# THE REACTIONS TO LEPROMIN IN MAN
## I. A SUGGESTION OF POSSIBLE DESENSITIZATION OF THE EARLY REACTION

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Recognition of the "early" or Fernandez reaction to lepromin has complicated the problem of the nature and mechanism of the response to that antigen. No explanation of the "late" or Mitsuda phenomenon has proved acceptable to all investigators, and now there is the question whether the early one is due to the same immunological condition or a different one. With regard to the practical application of the test, apart from theoretical considerations, it has been hoped that the early reaction would simplify the matter—that the results of the test could be determined, especially if a practicable filtrate or protein bacillus extract could be developed, in 48 hours as with the tuberculin reaction. Although there are those who apparently believe that has been accomplished with less refined antigens, that view is open to question.

Passing over the more or less incidental references of the earlier workers to an early erythematous reaction, the first particular observations of this phase were by Rodriguez (7), although it remained for Fernandez (1) to establish its significance. Rodriguez suggested that it is due to a protein factor of the leprosy bacillus, because clinically and histologically it resembles the response to injections of tuberculin, and that the late response is produced by some other substance or substances, possibly derived in part from the bacillary lipids. Fernandez, likewise, ascribed the two phases of the reaction to two antigenic elements, but without the same specification. One is the soluble element, since a filtrate of the lepromin causes little if anything more than the early reaction—as Hayashi (4) had shown—and the other an insoluble one, not dissociable from the bacillary bodies, the effect of which appears after a longer period.

Lowe and Dharmendra (6) disagreed with this view because they had found—in observations allied to those of Kitano and Inoue (5)—that the more thoroughly the bacilli are broken down the stronger is the early reaction and the less marked the late one, and that the extracted proteins give only the former. They

concluded that both early and late phases are manifestations of the same preexisting condition of allergic sensitization, the former induced by the dissolved proteins, the latter by those which are slowly released as the injected bacilli are broken down. Later, Fernandez (2, 3) concluded that the early reaction is due to preexisting sensitization of the tuberculin type, while for the late one he accepted the hypothesis of Wade (8, 9, 10) that it is due to an allergic condition induced by the injection of the antigen. The matter still needs clarification, and every possible approach should be explored.

The present report concerns observations made after a healthy individual had been given simultaneous injections of four different lots of lepromin. There were rather striking differences in the early reactions to the different lots and, to see if these differences would persist, reinjections were made twice at two-day intervals. The results are reported without further observations for what they may be worth, since there is no opportunity to pursue this particular line of inquiry in this institution.

## MATERIALS AND METHOD

In order to make a large, pooled batch of lepromin, specimens of lepromatous tissues were secured from three sources, and from one of them a control sample of lepromin actually in use. (Details will be given in the next report of this series.) Before pooling the three preparations made from these tissues they were tested separately against the control (antigen No. 4). Because four injections were to be made, and because multiple intracutaneous injections of small doses of an antigen are likely to sensitize more actively than single large doses while multiple large doses might induce excessive reactions, the preparations were all diluted to one-fifth strength with 0.5 per cent carbol-saline. These dilutions were administered in the usual 0.1 cc. dose.

A second set of injections was given, for the reason stated, after making the 48-hour readings of the first set, and because the responses to it after 48 hours were decidedly different a third set of injections was made at that time. The first injections were made on the antero-medial surface of the forearms, three on the left and the fourth (together with two unrelated antigens) on the right. Those areas having been so used, the reinjections of present interest were made on the anterior surface of the upper arms, in the same arrangement. From other experience no reason is seen to believe that the differences of location could have had any influence on the results.

The primary records of the early reaction were of the extent of the erythema. Some degree of edematous infiltration was always present, but at times it was so diffuse that a separate measurement of it could not be made with accuracy.

## RESULTS

The apparent effects of reinjection are to be seen only in a comparison of the primary injections with the first reinjections, at the respective 48-hour readings. The individual concerned was a fairly quick reactor, although not a strong one with respect to the late reaction, so that by the time the second reinjections were 48 hours old the primary sites—then 6 days old—had begun to show the onset of the late effect. That condition seemed to have influenced the response to the last reinjections, with increased erythema and more infiltration, and consequently they are not considered.

The recorded observations of the two phases of the reactions are shown in Table 1. The early ones are of the 48-hour period, as specified by Fernandez and others. Save in one instance the measurements were smaller then than at 24 hours, with however a greater distinction between the first and second sets of injections. In the sole exception—the first injection of the No. 3 antigen—the overall dimension had increased from 26 to 31 mm. during the second day, although there was a diminution of intensity of the erythema, suggesting that the maximum had occurred at some time in the interval.

TABLE 1.—*Comparing the effects of injections of four lots of lepromin and reinjections made after 48 hours.*

| Antigen | Injection | Date of injection | Early reaction | | Late reaction | | |
|---|---|---|---|---|---|---|---|
| | | | Erythema *a* | Infiltration *b* | Time of maximum | Erythema | Induration |
| #1 | 1st | 11/5 | Sl 13 | Slight (—) | 2 wks | Mk 6/9 | Mk 13 |
| | 2nd | 11/7 | Ft ± 11 | Trace *c* | 3 wks | Md 7 | Md 6.5 |
| #2 | 1st | 11/5 | Sl 13 | Slight (—) | 2 wks | Mk 7/11 | Mk 11 |
| | 2nd | 11/7 | Ft ± 11 | Trace *c* | 3 wks | Md 8 | Md 9 |
| #3 | 1st | 11/5 | Mk 12/31 | Md 12 | 2 wks | Md 9 | Md 10 |
| | 2nd | 11/7 | Mk 7/16 | Trace *c* | 3 wks | Md 6 | Md 6.5 |
| #4 | 1st | 11/5 | Mk 10/20 | Md 10 | 2 wks | Md 5 | Sl 6.5 |
| | 2nd | 11/7 | Mk 8/20 | Trace *c* | 3 wks | Md 7 | Md 6 |

*a* When two measurements are given the first refers to the extent of central erythema of the degree indicated, the second to the over-all measurement of the outer zone, usually fading off toward the edge.

*b* The sign (—) signifies no measurement, the condition not sufficiently well defined.

*c* Recorded as "negligible."

Conspicuous in the records of the first injections is the fact that the Nos. 3 and 4 antigens, both derived from the same source, caused more marked early responses than did the other two.[1] This difference also appeared with the reinjections, with respect only to the erythema. In all aspects the reinjections caused less response than the first ones, the differences with antigens Nos. 1 and 2 being more striking to the eye than could be indicated by the recorded data.

With regard to the late reactions, none of which ulcerated, those induced by the first injections had reached their maximum at the end of two weeks. Those of the reinjections were somewhat later, although in some instances the differences between the two-week and three-week readings were slight. With each antigen the late lesion induced by the first injection was the larger. In two instances the differences were too slight to be regarded as significant, but in the other two it was decidedly greater. Furthermore, the lesions of the reinjections resolved somewhat less slowly than did those of the original injections despite the somewhat later maxima. It may be added that, whereas the early reactions to the third injections were generally more marked than those of the second, the late lesions induced by these two sets were virtually identical.

A later observation is of some interest. About three months after these injections were made, when the late lesions seemed to have quite subsided and become residual, a series of reinjections with single doses of a one-third strength lepromin was begun, these injections being made at intervals of two weeks. At the time of the second of them the sites of the original tests showed reactivation. First they became somewhat brighter red and more shiny of surface than before, and definitely sensitive to friction. Shortly, without significant increase of induration, sites Nos. 1 and 2 underwent shallow ulceration and presented tiny crusts barely 2 mm. in diameter, and the No. 3 site soon had a 1 mm. spot of erosin. The site of the No. 4 antigen—the least concentrated of all—showed less change. The sites of the reinjections showed little evidence of reactivation, nothing more than slight desquamation of the Nos. 1 and 2 sites.

## DISCUSSION

The indications here seen that reinjection of lepromin as done

[1] This difference was observed regularly in subsequent work with both patients and other controls, and will be considered later with the collaboration of Dr. J. O. Nolasco, chief pathologist of this colony.

in this instance—in divided doses, multiple sites, after a 48-hour interval—tends to induce less reaction than the primary injections, at the early stage if not also later, can be regarded as no more than a suggestion that it may be possible to effect some degree of desensitization of a preexisting allergic condition. Such observations, in a single case, justify no conclusion whatever; they are recorded only because of the interest that there is in the possibility suggested.

It is well established that tuberculous animals can be desensitized with respect to tuberculin without losing the immunity induced by the sensitizing infection; it is not known if any investigation has been made of skin reactivity to injected tubercle bacilli in animals so desensitized. In connection with the problem of the essential nature of the two phases of the lepromin reaction, it would be of the greatest interest to know if desensitization with respect to the early phase can really be effected, and if so whether the late response is also materially changed.

That there may have been some effect on the late phase in this instance is suggested by the somewhat slower attainment of the maximum, the smaller average size of the resultant lesions at their largest, their quicker resolution, and finally, their virtual nonreactivation when the primary sites had "flared up" and most of them had ulcerated—as they had not done at first. Unless this series of circumstances was wholly fortuitous and without significance, it suggests the possibility that lessening of the early reaction results in the diffusion of a greater part of the injected antigen, or in other words less fixation of antigen at the site of inoculation. This idea, also, can only be regarded as a suggestion.

A conclusive desensitization experiment could obviously be made only on human volunteers, and several of them would be necessary because some might show quite different results from others. For the desensitizing injections whole lepromin would be entirely unsuitable, particularly because the late effects might interfere as they seemed to in the present instance. Active lepromin filtrates or extracted soluble proteins would be used hoping that, like tuberculin, they would not themselves be allergenic. The material would be administered daily, and obviously in relatively large doses—one regular dose into the skin to observe the reaction induced, and the rest subcutaneously in a different region. Whole lepromin, presumably, should be used only for a final test for the late reaction—which, of course, would then be without a comparative test unless that could be done at such a time that

it would not affect the experiment or be affected by it. Whether or not such an experiment would be feasible anywhere one cannot venture to say.

SUMMARY

In a preliminary test of four lepromin preparations in a single individual it was observed that two of them gave much stronger early reactions than the others.

To ascertain if the differences would persist, duplicate injections were made after the 48-hour readings had been made. A third set administered after another 48 hours is not considered, because the results were apparently affected by onset of the late reaction to the first one.

The reinjections caused materially less early reaction than did the original injections—the difference more distinct after 48 hours than after 24 hours—suggesting that a certain amount of desensitization had occurred. The late reactions also showed differences, in several respects.

No conclusions being possible from a single observation of this kind, this report is offered in the hope that others may be in a position to pursue the matter. The lines along which such an investigation might be made are suggested.

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