon. Dis. **73** (1956) 547-562.

This is an article to which the reader must be referred because no ordinary abstract would do it justice, nor would it serve much better to copy the authors' summary. Special note will be made of one feature which is stressed in the discussion. For protection from infection it is held that a balance of antibodies must be present, and in rabbits vaccinated with BCG there was such a balance except in animals known to be susceptible, and in them the antibodies to polysaccharide were missing or deficient. Furthermore, the tuberculopolysaccharide itself is capable of interfering with antigen-antibody relationships, for addition of this substance caused *in vitro* inhibition of the complement-fixation reaction of BCG antiserum with PPD-S. It is speculated if this polysaccharide may not perhaps be capable of interfering with immune reactions. If so, the presence of free tuberculopolysaccharides in the body would be detrimental. Certain experiments gave suggestive results. [Have biochemists given thought to this factor in leprosy? In the lepromatous form, with its myriads of bacilli, the blood should contain a much higher titer of that bacterial product than the tuberculoid form.] —H. W. W.

England, N. J., Muir, J. M. and Reynard, W. A. Serial tuberculin tests in adults. Tubercle **38** (1957) 213-216.

This note tells of tuberculin "conversions" from negative to (mostly) 1 + observed in 40% of first-test negatives when retested a year later, and some of the repeated negatives were positive on the third test after another year. A total of 57 out of 459 men showed this change, none with any accompanying radiologic change. "Reversions" from positive to negative were relatively few. No explanation is definitely established. Similar experiences by other workers are referred to. —H. W. W.

MAGNUS, K. Effect of intradermal tuberculin tests on BCG-induced allergy. Bull. Wld. Hlth. Org. **17** (1957) 249-254.

Certain reports suggest that intradermal testing of school children with 10 TU of tuberculin at yearly intervals prevents the expected decline of the allergy induced by BCG vaccination. Pending the results of studies designed to detect this "boosting" effect in school children, the author has carried out experiments with guinea-pigs, which show that the decline of allergy can be prevented by intradermal injection of tuberculin, with either 5 or 10 TU but not with 1 TU. Too many injections, on the other hand, cause weakening of the reactions (desensitization). A question raised is whether the tuberculin-induced variations in the levels of allergy are accompanied by corresponding variations in immunity.—[From abstract in *WHO Chronicle* **12** (1958) 57.]

HOYT, A., KNOWLES, R. G., MOORE, F. J. AND SMITH, C. R. The adjuvant effect of *n*-hexadecane in mouse vaccination tests with heat-killed BCG. American Rev. Tuberc. & Pulmon. Dis. **75** (1957) 624-629.

Mineral oils and waxy materials being variable mixtures of uncertain composition, the authors used *n*-hexadecane (Eastman practical), a purified hydrocarbon. Heat-killed BCG, on intraperitoneal injection, gives protection to mice, but combinations of BCG and the hexadecane gave significantly more, having an adjuvant effect.—H. W. W.

HOYT, A., DENNERLINE, R. L., MOORE, F. J. AND SMITH, C. R. Tubercle bacillus wax as an experimental vaccine against mouse tuberculosis. American Rev. Tuberc. & Pulmon. Dis. **76** (1957) 752-760.

Purified tubercle bacillus wax has been proved to be an effective antigenic adjuvant (Raffel and others). The present study began as an investigation of enhancement of heat-killed BCG by tubercle-bacillus wax. Equal amounts of wax plus hexadecane, and BCG plus hexadecane, engendered similar resistance levels in mice, although probably by different mechanisms. The wax (an original Anderson product) was found to contain myriads of almost completely nonacid-fast mycobacteria which could be seen if treated only with alcohol after carbol-fuchsin staining. By high-speed centrifuging, the wax solution could be cleared of almost all of its bacilli, after which the wax (in combination with hexadecane) failed to give mice the protection that it did before removal of the bacilli. [Which simply shows that the purified wax is not antigenic; it is only supposed to be an effective adjuvant.] —H. W. W.

FENNER, F. Homologous and heterologous immunity in infections of mice with *Mycobacterium ulcerans* and *Mycobacterium balnei*. American Rev. Tuberc. & Pulmon. Dis. **76** (1957) 76-89.

Footpad inoculation with *M. balnei* and *M. ulcerans* was used to study homologous and heterologous immunity in mice infected with a variety of mycobacteria. The shorter incubation period in *M. balnei* infection made that one advantageous. Homologous immunity was best demonstrated with smaller dosage of the challenge inoculation. With large challenge inoculations there was some protection after an interval of three days between infection and challenge, but a tuberculin type of sensitization was not demonstrable until after 21 days. With small challenge inoculations a high degree of protection was evident after 4 days, and persisted unchanged for at least 30 weeks. Heterologous protection was demonstrated with both organisms. *M. ulcerans, M. balnei,* and BCG all induced a high degree of protection against small doses of *M. ulcerans*. Substantial protection against *M. balnei* was produced by *M. balnei, M. avium,* and BCG. Protection induced by *M. ulcerans* and *M. balnei* was associated with an accelerated reaction to the challenge inoculum, but mice immunized with BCG showed a high degree of protection without sensitization. Prior inoculation with *M. ranae* and *M. phlei* failed to immunize mice against *M. balnei*. Prior infection with *M. ulcerans* or *M. balnei* failed to prolong the survival time of mice challenged by the intravenous inoculation of

virulent tubercle bacilli, although both BCG and *M. avium* produced significant prolongation of the survival time.—[From author's summary.]

WHO Tuberculosis Research Office. Assessment of BCG vaccination in India; second report. Bull. Wld. Hlth. Org. **17** (1957) 203-224.

The last paragraph of the synopsis of this report reads as follows: "Testing of unvaccinated village populations in Madras and Mysore confirms previous observations that low-grade, non-specific tuberculin sensitivity is widely prevalent in South India, making it virtually impossible to separate the infected from the uninfected with the tuberculin tests in use today." Of the presumably uninfected persons (small reactions with 5 TU), nearly all over 15 years of age gave strong reactions to the 100 TU test. Of the 724 people in one village tested with 100 TU, all of the 61 persons 30 years and more of age gave reactions 10 mm. or larger. This disturbing situation, i.e., the ineffectiveness of the tuberculin test, has been observed in many other tropical areas—Burma, Indonesia, the Philippines, Thailand, Viet Nam, the Sudan, East Pakistan, Mauritius and Nigeria. It may perhaps be possible to fractionate tuberculin so as to separate the components that are specific for tuberculosis and those that are responsible for the nonspecific reactions, and studies of that matter are under way. —H. W. W.

Roelsgaard, E., Christensen, H. and Iversen, E. BCG-vaccination programme in Pakistan. Bull. Wld Hlth Org. **17** (1957) 187-202.

The following portion of this report is quoted, in connection with the subject matter of the preceding abstract. "The following conclusions can be drawn . . . . (*a*) In West Pakistan testing with 5 TU provides a fairly efficient separation of infected and non-infected individuals, and thus practically all non-infected persons are vaccinated and the percentage of positive reactors can be used as a fairly accurate measure of the prevalence of tuberculous infection. (*b*) In East Bengal the tuberculin test gives a much less satisfactory separation; owing to the occurrence of non-specific sensitivity many non-infected persons are classified as positive. As a result, far from all non-infected persons are vaccinated and the percentage of positives gives an exaggerated measure of the frequency of tuberculous infection." —H. W. W.

McMillen, S. and Kushner, D. S. Atypical acid-fast bacilli. I. A. cultural scheme for rapid identification. American Rev. Tubere. & Pulmon. Dis. **76** (1957) 103-107.

Kushner, D. S., McMillen, S. and Senderi, M. Atypical acid-fast bacilli. II. *Mycobacterium fortuitum:* Bacteriologic characteristics and pathogenicity for laboratory animals. *Ibid.* 108-122.

[The interested reader is referred to the original articles.]

Nishimura, S. The significance of experimental murine leprosy and screening test in studies on chemotherapeutic agents for leprosy. Med. J. Osaka Univ. **7** (1957) 753-776.

Reviewing the evidence for using *M. leprae murium* as a test organism for forecasting the activity of drugs for human leprosy, the author concludes that its use for this purpose is a mistake. Nevertheless, the experiments are useful, and the author advocates his own method of using murine leprosy for screening drugs as the best. Little is known of the bacteriology of the human and murine bacilli, especially as regards their growth metabolism. Regarding their pathology, the two diseases have some similarities, but also some important differences; e.g., *M. leprae* multiplies in both mesodermal and ectodermal cells, whereas *M. leprae murium* proliferates only in the mesenchymal cells of the mesoderm. Of the drugs used in murine leprosy, only isoniazid and chaulmoogra oil have consistently shown activity. Considering the activities of all the drugs for this disease, human leprosy, and tuberculosis, only those which have weak activity against the tubercle bacillus, except TB-1 (thioacetazone), are active in human leprosy, and none of them is active in murine leprosy; thioacetazone is active in tuberculosis and

human leprosy, but not in murine leprosy. The fact that more drugs are active against the tubercle bacillus than the leprosy bacillus suggests that the metabolic processes of the tubercle bacillus are the more complex.—[From abstract in *Trop. Dis. Bull.* **55** (1958) 277.]

NAKAGAWA, Y. AND NAKAMURA, M. Preservation of *Myc. lepraemurium* by means of lyophilization. II Report. Kurume Med. J. **4** (1957) 69-74.

*M. leprae murium* was lyophilized in saline, serum water, Kirchner's medium, and glycerine water and stored in a refrigerator for about five and one-half years. On testing by inoculation into normal white rats, it was found that infectivity had been maintained in all the solutions except the glycerine water. [See THE JOURNAL **23** (1955) 485.]—[From authors' summary in *Trop. Dis. Bull.* **55** (1958) 277.]

KAWAGUCHI, Y. Strains of mice for experimental murine leprosy. Part 1. Susceptibility of various uniform strains of mice to murine leprosy bacilli. La Lepro **26** (1957) 318-324 (in Japanese; English abstract).

The differences in susceptibility of six strains of mice (C57BL, CF1, CFW, C3H, dd, and A) to subcutaneous inoculation with murine leprosy bacilli were studied. Under similar conditions, mice of the C57BL strain showed the earliest but smallest lesions. Lesions of the C3H strain developed later but were larger than all others. These two strains presented a striking contrast. The other four strains were intermediate. The susceptibility of the C57BL strain to ulcer formation was much greater than that of the others, and the dd strain ranked second in severity.—[From the English abstract.]

ITO, T. AND SONODA, R. Biochemical investigations of the murine leprosy bacillus. (Part 3) Enzyme activity of *M. leprae murium* upon asparagin and asparaginic acid and adaptation to several organic acids. La Lepro **26** (1957) 311-314 (in Japanese; English abstract).

By the enzyme activity of *M. leprae murium* asparagin is converted to asparaginic acid, with freeing of ammonia. It was also found, by the Friedman-Haugen method, that transamination takes place between asparaginic acid and $\alpha$-ketoglutaric acid to give oxalacetic acid. Attempts were made to adapt *M. lepre murium* to several organic acids, but results suggestive of production or existence of adaptive enzymes were not obtained.—[From the English abstract.]

HIRANO, N. AND SUSHIDA, K. The chemotherapy of murine leprosy. 3. The action of isonicotinoyl-3-methoxy-4-ethoxybenzal hydrazone and isonicotinoyl-3, 4-diethoxybenzal hydrazone. La Lepro **26** (1957) 315-317 (in Japanese; English abstract).

In a previous report, it was shown that the action against murine leprosy of isonicotinoyl-3, 4-dimethoxybenzal hydrazone (No. 282) was greater than that of isonicotinoyl-3-methoxy-4-hydroxybenzal hydrazone (No. 254). In the present experiment was studied the action against murine leprosy of isonicotinoyl-3-methoxy-4-ethoxylbenzal hydrazone (No. 283), and isonicotinoyl-3, 4-diethoxybenzal hydrazone (No. 284), in which the methoxy radical has been replaced by the ethoxy radical. The toxicity of these two compounds is very low, and doses of 4.5 gm./kgm. orally are well tolerated by mice. Studies were also conducted with INH, No. 254 and No. 282 at the same time. It was found that acid-fast bacilli were present after treatment for 3 months with INH, No. 254, No. 282 and No. 283, but could not be found in 3 out of 6 mice given No. 284. Leproma formation was detected in the remaining 3 animals, but only small numbers of acid-fast bacilli could be found. If a reduction or disappearance of acid-fast bacilli should take place when No. 284 is administered in mice in which severe leprotic changes are already present, this agent may be useful in the treatment of human leprosy, but further study is required.—[From the English abstract.]

Ito, T. and Sonada, R. Biochemical studies on murine leprosy bacilli. IV. (1) Effect of testicular extract on the respiration of murine leprosy bacilli. (2) Catalase activity of murine leprosy bacilli. La Lepro **27** (1958) 136-138 (in Japanese; English abstract).

Extracts were prepared from the testicular leproma of rats inoculated with murine leprosy and of tests of normal rats. After dialysis and acetone treatment the effect on the respiration of murine leprosy bacilli was examined. It was found that the dialysis removed a respiration-accelerating substance from the extract, and treatment with acetone showed that the accelerating substance was contained in the acetone-soluble portion. Studies with the Warburg monometer showed that murine leprosy bacilli possess a catalase activity.—[From abstract.]

Shepard, C. C. Growth characteristics of tubercle bacilli and certain other mycobacteria in HeLa cells. J. Exper. Med. **105** (1957) 39-48.

By making use of the increased phagocytosis which follows the exposure of HeLa cells to tissue culture media containing selected horse sera, it was possible to introduce all of the mycobacterial species studied into the cells, where many of them proceeded to grow. Fully virulent strains of tubercle bacilli filled much of the cytoplasm in a few days and formed characteristic cords not seen with other strains. The strains said to be less virulent, R1Rv, BCG, H37Ra, and R1Ra, grew less rapidly and in characteristic patterns. Their rates of multiplication in HeLa cells were in the order named and correlated well with their reported pathogenicity for mice and guinea pigs. Six INH-resistant strains grew at rates characteristic of fully virulent strains. Among the "rapidly growing" species, *M. phlei* and *M. smegmatis* did not show evidence of growth in the cells, although *M. fortuitum* did. Some strains with optimal temperatures on bacteriological media below 37°C, *M. balnei, M. marinum,* and *M. platypoecilus,* grew rapidly in HeLa cells, especially at temperatures of 31 to 35°C. The growth patterns of the bacilli in HeLa cells appear sufficiently specific to be useful in differentiation among the mycobacteria.—[Author's summary.]

Shepard, C. C. A comparison of the growth of selected mycobacteria HeLa, monkey kidney, and human amnion cells in tissue culture. J. Exper. Med. **107** (1958) 237-246.

The tissue cultures mentioned were compared as sites for the growth of tubercle bacilli and certain other pathogenic mycobacteria, including leprosy bacilli. These last were obtained from lepromas by a process to be described later, which involves centrifugal washing in solutions of bovine albumin. There was satisfactory ingestion of bacilli. HeLe cell cultures could be kept for 1-2 months, although they become overcrowded with cells; the human amnion and monkey kidney cultures, which did no become overcrowded, were kept for 6 months and more while still containing bacilli. Definite evidence of growth was not observed.—H. W. W.

Lack, C. H., Newman, B. J., Tanner, F. A. and Towers, A. G. Atypical acid-alcohol-fast bacilli cultured from human urines. J. Clin. Path. **10** (1957) 204-207.

An acid-alcohol-fast bacillus, 1 to 3 $\mu$ in length, was found in urines from 45 patients and has the following characteristics: In liquid media (Glover's blood) it grows in amorphous masses; on glycerol-containing solid media it forms a smooth, moist buff-colored colony, grows well at room temperature and survives at 60°C. for four hours; it can survive for at least seven weeks in mice but is nonpathogenic for guinea pigs. Lack of pigment, inability to ferment rhamnose and arabinose, plus other features distinguish it from *M. smegmatis* and *M. phlei.* Colonially this bacillus appears to be more like an avian bacillus or *M. fortuitum,* but its growth at 22°C. distinguishes it from the former and it is shorter and more uniform in size than the latter. It is resistant to most antimicrobials and could be taken for a drug-resistant tubercle bacillus. It differs from

isoniazid-resistant tubercle bacilli in that it combines catalase production with non pathogenicity for guinea pigs. Being able to grow at 22°C. on glycerol-containing media is the important characteristic that distinguishes it from drug-resistant variants of *M. tuberculosis*.—[Abstract from *American Rev. Tuberc. & Pulmon. Dis.* **77** (1958) 118-119.]

DUNBAR, J.-M. L'apparition de formes non acido-résistantes de *Mycobacterium tuberculosis* en présence d'isoniazide, de cyclosérine et du thioamide de l'acide α-éthyl iso-nicotique. [The appearance of nonacid-fast forms of *M. tuberculosis* in the presence of isoniazid, cycloserine and thiamide of α ethyl-isonicotinic acid.] Ann. Inst. Pasteur **92** (1957) 451-457.

The H37Rv strain of *Mycobacterium tuberculosis* when grown in a suitable concentration of α-ethyl-thioisonicotinamide (2.0 γ per mil.) loses the property of acid-fastness in the Ziehl-Neelsen staining technique and is demostrable only by the use of a counterstain. Such nonacid-fast forms present a similar morphology to diphtheria group organisms. These nonacid-fast forms are readily demonstable by simple Gram and Giemsa staining procedures. The presence of streptomycin (5 γ per ml.) prevents this loss of acid-fastness associated with the presence of α-ethyl-thioisonicotinamide (2.0 γ per ml.) while PAS (5 γ per ml.) fails to do so. These facts show that α-ethyl-thioisonicotinamide, if not working by the same mechanism, produces the same phenomena as isoniazid. Liquid culture growths using Youmans' medium with 10 per cent bovine serum in the presence of cycloserine 50 γ per ml. yields large numbers of "ghost cords" with only a few bacillary outlines discernible within the serpentine "cord" boundaries. The phenomenon is not modified by the presence of streptomycin. The staining reactions of "resting bacilli" remain unchanged in the presence of isoniazid, thioisonicotinamide and cycloserine.—[Author's summary, from *American Rev. Tuberc. & Pulmon. Dis.* **77** (1958) 119.]

GUNDERS, A. E. Progressive experimental infection with *Mycobacterium leprae* in a chimpanzee; a preliminary report. J. Trop. Med. & Hyg. **61** (1958) 228-230.

A young female chimpanzee 6-7 months old was inoculated with a leproma suspension, partly into (or under?) the temporal bone, partly around an ulnar nerve, and partly intravenously. After 11 months there was evidence of active infection, in multiple (100 or more) intracutaneous nodules ranging in size from small peas to hazel nuts located on the ears, dorsa of hands and feet, and forearms and legs, and also small areas of depigmentation in various locations. Biopsy of a skin nodule showed two types of cells predominating, one of them epithelioid (without giant cells), and bacilli lying singly or in "cigarette-bunch" formation [shown in small numbers in photomicrographs reproduced]. Three months later there was a general tendency to regression; some of the nodules had subsided, leaving areas of depigmentation, and others were smaller than before. A second, older, chimpanzee which was also inoculated, but in different ways, had shown no sign of infection.—H. W. W.

KELKAR, S. and RANADIVE, K. J. Biological factors in transmission of human leprosy to laboratory animals. I. Indian J. Med. Sci. **12** (1958) 873-883. ———— and ———— Studies on transmission of human leprosy to laboratory animals. II. Ibid. 884-891.

After considering various factors which would favor infection, the authors applied them in their experiments. Because children are particularly susceptible, they used mainly newborn animals (75 rats, 36 hamsters, and 24 mice), and because repeated contact favors infection they were given multiple inoculations. The general body resistance was lowered by means of cortisone, hyaluronidase, a diet deficient in vitamin $B_1$, sensitization with lepromin, and exposure to x-rays. The inoculations were with lepromatous nodule implanted subcutaneously, and a suspension of bacilli given intramuscularly or intraperitoneally. Young adult rats thus inoculated served as controls. Both of the rat

groups were observed for 8 to 18 months. The hamsters, 7 of which served as controls, were treated with varying doses of x-ray. Both groups were observed for six months and killed when the animals looked emaciated. *Results:* Rats: Of the 75 rats, 42 showed acid-fast granules in macrophages with diffuse pink cytoplasm, and a nonspecific inflammatory cellular reaction. The few organisms seen were in and around nerves. The control rats and those treated with cortisone and the vitamin $B_1$-deficient diet did not show any significant difference in their tissue reaction to repeated inoculation. The reaction was more pronounced, however, and more animals were affected, when cortisone and a vitamin $B_1$-deficiency diet was given prior to inoculation. The intraperitoneal inoculation was found to be the most effective route. Mice: There was a complete absence of tissue reaction in the mice treated with hyaluronidase or lepromin prior to inoculation. Hamsters: There was evidence of survival and even proliferation of acid-fast organisms in over 50% of those in the irradiated group, but not in those treated with cortisone or the controls. In young hamsters showing evidence of disease, the bacilli—after disintegration of the transplanted nodule—were liberated into the subcutaneous tissue, some to be ingested by macrophages and others to be transported to regional axillary lymph nodes. The older hamsters so treated showed marked tissue damage, the tissue reactions suggesting a progressive generalized infection. A dose of 100 r whole body irradiation given to young adult hamsters prior to inoculation thus proved to be a means of lowering of the body resistance of those animals.—[From Foreign Letters, *J. American Med. Assoc.* 169 (1959) 273; supplied by Sr. Hilary Ross.]

HAWKING, F. Growth of rat leprosy bacillus in tissue culture. Leprosy Rev. **29** (1958) 71-80.

Glass slips were embedded in a thin layer of fowl plasma clotted by embryo extract on the bottom of a Carrel flask, and small pieces of rat leproma were mounted on these slips, after which growth medium was added. At intervals one of the coverslips was removed and examined for evidence of growth of the *M. leprae murium*. The total number of bacilli in the culture was estimated by the method of Sandford (details given). The results of some early experiments with small pieces from the edge of the rat leproma to which were added pieces of normal spleen or liver of young rats showed that it was difficult by this technique to estimate the number of bacilli at the beginning of the experiment in order to compare it with the number present at a later date. The later experiments, consequently, were done with macrophages infected with the bacilli *in vivo*. The macrophages were produced by intraperitoneal injection of liquid paraffin into rats, a leproma suspension being injected intraperitoneally 3 days before the cells were aspirated. These cells became degenerate after about 4-5 weeks' incubation in the tissue culture, and in some experiments they were replaced with fresh uninfected cells; in 1 experiment the culture was maintained for 4 months by this method. Although there was an inevitable loss of bacilli through changing the medium, from the number present at the end of the experiment (52 days), the author judged that they had doubled their numbers. Bacilli that were present after 42 days were still infective to rats. The author holds that this type of culture can be maintained indefinitely, and that the bacilli pass readily from one crop of cells to the next in spite of the presence of 40% serum in the medium. Thick fibrin around the cells encourages the growth of the bacilli, and an extract of lepromatous tissue or mycobactin probably promotes multiplication of the bacilli; penicillin 100 units per cc. does not seem to interfere with growth.—[From abstract in *Trop. Dis. Bull.* **55** (1958) 901.]

GARBUTT, E. W., REES, R. J. W. and BARR, Y. M. Multiplication of rat-leprosy bacilli in cultures of rat fibroblasts. Lancet **2** (1958) 127-128.

Recently there have been reports of significant but limited growths of *M. leprae murium* in tissue cultures of rat spleen cells and in monocytes. The authors report having grown this organism in rat fibroblast cells without hydrocortisone, thus contrasting with the results of Wallace *et al.* [THE JOURNAL **26** (1958) 292] who detected growth

only when sufficient hydrocortisone was present to damage the cells. The 14pf strain of fibroblasts was used, grown in 50% human cord serum plus 50% Hanks' balanced salt solution for 7-10 days. During the time of infection with the bacilli, which were derived from livers of mice inoculated intravenously, the cells were in the salt solution containing 0.0125% albumin, and ingestion of the bacilli was allowed to take place for 24 hours, during which period 90% of the cells ingested from 1 to more than 10 bacilli each. The cells were then trypsinized, washed in the salt solution to remove extracellular bacilli, and resuspended in the growth medium. Total bacterial counts were made after 13, 26 and 39 days' incubation, the cells having been disintegrated in an ultrasonic vibrator. In 8 of the 9 experiments reported there was significant increase in the number of bacilli, ranging from 2.0- to 5.3-fold. In most of these experiments no increase occurred after 13 days' incubation, but in 3 the number present at 39 days was greater than at 26 days, the increases being 3.1-, 4.4- and 5.5-fold. No increase was observed in the presence of streptomycin and isoniazid. Hydrocortisone, 0.05 and 0.1 mgm. per cc., had no effect on the multiplication.—[From abstract in *Trop. Dis. Bull.* **55** (1958) 1339.]

KAWAGUCHI, Y. Strains of mice for experimental murine leprosy. Part 2. The relation between strains of mice and effects of BCG in murine leprosy. Le Lepro **27** (1958) 44-48 (in Japanese; English abstract.)

For the purpose of selecting a mouse strain suitable for immunology experiments, comparative observations were made of the effects of BCG vaccination on the onset of murine leprosy in various uniform strains of mice (C57BL, dd, CFW, CF1, and C3H). The animals used were of approximately the same age, and lived under the same conditions of diet and environment. Two strains, C3H and CF1, were found suitable, in that the lepromas developed to a much larger size whereas ulcers developed more slowly. In strains in which the lepromas generally remained small and ulcerated early, the differences between the vaccinated and unvaccinated mice were less marked, and they were deemed unsuitable for the study of immune responses.—[From the English abstract.]

KAWAGUCHI, Y. Strains of mice for experimental murine leprosy. Part 3. Strains of mice for experimental chemotherapy of murine leprosy. La Lepro **27** (1958) 49-53 (in Japanese; English abstract).

Mice of different strains (C57BL, dd, CFW, CF1 and C3H), inoculated subcutaneously with murine leprosy bacilli, were watched under treatment with isoniazid and pyrazinamide to select a strain suitable for the determination of efficacy of therapeutic agents. The experiments were carried out in two series. (1) Inhibition of onset. No significant difference was recognized among the mouse strains tested. A strain, such as C57BL, with a tendency toward earlier leproma formation was therefore deemed suitable for leproma inhibition experiments. (2) Therapy: Only a slight difference was found between the treated and untreated mice of the C57BL strain, whereas a remarkable one was found in the C3H strain. The latter, therefore, was the better for therapy experiments. —[From the English abstract.]