

CORRELATION BETWEEN THE REACTIONS TO LEPROLIN
AND LEPROMIN IN PERSONS HYPERSENSITIVE TO
*M. LEPRAE*¹

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In connection with the use of our total protein antigen, prepared from lepromas for the investigation of hypersensitivity in leprosy (⁸), and in accord with the suggestion of Wade (¹³), we classify the leprosy antigens, according to their antigenic function, in two categories. These are: (1) *Leprolins*, antigens which reveal the state of protein hypersensitivity of tuberculin type, but which are incapable themselves of provoking such a state of altered reactivity in sensitive organisms; they do not contain bacillary bodies, only their antigenic derivatives, and they act as haptens. (2) *Lepromins*, antigens which have the property of provoking states of altered reactivity (hypersensitivity) to *M. leprae* itself (bacillus-body hypersensitivity) or to its protein derivatives (protein hypersensitivity). They are composed of bacillary bodies, and act as complete antigens. They are capable of revealing hypersensitivity when injected intradermally in sensitized organisms. The intradermal reactions provoked by these two types of antigens are called the leprolin reaction and the lepromin reaction, respectively (^{9, 13}).

The positive leprolin reaction is characterized by a 48-hour reaction of the tuberculin type, and it signifies protein hypersensitivity of that type. The positive lepromin reaction is characterized by an accelerated formation of the tubercle by the end of the first week, and it signifies hypersensitivity to the bacillary body. Generally it is preceded by the tuberculin type of reaction (the early, or Fernandez reaction). The late nodular reaction (the Mitsuda reaction), interpreted as a test of immunity, is nothing but the terminal evolution of the accelerated formation of the tubercle (^{9, 10}).

A problem of interest posed by the intradermal tests with leprosy antigens is to establish whether the positive leprolin reaction indicates a certain degree of resistance to infection, as it does in tuberculosis. Also there is the interesting question of whether or not the intradermal injection of lepromins in hypersensitive persons is capable of increasing their degree of protein hypersensitivity.

PLAN OF STUDY

Antigens used.—(1) The leprolin test was performed with total protein leprolin (⁸).

¹Translated from the Spanish.

²Dr. Olmos Castro died on December 31, 1962.—EDITOR.

The results are obtained by measuring both of the greater diameters of the reaction and taking the average in millimeters, and by plus marks for the early Fernandez reaction, according to the suggestion of the Committee on Immunology of the Tokyo Congress ⁽¹⁾.

(2) The lepromin test was performed with the Mitsuda-Hayashi type lepromin. The Fernandez reaction, the accelerated formation of the nodule, and the Mitsuda reaction are recorded by noting the average in millimeters of their greater diameters.

Subjects utilized.—The experiment was carried out on a group of 23 persons taken at random who were leprolin positive: 19 contacts and 4 tuberculoids.

Method.—To the individuals known to be leprolin positive, 0.1 cc. of lepromin was injected intradermally in the right scapular area. In the 21st day after the injection of the lepromin, the leprolin test was repeated, and—with about one-half of the group—it was repeated again after 6 weeks.

RESULTS

The results obtained in the 23 cases tested are shown in detail in Table 1.

TABLE 1.—Results of the leprolin test performed before the lepromin test, and 21 days and 6 weeks afterward; also the results of the lepromin test.

Case No. and kind ^a	First leprolin test		Lepromin test (reactions, mm.)			Leprolin tests after lepromin test			
						21 days		6 weeks	
	mm.	+	Early ^b	Accel. ^c	Late ^d	mm.	+	mm.	+
1 C	15	2+	18	5	6 U ^e	20.5	3+		
2 C	12.5	1+	27.5	5	9 U	22.5	3+		
3 C	10	1+	35	7	10 U	17.5	2+		
4 C	17.5	2+	27.5	9	15 U	50	3+		
5 C	20	3+	30	10	15 U	75	3+		
6 C	52.5	3+	25	7 U ^e	10 U	22.5	3+	75	3+
7 C	15	2+	14	5.5	7	20	3+	47.5	3+
8 C	13.5	1+	30	14 U	17.5 U	70GR ^f	3+		
9 C	15	2+	30	7	11 U	60GR ^f	3+		
10 C	10	1+	20	9	11	10	1+	27.5	3+
11 C	25	3+	37.5	15	30 U	27.5	3+	56.5	3+
12 C	20	3+	30	10	12 U	10	1+	25	3+
13 C	15	2+	25	10	12	13	1+	50	3+
14 C	15	2+	15	10	15 U	15	2+	46	3+
15 C	15	2+	15	8	9 U	11	1+	47.5	3+
16 C	11	1+	25	8	10 U	37.5	3+	50	3+
17 C	10	1+	20	8	10	10	1+	55	3+
18 C	20	2+	37.5	10 U	12 U	22.5	3+	45	3+
19 C	8	1+	17.5	10	13	50	3+		
20 T	23.5	3+	27.5	7	8	15	2+	25	3+
21 T	25	3+	25	5.5	7 U	13.5	1+	20	3+
22 T	17	2+	20	15 U	13 U	15	2+		
23 T	28.5	3+	13.5	8	10	60	3+		

^aC = contacts; T = tuberculoids. ^bEarly or Fernandez reaction. ^cAccelerated formation of the tubercle. ^dLate or Mitsuda reaction. ^eU = ulceration. ^fGR = generalized reaction induced by leprolin after lepromin.

The results show a high concordance of positivity between the leprolin and the lepromin reactions.

The leprolin tests made 21 days after the lepromin tests (i.e., at the time the Mitsuda reactions were read), showed a frank increase in the intensity of the reactions.

In most of the 13 cases recorded as having been tested with leprolin 6 weeks³ after the lepromin testing, the early reactions had increased greatly. For example, in the first recorded case in the list the increase was from 22.5 mm. to 75 mm., and in the second case from 20 mm. to 47.5 mm. The average was from 17.5 mm. to 43.8 mm.

DISCUSSION

1. For years we have insisted that the two most important facts in the immunology of leprosy are (1) the experiments carried out in the dog by Wade (^{11,12}) which demonstrated the sensitizing capacity of *M. leprae*, and (2) the experiments of Fernandez (^{3,4}) which established the existence of the tuberculin-type reaction of hypersensitivity. Nevertheless, leprologists have shown little interest in extending these investigations, which may significantly contribute not only to the knowledge of the states of altered reactivity provoked by *M. leprae*, but also of the pathogenesis of leprosy.

It has been established that *M. leprae*, in sensitive organisms, is capable of provoking a state of altered reactivity to the bacillary bodies or to their protein derivatives.

Hypersensitivity to the bacillus body (corporeal hypersensitivity) can be investigated by means of intradermal injection of heat-killed *M. leprae* (lepromins), and it is manifested by a nodular reaction at the end of the first week, a reaction which we call "accelerated formation of the tubercle." This 7th-day nodular reaction continues for 21 days or more, constituting the late, or Mitsuda reaction. Since the Mitsuda reaction, which is interpreted as a test of resistance to infection, is the final evolutive consequence of the accelerated tubercle formation, both reactions have the same significance.

Hypersensitivity to the protein derivative of *M. leprae* can be investigated by means of intradermal injection of a leprolin (i.e., a bacillus-free filtrate of lepromin, or our own special preparation (⁸), which provokes in hypersensitized persons an early, tuberculin-type reaction known as the Fernandez reaction.

2. Although the pathogenic mechanism and immunologic significance of the hypersensitivity to the bacillary body and that to the protein derivative are different, yet the two phenomena are frequently associated.

³It is stated in the original text that, of the 23 cases tested 6 weeks after the lepromin injection, the leprolin reactions had increased in intensity in 21 cases; in 1 case there was a slight decrease, and in 1 case the reaction was not controlled.

The observations made in the present study show that individuals hypersensitive to leprolin are also hypersensitive to lepromin. In fact, leprolin-positive persons give the accelerated tubercle formation when injected with lepromin, and therefore they give the positive Mitsuda reaction later. It is for this reason that the positive leprolin reaction, which specifically expresses bacterial protein hypersensitivity of the tuberculin type, presupposes resistance.

3. It seems that there is now agreement among leprologists regarding the sensitizing capability of *M. leprae*, but it is still a matter of debate whether or not repeated injections of lepromin to hypersensitive individuals will provoke an increase of the hypersensitivity.

The observations here reported clearly show that injection of leprolin-positive persons with lepromin increases their degree of hypersensitivity to leprolin.

4. These findings are considered of real significance, not only theoretical but also highly practical.

The Mitsuda test is used by most leprologists as a routine procedure. Nevertheless, its indiscriminate use has great inconveniences, which we here describe briefly: (a) The use of lepromin, a sensitizing antigen, provokes an increase of hypersensitivity in individuals who are already hypersensitive. In tuberculoid cases, as shown by Fernandez as far back as 1938 (²), the *subcutaneous* injection of sufficient lepromin (1.5 cc.) is capable of precipitating a generalized tuberculoid reaction of preexisting lesions. We ourselves have seen grave reactivations of tuberculoid lesions, with tissue necrosis, as a result of an intradermal injection of lepromin. Two of the cases of Table 1 had general reactions on retesting with leprolin. That is apparently unusual, for Garrod and Wade (⁵) seeking—in Africans—evidence of increase of the late reaction in tuberculoid cases by repeated lepromin testing, encountered a general reaction in only 1 of the 15 cases studied, and that only after the 5th of the series of six tests made at 3-week intervals. Nevertheless, it is still necessary to perform many clinical observations on the role played by hypersensitivity in the evolution of leprosy before being assured of the innocuousness of the injection of tuberculoid cases with lepromin.

(b) The use of lepromin in contacts is inadvisable, since its sensitizing effect prevents all later investigations on the sensitizing influence of the focus (^{6,7}).

(c) The use of the Mitsuda reaction in healthy persons as a test of resistance to infection we believe to be wrong. A 21-day nodular reaction induced by lepromin in healthy persons can be the result of the altered reactivation provoked by the injection of the antigen itself (Wade phenomenon), or it can be the result of the final evolution of the accelerated formation of the tubercle. Only by previous observa-

tion of the leprolin reaction, or, in very special cases, by the observation of the presence or absence of the accelerated formation of the tubercle, can this matter be explained (^{9,10}).

5. From all that has been said, there stands out the importance of the Fernandez reaction, performed with leprolin, in the investigation of hypersensitivity and resistance in leprosy. For this reason we insist that the routine investigation of pre-existent hypersensitivity and resistance in leprosy should always be started with the use of a leprolin. In the case of leprolin-positive persons, the later use of a lepromin is harmless. In leprolin-negative persons, only in very special cases should we resort to lepromin in order to determine the presence or absence of the accelerated formation of the tubercle.

CONCLUSIONS

1. The positive leprolin reaction, a protein hypersensitivity reaction of the tuberculin type, also demonstrates the existence of a certain degree of resistance.

2. The intradermal injection of heat-killed *M. leprae* (lepromin) to a leprolin-positive person increases the degree of protein hypersensitivity.

3. The routine investigation of hypersensitivity and resistance in leprosy should always be started with the use of a leprolin, reserving lepromin for very special cases.

SUMMARY

In 23 persons known to be leprolin-positive (19 contacts and 4 tuberculoid cases), Mitsuda-Hayashi lepromin was injected intradermally. After 21 days, and again after 6 weeks following the lepromin injection, the leprolin test was repeated. It was found that: (1) leprolin-positive persons also react positively to lepromin, and (2) the injection of lepromin increases the degree of reactivity to leprolin, much more after six weeks than after 3 weeks.

It is concluded: (a) that the positive leprolin reaction, a test of protein hypersensitivity, also presupposes a certain degree of resistance; and (b) that, due to the sensitizing capability of lepromin, the investigation of hypersensitivity and resistance in leprosy should always be started with the use of a leprolin, reserving lepromin for very special cases.

The value of the Fernandez reaction, performed with a leprolin, is stressed. The positive reaction, besides signifying protein hypersensitivity, also indicates a certain degree of resistance.

RESUMEN

A 23 individuos, 19 convivientes y 4 tuberculoideos, leprolínicos positivos, se les inyecta en forma intradérmica lepromina de Mitsuda-Hayashi. 21 días y 6 semanas

después, se repiten la leprolinorreacción. Los autores comprueban: (a) los leprolínicos positivos, también reaccionan positivamente a la lepromina. (b) la inyección de lepromina, aumenta el grado de reactividad a la leprolina. Concluyen que: (a) la leprolinorreacción positiva, test de hipersensibilidad proteica, también presupone cierto grado de resistencia; (b) debido a las inconveniencias del uso de leprominas, por su capacidad sensibilizante, la investigación de la hipersensibilidad y resistencia en lepra, debería iniciarse siempre con el uso de leprolinas, reservando las leprominas para casos muy especiales; (c) insisten sobre el valor de la reacción de Fernández realizada con leprolinas que, además de significar hipersensibilidad proteica, presupone cierto grado de resistencia.

RESUMÉ

De la lépromine de Mitsuda-Hayashi a été injectée par voie intra-dermique à 23 personnes dont on savait qu'elles réagissaient positivement à la léproline (19 contacts et 4 malades tuberculoïdes). Vingt et un jours après l'injection de lépromine, et de nouveau 6 semaines après celle-ci, l'épreuve à la léproline a été répétée. On a observé que: (1) les personnes positives par la léproline réagissent positivement aussi à la lépromine; (2) l'injection de lépromine accroît le degré de réactivité à la léproline, et ceci est beaucoup plus marqué après 6 semaines qu'après 3 semaines.

On en conclut: (a) que la réaction positive à la léproline, qui est une épreuve d'hypersensibilité aux protéines, suppose aussi au préalable un certain degré de résistance; et (b) que, par suite du pouvoir sensibilisant de la lépromine, l'exploration de l'hypersensibilité et de la résistance dans la lèpre devrait toujours faire d'abord appel à la léproline, la lépromine étant réservée à des cas très particuliers.

Les auteurs insistent sur la valeur de la réaction de Fernandez effectuée avec la léproline. Réaction positive, en outre qu'elle témoigne d'une hypersensibilité aux protéines, indique aussi un certain degré de résistance.

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