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Double Blind Trials to Determine the Late Reactivity of Leprosy Patients and Unaffected Persons to Different Concentrations of Armadillo Lepromin in Comparison to Human Lepromin[1]

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The objective of the present study was to compare the late (30 days) reactivity of leprosy patients and unaffected persons to different concentrations of armadillo lepromin and human lepromin (40 million bacilli/ml). With these trials we expected to determine the concentrations of armadillo lepromin (A) most suitable for testing unaffected persons, for determining the classification and prognosis of leprosy cases, and for epidemiological and vaccine studies. The importance of the investigation for leprosy control and for research purposes is obvious.

To date a similar study does not appear to have been undertaken. Several authors have compared armadillo and human lepromins but only using preparations with a fixed bacillary count. Meyers, *et al.* ([10]) studied 115 leprosy patients tested with human (H) and armadillo (A) lepromins. The patients were classified according to Ridley and Jopling's criteria ([14]) as follows: LL, 39; BL, 8; BB, 21; BT, 19; and TT, 28. The human lepromin contained 175 million bacilli/ml. The initial 22 patients were tested with lepromin-A containing 220 million bacilli/ml; for the remaining 93 patients this suspension was adjusted to 175 million bacilli/ml. Lepromin-A consistently provoked a more intense Mitsuda reaction than lepromin-H. The differences were significant for all categories of patients except the small group of BL patients. It should be noted that out of 10 LL patients, 6 had positive Mitsuda responses (3 to 5 mm). "Histologic evaluations of the biopsy specimens of Mitsuda reactions to lepromin-H and lepromin-A, over the range of the clinical forms of leprosy revealed similar cellular responses in each instance." The individual responses to normal armadillo tissue extract were compared with lepromin-A in 35 patients with the different classes of leprosy. "The results of this series suggest that normal armadillo tissue components participate in the reaction to lepromin-A. This is particularly true in the BT and TT patients."

In a double blind trial, Millar, *et al.* ([11]) tested 112 patients (L, 67 cases; T, 39; I, 1; B, 5) with lepromin-A and lepromin-H containing 160×10^6 bacilli/ml. Summarizing the findings, "36 cases (32.3%) of the total 112 cases demonstrated no difference in the induration stimulated by the armadillo antigen and the human antigen. Fifty-nine cases (52.7%) showed a larger induration from the armadillo antigen, whereas 17 cases (15%) showed a smaller reaction." Among the 39 tuberculoid cases, only 1 (2.6%) had a negative response (less than 3 mm) with lepromin-A and 4 (10.2%) with lepromin-H. Surprising results were reported regarding the 67 lepromatous cases: positive reactions ($\geq$3 mm) in 53.7% of those tested with lepromin-A and in 52.2% of the

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cases injected with lepromin-H. It is known that L cases as a rule are not reactors to lepromin. The histologic examination of the positive lepromin response revealed a similar reaction with both antigens, but lepromin-A stimulated a more severe response. The histologic appearance of the negative reaction—foamy histiocytes and, in several fields, bacilli in phagocytes—was similar for both lepromins.

Antigen-A (40 million bacilli/ml) and antigen-H were used by Pereira and Pereira ([13]) to test 62 contacts, 5 to 17 years old. The late lepromin was read after 21 days, and the mean size in mm was 6.8 for the former lepromin and 5.7 for the latter; the difference was significant. Stronger reactions were observed with lepromin-A. The proportion of negative and doubtful responses was 3.3% with lepromin-A and 8.3% with lepromin-H. The histological granulomatous response in the reactors was similar for both lepromins.

In an unpublished study conducted by the World Health Organization (WHO) in Burma and Japan ([16]), comparing lepromin-H (prepared by Dr. Abe, Tokyo, Japan) with lepromin-A (prepared by Dr. Kirchheimer, Carville, Louisiana, U.S.A.) both with 40×10^6 bacilli/ml, the results were considered similar regarding late reactions. However, in 27 tuberculoid patients, the mean size was respectively 6.8 and 8.3. Among 27 L patients the mean size of the reaction in mm was 0.2 with lepromin-H and 0.3 with lepromin-A.

From the above studies it appears that, either in leprosy patients or in contacts, lepromin-A usually provoked stronger reactions than lepromin-H, when both preparations had the same bacillary content.

MATERIALS AND METHODS

In a first trial the lepromin test was performed on 16 February 1977 in 103 leprosy patients and 35 unaffected young males (normal subjects), 14 to 17 years old.

Leprosy cases were classified taking into account clinical (L.M.B., P.M.G.P., and E.M., Jr.), bacteriological, and histological examinations (R.G.N.) done in previous years before starting the lepromin testing as well as the evolution of their disease. The Madrid classification was adopted for this trial, but the Ridley and Jopling classification was also studied, and comparative results will be reported in the future.

Four preparations were utilized in the first trial: human lepromin (H) in a concentration of 40 million bacilli per ml and armadillo lepromin (A) in the concentrations of 160, 40, and, 20 million bacilli per ml, designated as H 40, A 160, A 40, and A 20, respectively.

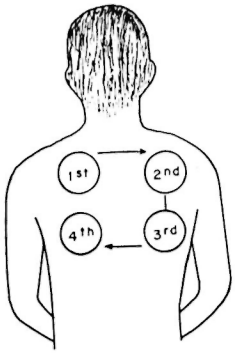
Armadillo lepromin and human lepromin were prepared, enumerated, and provided respectively by Dr. W. F. Kirchheimer (U.S. Public Health Service Hospital, Carville, Louisiana, U.S.A.) through WHO and by Dr. Lygia M. C. Andrade (Director of the former Instituto de Leprologia, Rio de Janeiro, Brazil).

In view of the findings obtained in the field trial, a second group of 47 children and young males, 10 to 17 years old, were tested on 22 November 1977 in another city, Batatais. Later, on 1 August 1978 a third group of 38 young males, 14 to 18 years old, were tested in Ribeirão Preto with lower concentrations of armadillo lepromin. Lepromins-A containing 160, 40, and 20 million bacilli/ml were used in the first trial; preparations containing 20, 10, and 5 million bacilli/ml were tested in the second one; and lepromin-A with 5, 2.5, and 1 million bacilli/ml were used in the third trial.

Each patient or healthy boy received 4 preparations (coded A, B, C, and D) simultaneously in the interscapular region in a randomized order of 24 possible permutations (Fig. 1). The studies were carried out as double blind trials. Only 1 of the investigators (L.M.B.) read the lepromin reactions, and only 1 of the others (N.H.) was aware of the identity of all of the factors involved in the investigation. All the lepromin preparations were injected intradermally in a volume of 0.1 ml.

Almost all the readings of the late lepromin reactions were performed by 1 of the investigators (L.M.B.) These were done 30 days after the injections by measuring 2 diameters of the response and recording the presence or absence of necrosis. The reading criteria were those proposed in the Madrid (1953) and Tokyo (1958) Congresses, slightly modified by Hanks, *et al.* ([4]).

Statistical analyses for comparing the means of the reaction readings to the dif-



ABCD BACD CABD DABC
ABDC BADC CADB DACB
ACBD BCAD CBAD DBAC
ACDB BCDA CBDA DBCA
ADBC BDAC CDAB DCAB
ADCB BDCA CDBA DCBA

24 POSSIBLE PERMUTATIONS FROM WHICH ONE SEQUENCE WAS SELECTED FOR EACH INDIVIDUAL BY RANDOMIZED PROCESS.

FIG. 1. Location order for intradermal injections (0.1 ml).

ferent concentrations of armadillo and human lepromins have taken into account the correlation that exists between 2 populations when each pair of means is compared since the 4 antigens were applied in the same subjects. Therefore, the differences of pairs of observations for each comparison were calculated for the new variable d, and the mean $\bar{d}$ and the standard deviation s_d have also been calculated.

The t test for the null hypothesis was performed by:

$$t_{n-1} = \frac{\bar{d} - 0}{s_d/\sqrt{n}}, \text{ with } n - 1 \text{ degrees of freedom}$$

where

$$s_d = \sqrt{\frac{\Sigma(d - \bar{d})^2}{n - 1}}$$

This is the appropriate testing of the hypothesis for comparison of means when there are correlated populations, and the standard error of the means for the testing should not be used because the populations are not independent.

RESULTS AND DISCUSSION

FINDINGS IN THE FIRST TRIAL

Non-affected persons and T + Tr (BT) cases (Table 1). The late reactions to armadillo lepromins, even to the weaker concentration (A 20), were stronger than those observed in response to the human lepromin (H 40).

The test for the difference of 2 means regarding T and Tr leprosy patients was as follows:

H 40 and A 160
H 40 and A 40
H 40 and A 20
A 160 and A 20
} significant ($p < 0.001$)

A 160 and A 40—significant ($p < 0.05$)
A 40 and A 20—significant ($p < 0.01$)

For the unaffected persons the test for the difference of 2 means for correlated populations (30 day reading) was as follows:

H 40 and A 160
H 40 and A 40
H 40 and A 20
A 160 and A 40
A 160 and A 20
} significant ($p < 0.001$)

A 40 and A 20—significant ($p < 0.01$)

Thus in both leprosy patients and in normal subjects, the late reactions to the armadillo lepromins, even to the weakest concentrations used, were stronger than those induced by human lepromin (H 40).

TABLE 1. *Mean size (mm) of lepromin reactions (30 day reading) using human and armadillo lepromins in T and Tr (BT) leprosy cases and unaffected persons (normal subjects).*

Tested persons	Total	Lepromins[a]			
		H 40	A 160	A 40	A 20
T and Tr cases	37				
Mean (± standard deviation)		7.22 (±4.42)	12.84 (±6.03)	11.57 (±5.55)	10.05 (±4.63)
Unaffected persons	35				
Mean (± standard deviation)		7.11 (±3.74)	13.74 (±5.93)	11.94 (±4.95)	10.57 (±4.53)

[a] H 40 = human lepromin, 40 × 10^6 bacilli/ml. A 160 = armadillo lepromin, 160 × 10^6 bacilli/ml. A 40 = armadillo lepromin, 40 × 10^6 bacilli/ml. A 20 = armadillo lepromin, 20 × 10^6 bacilli/ml.

TABLE 2. *Lepromin testing (30 day reading) of 56 lepromatous cases with human and armadillo lepromins.*

Lepromins[a]	No. with nodule ≥ 3 mm (size range)	% positive
H 40	1 (4 mm)	1.8
A 160	5 (3–7 mm)	9.0
A 40	3 (4–6 mm)	5.4
A 20	1 (6 mm)	1.8

[a] See footnote, Table 1.

Lepromatous cases (Table 2). The higher concentration of armadillo lepromin (A 160) was more prone to cause false positive reactions than human lepromin (H 40) and A 20. It should be noted that Meyers, *et al.* ([10]) found that 6 out of 10 lepromatous cases had positive Mitsuda reactions (3–6 mm) with lepromin-A 160.

Results of late lepromin reactivity in leprosy patients according to the reading criteria proposed in the Madrid (1953) and Tokyo (1958) Congresses, slightly modified by Hanks, *et al.* ([4]) (Table 3). It is interesting to consider the results following these reading criteria because no matter the size of the reaction, the reading is 3+ when there is necrosis.

It appears that armadillo lepromin A 160, A 40, and A 20, in contrast to several antigens prepared from supposed cultures of *M. leprae* and material obtained from other experimental animals, caused only a few positive Mitsuda reactions in lepromatous patients. This positivity was more often observed with A 160 and A 40. Additionally, the degrees of intensity of the responses with A 160 and A 40 were similar, no matter the types or forms of leprosy, and were more intense than those noted with H 40. The degree of intensity of the reactions in I, T, and Tr cases with A 20 was lower than that observed with A 160 and even A 40 but higher than that seen with H 40. In L cases the number of false positive reactions with A 20 is similar to that with H 40 and lower than that seen with A 160 and A 40. On the whole, considering the findings in the types and forms of leprosy studied, A 20 was the armadillo lepromin that gave results most similar to those of H 40.

Results of late lepromin reactivity in unaffected persons according to the reading criteria proposed in the Madrid (1953) and Tokyo (1958) Congresses, slightly modified by Hanks, *et al.* ([4]) (Table 4). Strong (3+) reactions were observed in identical proportions (72.2%) with A 160 and A 40, and were slightly lower (66.7%) with A 20. All 3 lepromin-A preparations induced higher proportions of strong reactions (3+) than H 40 (44.4%). Further comments on the data in Table 4 will be made when considering the findings obtained with lower concentrations of lepromin-A.

From the above, it is evident that armadillo lepromin in the concentrations used

TABLE 3. *Late lepromin reactivity in 103 patients with leprosy according to clinical forms and the concentrations and types of antigens. Data presented as number of observations.*

	Mitsuda reaction					
Lepromins[a]	– (0 mm)	± (1–2 mm)	+ (3–5 mm)	++ (6–10 mm)	+++ (>10 mm or ulceration)	Total
			Indeterminate			
H 40	1	0	4	0	2	7
A 160	1	0	0	3	3	7
40	1	0	0	3	3	7
20	1	0	0	4	2	7
			Polar tuberculoid			
H 40	0	0	2	8	5	15
A 160	0	0	0	2	13	15
40	0	0	0	3	12	15
20	0	0	0	5	10	15
			Tuberculoid in reaction			
H 40	1	4	5	5	7	22
A 160	0	0	3	3	16	22
40	0	0	2	5	15	22
20	1	0	3	7	11	22
			Lepromatous			
H 40	51	2	1	0	0	54
A 160	48	1	3	2	0	54
40	51	0	2	1	0	54
20	52	1	0	1	0	54
			Borderline			
H 40	2	1	0	0	0	3
A 160	0	2	0	1	0	3
40	2	0	0	1	0	3
20	2	0	1	0	0	3
			Lepromatous with reversal reaction			
H 40	2	0	0	0	0	2
A 160	1	0	0	1	0	2
40	1	0	0	1	0	2
20	1	0	0	1	0	2

[a] See footnote, Table 1.

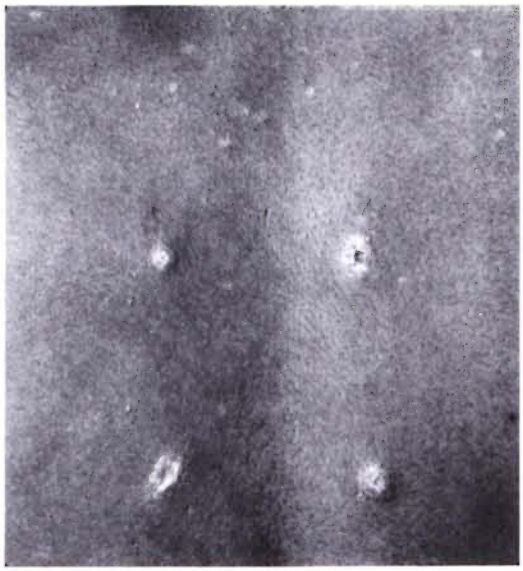


FIG. 2. Thirty day reading (Mitsuda reaction) of lepromin tests with human antigen (H 40) and armadillo antigen (5, 2.5, and 1 × 10^6 bacilli/ml). Location order given in Fig. 1.

induced stronger lepromin reactions than human lepromin, in agreement with the authors quoted above. Such strong reactions (Fig. 2) and subsequent large scars often represent an inconvenience for the current use of armadillo lepromin (mainly 160 and 40 million bacilli/ml) and a nuisance for many patients and unaffected persons. For this reason, it was decided to continue the study in an effort to determine a lower optimum concentration of lepromin-A for clinical use. Besides avoiding the inconvenience and nuisance of ulceration and scarring for many individuals, a lower concentration would allow a substantial saving of lepromin-A and could reduce the proportion of false positive reactions, mainly in L cases.

FINDINGS IN THE SECOND TRIAL

Findings with A 20, A 10, and A 5 (with 20, 10, and 5 million bacilli/ml) as compared to human lepromin (H 40) in unaffected children and young males. In Table 5 it can be seen that the late reactions to armadillo lepromin, even to the weakest concentration

TABLE 4. *Mitsuda reaction (30 day reading) in unaffected persons (normal subjects) in the 3 trials. Data presented as number of observations (percentage of total observations).*

Lepromins[a]	Mitsuda reaction					Total
	– (0 mm)	± (1–2 mm)	+ (3–5 mm)	++ (6–10 mm)	+++ (>10 mm or ulceration)	
First trial Ribeirão Preto						
H 40	3 (8.3)	0 (0.0)	6 (16.7)	11 (30.5)	16 (44.4)	36 (100)
A 160	0 (0.0)	0 (0.0)	1 (2.8)	9 (25.0)	26 (72.2)	36 (100)
40	1 (2.8)	0 (0.0)	0 (0.0)	9 (25.0)	26 (72.2)	36 (100)
20	2 (5.6)	0 (0.0)	0 (0.0)	10 (27.8)	24 (66.7)	36 (100)
Second trial Batatais						
H 40	0 (0.0)	2 (4.3)	19 (41.3)	11 (23.9)	14 (30.4)	46 (100)
A 20	0 (0.0)	0 (0.0)	3 (6.5)	21 (45.6)	22 (47.8)	46 (100)
10	0 (0.0)	1 (2.2)	4 (8.7)	21 (45.6)	20 (43.5)	46 (100)
5	0 (0.0)	1 (2.2)	8 (17.4)	21 (45.6)	16 (34.8)	46 (100)
Third trial Ribeirão Preto						
H 40	2 (5.3)	1 (2.6)	21 (55.3)	4 (10.5)	10 (26.3)	38 (100)
A 5	1 (2.6)	2 (5.3)	10 (26.3)	17 (44.7)	8 (21.0)	38 (100)
2.5	2 (5.3)	1 (2.6)	18 (47.4)	10 (26.3)	7 (18.4)	38 (100)
1	3 (7.9)	2 (5.3)	18 (47.4)	9 (23.7)	6 (15.8)	38 (100)

[a] See footnote, Table 1. A 10 = armadillo lepromin, 10 × 10^6 bacilli/ml. A 5 = armadillo lepromin, 5 × 10^6 bacilli/ml. A 2.5 = armadillo lepromin, 2.5 × 10^6 bacilli/ml. a 1 = armadillo lepromin, 1 × 10^6 bacilli/ml.

TABLE 5. *Mean size (mm) of late lepromin reactions induced by human and armadillo lepromins in unaffected persons (second and third trials).*

Tested persons	Total tested	Lepromins[a]			
		H 40	A 20	A 10	A 5
Second trial					
Batatais	47				
Mean		6.04	9.70	8.98	7.13
(± standard deviation)		(±2.35)	(±4.32)	(±3.39)	(±2.78)
Third trial					
Ribeirão Preto	38				
Mean		4.29	5.76	5.05	4.48
(± standard deviation)		(±1.59)	(±1.85)	(±2.08)	(±1.87)

[a] See footnote, Table 4.

tested (A 5), were still stronger than those induced by human lepromin (H 40) ($p < 0.001$). The differences between A 20 and A 5 and between A 10 and A 5 were significant ($p < 0.001$).

With the Madrid (1953) and Tokyo (1958) Congresses' reading criteria (Table 4), strong reactions (3+) were more often observed with A 20 and A 10. Similar proportions of strong reactions (3+) were seen with A 5 and H 40, but the latter induced a lower percentage of 2+ reactions (23.9) than the former (45.6) and a higher proportion of 1+ reactions (41.3) than A 5 (17.4). The proportions of negative and doubtful reactions were low, as expected, and did not differ with H 40, A 10, and A 5.

These findings led to a third trial to test even lower concentrations of armadillo lepromins.

FINDINGS IN THE THIRD TRIAL

Findings with A 5, A 2.5, and A 1 (with 5, 2.5, and 1 million bacilli/ml, respectively) as compared to human lepromin (H 40) in unaffected young males. In Table 5 it may be seen that significantly stronger reactions were induced by A 5 and A 2.5 lepromin than by H 40 (A 5 compared to H 40: $p < 0.001$; A 2.5 compared to H 40: $p < 0.01$). Results were similar with A 1 and H 40 ($p > 0.05$).

With the Madrid and Tokyo Congresses' reading criteria (Table 4), strong reactions (3+) were slightly more frequent with H 40. When 2+ and 3+ reactions are considered together, however, the percentage was higher with A 5 (65.7), still high with A 2.5 (44.7), and similar with A 1 (39.5) as compared to the 36.8% positives seen with H 40. The 1+ responses were least frequent with A 5. The percentage of negative and doubtful (±) reactions combined was the same (7.9) with H 40, A 5, and A 2.5, and slightly higher with A 1 (13.2). On the whole, the A 1 lepromin would seem to be the most suitable for current use in unaffected persons.

For leprosy patients, the lowest concentration tested was A 20, which caused stronger reactions in T and Tr than H 40. Taking into account that the results in Table 1 with A 160, A 40, and A 20 were similar in these patients and in unaffected persons, lower concentrations of armadillo lepromin (A 5, A 2.5, and perhaps A 1) could be suggested for testing leprosy patients as well. Additionally, the lower concentration of lepromin-A (A 20 compared to A 160 and A 40) was less prone to cause false positive responses in L patients.

Mitsuda reaction and tissue elements of the armadillo lepromin. From the 3 trials it appeared that reactions induced by H 40 were always less intense than those observed with armadillo lepromins, even at concentrations equal to or lower than 40 million bacilli/ml, except for A 1 (1 million bacilli/ml). This suggests that the stronger reactions with armadillo lepromins might be due to the armadillo tissue elements in the skin test preparations as well as their bacillary contents. This hypothesis is supported by the findings of Meyers, *et al.* ([10]), who also suggested that normal armadillo tissue components may contribute to the reaction in lepromin-A. In a blind examination of smears from H 40, A 20, A 10,

and A 5, it does not seem that the content of solid-staining bacilli could be responsible for the results observed.

Previous studies have suggested the importance of tissue elements in the lepromin reaction ([15]). Davey ([1,2]), Lopes de Faria ([8,9]), and later Kooij and Gerritsen ([5]) have reported that tissue elements of a normal triturate and of normal liver particles prepared in the same way as lepromin, produced an inflammatory lesion in 50% of tuberculoid cases, slightly intense and with a tuberculoid structure. These results have not been confirmed by Olmos Castro, *et al.* ([12]), Leiker ([6,7]), and Hadler ([3]). Hadler ([3]), working with guinea pigs, stated that the lesion caused by tissue elements was more fugacious and less intense. He felt that the tissue components, predominantly proteins, do not have antigenicity because of the heat denaturation which occurs during the preparation of the lepromin. Due to thermocoagulation, the tissue elements were felt to only act as a foreign body and as a nonspecific irritant.

Due to their immune deficit, only a few of the lepromatous patients reacted to the armadillo lepromin (though more often with A 160 and A 40 than with A 20). The reactivity of these few lepromatous patients suggests that the combination of the 2 factors (bacillary content plus foreign protein) might be able to induce the late reaction to lepromin in this polar type.

Possible enhancement of the late reaction due to simultaneous injections of 4 lepromins. From the findings in the 3 trials (Table 4) regarding the human lepromin, H 40, always in the same concentration, it appears:

1) The proportion of negative and doubtful reactions combined was similar in the 3 trials.
2) The percentage of 2+ and 3+ responses combined decreased from the first trial (74.9) to the third one (36.8). It should be noted that the groups in these 2 trials (the first and the third) were comparable not only as to their socio-economic background but also as to age. The group tested in the second trial was also comparable to the others, but it also included males of a lower age. The mean size of responses to H 40 decreased also from 7.11 (first trial) to 6.04 (second trial), to 4.29 mm (third trial). (Tables 1 and 5).
3) Obviously, since the percentage of negative and doubtful reactions combined was similar in the 3 trials and that of 2+ and 3+ reactions combined decreased from the first to the third study, there were marked increases in the proportions of 1+ responses from the first (16.7) to the second (41.3) and to the third (55.3) studies.

How can one explain the decreases in the proportions of 2+ and 3+ reactions from the first to the third trials? It does not seem that they depend on a variation in the composition of the tested groups. A decrease in the bacillary content of the stored human lepromin, H 40, could perhaps be partially responsible for the differences observed. When considered together with the results obtained with A 160, A 40, and A 20, A 20, A 10, and A 5, and A 5, A 2.5, and A 1, the findings with H 40 could be at least in part explained by an enhancement of the late reaction produced by higher concentrations of armadillo lepromin injected simultaneously ("potentialization"?, booster-like effect?). Indeed, the reactivities to the different concentrations of armadillo lepromin could also have been enhanced by the simultaneous injections. Each of them could correspond to a "micro-vaccination," and the responses could perhaps be interdependent in each person.

SUMMARY

The authors carried out 3 double blind trials to determine the late reactivity of 103 leprosy patients and unaffected persons to different concentrations of armadillo lepromin (160, 40, 20, 10, 5, 2.5, and 1 $\times$ 10^6 bacilli/ml) in comparison to human lepromin (40 $\times$ 10^6 bacilli/ml). Their conclusions are as follows:

1) From the comparison of several concentrations of armadillo lepromin (160, 40, 20, 10, 5, 2.5, and 1 million bacilli/ml) to human lepromin (40 million bacilli/ml), it seems that the content of 1 million bacilli/ml would be the most adequate for current use in unaffected persons; it allows a sub-

stantial saving of antigen and could reduce the proportion of false positive reactions.

2) For leprosy patients the lowest concentration of armadillo lepromin tested was 20 million bacilli/ml, but when analyzing all the findings, it seems that lower concentrations (5, 2.5, and perhaps better, 1 million bacilli/ml) could be suggested for their testing in routine work, also avoiding false positive reactions in lepromatous patients.

3) The late reactions to armadillo lepromins (from 160 to 2.5 million bacilli/ml) were stronger than those observed in response to human lepromin (40 million bacilli/ml); tissue components of armadillo lepromin (mainly proteins) might be responsible for this finding.

4) Stronger reactions to human lepromin (40 million bacilli/ml) were coincident with the higher bacillary contents of simultaneously administered armadillo lepromins (160, 40, and 20 million bacilli/ml), and their intensity decreased in each trial with the lower concentrations of the armadillo lepromins. The reactivities to human lepromin and perhaps also to armadillo lepromins might have been enhanced by their simultaneous injections.

RESÚMEN

Los autores hicieron tres estudios "en doble ciego" para determinar la reactividad tardía de 103 pacientes con lepra y de personas sanas a diferentes concentraciones de lepromina de armadillo (160, 40, 20, 10, 5, 2.5, y 1.0 × 10^6 bacilos por ml) en comparación con la reactividad a la lepromina humana (40 × 10^6 bacilos por ml). Sus conclusiones fueron:

1) Comparando las diferentes concentraciones de las leprominas de armadillo (160, 40, 20, 10, 5, 2.5, y 1.0 × 10^6 bacilos por ml) con la lepromina humana (40 × 10^6 bacilos por ml), se deduce que la concentración de 1.0 millón de bacilos por ml es la más adecuada para su uso rutinario en personas no afectadas por la enfermedad; esto permite un ahorro substancial de antígeno y podría reducir la proporción de reacciones falsas positivas.

2) En los pacientes con lepra, la menor concentración de lepromina de armadillo probada fue de 20 millones de bacilos por ml pero analizando todos los resultados se hace evidente que podrían recomendarse concentraciones menores (5, 2.5 y quizá mejor aún 1.0 × 10^6 bacilos por ml) para su uso rutinario, evitando también reacciones falsas positivas en los pacientes L.

3) Las reacciones tardías a las leprominas de armadillo con 160 a 2.5 × 10^6 bacilos por ml, fueron más intensas que aquellas observadas en respuesta a la lepromina humana con 40 × 10^6 bacilos por ml; los componentes tisulares en las leprominas de armadillo (principalmente proteínas) podrían ser los responsables de este hecho.

4) Las reacciones más intensas a la lepromina humana (40 × 10^6 bacilos por ml) fueron coincidentes con la mayor reactividad a las leprominas de armadillo que tuvieron las mayores concentraciones bacilares (160, 40, y 20 × 10^6 bacilos por ml) y su intensidad disminuyó proporcionalmente con las menores concentraciones de bacilos. La reactividad a la lepromina humana y quizá también a las leprominas de armadillo pudieran haberse incrementado si ambas leprominas se hubieran inyectado en forma simultánea.

RÉSUMÉ

Les auteurs ont mené trois essais par la méthode du double incognito pour déterminer la réactivité tardive de 103 malades de la lèpre et de personnes ne souffrant pas de lèpre, à différentes concentrations de lépromine d'armadillo (160, 40, 20, 10, 5, 2,5, et 1 × 10^6 bacilles/ml). Cette réactivité a été comparée à celle montrée à l'égard de la lépromine humaine (40 × 10^6 bacilles/ml). Ces conclusions sont les suivantes:

1) En se basant sur la comparaison de plusieurs concentrations de lépromine d'armadillo (160, 40, 20, 10, 5, 2,5 et 1 × 10^6 bacilles/ml) avec la lépromine humaine (40 millions de bacilles/ml) il semble que le contenu de 1 million de bacilles/ml serait le plus approprié pour l'usage courant chez des personnes ne souffrant pas de lèpre. Il permet d'épargner de manière notable l'antigène, et pourrait réduire la proportion de réactions faussement positives.

2) En ce qui concerne les malades de la lèpre, la concentration la plus faible de lépromine d'armadillo qui ait été essayée était de 20 millions de bacilles/ml. Néanmoins, lorsque l'on analyse toutes les observations, il semble que les concentrations les plus faibles (5, 2,5 et peut-être encore mieux un million de bacilles/ml) pourraient être indiquées pour les épreuves de routine, car elles éviteraient également des réactions faussement positives chez les malades lépromateux.

3) Les réactions tardives aux lépromines d'armadillo (à des concentrations de 160 à 2,5 millions de bacilles/ml), étaient plus prononcées que celles observées à la suite de l'injection de lépromine humaine (40 millions de bacilles/ml). Les composants tissulaires de la lépromine d'armadillo (surtout les protéines) pourraient expliquer cette observation.

4) Les réactions plus prononcées à la lépromine humaine (40 millions de bacilles/ml) coïncidaient avec un contenu bacillaire plus élevé dans les lépromines d'armadillo (160, 40, et 20 millions de bacilles/ml) et l'intensité de ces réactions décroissait dans chaque épreuve avec les concentrations les plus faibles de ces dernières. La réactivité à la lépromine humaine, et peut-être également la réactivité aux lépromines d'armadillo, pourraient avoir été stimulées par le fait que ces deux lépromines ont été injectées simultanément.

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