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Relation between Pre-vaccination and Post-vaccination Tuberculin Sensitivity

A Contribution to the Ecology of BCG Vaccination

GERARD WIJSMULLER [1]

BCG vaccination is commonly assessed in terms of post-vaccination sensitivity to tuberculin. If vaccination is followed by the development of a high degree of tuberculin sensitivity, it is assumed that the vaccination was successful. If, on the other hand, tuberculin sensitivity does not develop, it is assumed that the vaccination was unsuccessful. Both these assumptions equate post-vaccination tuberculin sensitivity with BCG-induced tuberculin sensitivity and disregard the possibility that environmental factors, such as the prevalence of low-grade naturally acquired tuberculin sensitivity, may affect the outcome of vaccination. Thus, while it seems reasonable to equate post-vaccination and BCG-induced tuberculin sensitivity in areas where low-grade sensitivity is uncommon, it might be unjustifiable to do so in areas where such sensitivity is prevalent.

This paper analyses the relation between pre- and post-vaccination tuberculin sensitivity in a community with a high prevalence of naturally acquired low-grade tuberculin sensitivity. From this analysis it appears that post-vaccination tuberculin sensitivity may be only partly BCG-induced and cannot therefore be considered a reliable measure of the success of BCG vaccination in the presence of naturally acquired low-grade sensitivity. The author discusses the implications of this finding and concludes that a further evaluation of the protection afforded by BCG in areas where low-grade tuberculin sensitivity is prevalent is much needed.

Since 1956, mass BCG vaccination and revaccination have been practised in the former Netherlands New Guinea (now the West Irian Province of the Republic of Indonesia). When areas covered during the initial campaign were re-examined one year later, it became apparent that BCG vaccination did not always induce the same degree of tuberculin sensitivity. Unexpectedly, the degree of post-vaccination tuberculin sensitivity showed geographic variation, despite the fact that the same vaccines had been administered by the same personnel according to a strict protocol. Experience in later years confirmed these earlier observations.

This geographic variation in post-vaccination tuberculin sensitivity seemed somehow related to the percentage of the population that had not been vaccinated. If many people had been excluded from the vaccination programme, tuberculin sensitivity among the vaccinees one year later was rather strong. If, however, few people had been excluded, tuberculin sensitivity among the vaccinees one year later was rather weak. In other words, a positive correlation seemed to exist between the total amount of tuberculin sensitivity present in a given community at the initial campaign and the capacity of BCG vaccine to induce tuberculin sensitivity in those vaccinated in that community.

One explanation is that superinfection with human tubercle bacilli takes place in the interval between vaccination and retest. Although this possibility certainly exists, it is not likely to be the only or even the most important cause of the described observation. Studies on BCG-induced tuberculin sensitivity, designed to measure the contribution of superinfection with human tubercle bacilli to the total amount of tuberculin sensitivity present some time after vaccination, have suggested that superinfections did not contribute materially to the recorded post-

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vaccination sensitivity (Wijsmuller, 1959b). Therefore, it seems more likely that some other factor or factors, unrelated to the prevalence of tuberculous disease and yet directly associated with the total amount of tuberculin sensitivity measured at the initial campaign, may be responsible for the observed geographic variation in the change in sensitivity among the vaccinated populations.

In addition to these geographic variations, a relation between the degree of post-vaccination tuberculin sensitivity and the age of the vaccinees was apparent. Among BCG-vaccinated infants and young children, the post-vaccination reactions were always smaller than among adolescents and adults.

As the geographic variation in post-vaccination tuberculin sensitivity indicated a relation between pre- and post-vaccination sensitivity in the *communities* examined, it was desirable to determine whether a similar relation could be established for the vaccinated *individuals*. The difference in response to vaccination that had been observed among different age-groups might then be related to differences in pre-vaccination sensitivity rather than to age.

As reported earlier (Wijsmuller 1959a, 1961, 1963), low-grade sensitivity to tuberculin is very prevalent in New Guinea. This type of sensitivity is not related to the presence of tuberculous infections in the community but to some unidentified agent or agents in the environment. While a low dose of human purified protein derivative (PPD) may produce an induration as large as 16 mm in diameter in some individuals who have been sensitized by this still-obscure mechanism, the majority will show substantially smaller reactions or no reaction at all. If, however, the same individuals are retested with a higher dose, it is apparent that many who had not reacted to a low dose of human PPD do possess substantial sensitivity to tuberculin, but that a higher dose of tuberculin must be employed to demonstrate its presence. Thus, almost 100% of adult males, non-reactors to a low dose of human PPD and living at a low altitude in the coastal districts of New Guinea, do react to a twentyfold higher concentration of the same tuberculin preparation (Wijsmuller, 1963).

The measurement of an individual tuberculin reaction is always subject to a considerable amount of unavoidable error. In persons with tuberculin sensitivity, the error can be either negative or positive. In persons *without* tuberculin sensitivity, the error can only be positive, as one cannot measure less than 0 mm. In recognition of these unavoidable errors, one should interpret individual reactions, and comparisons among them, with caution.

If, however, one considers a distribution of reactions according to size, the situation is somewhat different. Positive and negative errors are of the same relative size, and it is therefore possible to compare distributions of reactions obtained among similar populations more reliably than one can compare individual reactions.

One can apply the same reasoning to a particular portion of a distribution of reactions, such as, for example, the range of reaction size taken as criterion for vaccination (in the present study, 0-7 mm in one population, 0-11 mm in the other). In general, larger reactions must represent the presence of more sensitivity, smaller reactions the presence of less sensitivity, to tuberculin; only the test results in the younger infants and in the older adults may be exempted from this general rule. In young infants, who have not yet had enough time to acquire low-grade tuberculin sensitivity, any reaction within the range of the vaccination criterion is likely to be traumatic in origin (i.e., excess of positive errors). In adults, on the other hand, naturally acquired low-grade tuberculin sensitivity has attained a maximum value, and the absence of a reaction may therefore not indicate the absence of sensitivity but rather our failure to demonstrate its presence (i.e., excess of negative errors).

It is with these limitations in mind that we have endeavoured to compare the reaction to a low dose of human PPD prior to and some time after vaccination with BCG among males and females of different age. The present paper discusses the findings of this investigation.

MATERIALS AND METHODS

Tuberculin

Human tuberculin PPD, batches RT 22 and RT 23, both prepared at the Statens Seruminstitut, Copenhagen, was employed. These materials were diluted in phosphate buffer to concentrations of, respectively, 5 and 1 tuberculin units (TU) per 0.1 ml. To the dilutions of RT 23, 0.005% Tween 80 (a stabilizing agent) was added, as recommended by the producer (Guld et al., 1958).

All tuberculin dilutions were kept under refrigeration from the time of preparation until used. The vaccinating teams were instructed to discard the second half of each bottle of diluted tuberculin so as to reduce the influence of adsorption (Guld et al.,

1955; Magnus et al., 1956). This practice was continued also after RT 23 (with Tween 80) had replaced the previous batch of RT 22 (without Tween 80). Fresh bottles of tuberculin were used on each working day.

Syringes

All syringes were tested for leakage by the method of Guld & Rud (1953). In all but one study, the only syringes used were those that leaked less than 0.2 ml after an internal pressure of 5.5-6.0 kgf/cm² had been applied for six minutes. This exception concerned a mass campaign in the Sentanie district, the results of which are described separately below. In this instance, difficulties in the supply of syringes made it impossible to adhere to the above criterion, and it was necessary to accept syringes with leakage rates as high as 0.35 ml.

Syringes were generally retested after they had been used for approximately 400 injections; they were used again only if they still conformed to the above-mentioned criteria.

Test procedures

All tests were performed on the dorsum of the forearm by the careful injection of 0.1 ml of the appropriate dilution of tuberculin (Guld, 1954). Readings were made 72 hours after injection, at which time the transverse and the longitudinal diameters of the induration were measured with a plastic ruler and the mean size recorded (rounded off to the nearest millimetre).

Special care was taken not to retest at sites that had been used previously (WHO Tuberculosis Research Office, 1955).

BCG preparations

Liquid BCG Manila (Alabang) was used in the Sentanie district study, and liquid BCG Copenhagen (Statens Seruminstitut) was used in the Noemfoor Island study. Both vaccines were received in refrigerated containers and kept under constant refrigeration until used.

Ampoules were used only on the day on which they were opened. Strict precautions were taken to protect the vaccine from possible damage by sunlight. Vaccinations were given inside a building, usually a church or a school. The ampoules were kept on the table in wooden blocks, and the syringes used for vaccination were covered with rubber tubing, cut over-length in order to protect the vaccine while in the syringe.

Study populations

Pre- and post-vaccination reactions were compared among the vaccinees in two areas — namely, the Sentanie district and Noemfoor Island.

The Sentanie population was tuberculin-tested and BCG-vaccinated in 1956. Vaccinations were given to approximately 50% of those examined. Practically all (95%) of the population participated in this initial campaign.

Three samples of the total population were retested some time after vaccination as follows:

(*a*) Eight to ten weeks after the initial campaign, a number of villages were re-examined to determine the reaction to tuberculin shortly after vaccination. This study was undertaken by a special BCG assessment team and will be referred to as the 1956 BCG Assessment Study.

(*b*) Approximately one year after the initial campaign, a similar sample of the population of the Sentanie district was retested. A large proportion of these people (close to 80%) had also been included in the 1956 BCG Assessment Study. We will refer to this group as the 1957 BCG Assessment Study.

(*c*) The remainder of the vaccinated population was retested during a mass revaccination campaign in 1957. This campaign, conducted with syringes of rather poorer quality and by a team with somewhat less experience than the one that did the assessment studies, will be referred to as the 1957 Mass Revaccination Campaign.

While 5 TU of RT 22 in 0.1 ml of phosphate buffer (not containing Tween 80) and BCG Manila (Alabang) were used in Sentanie, 1 TU of RT 23 in 0.1 ml of phosphate buffer (containing Tween 80) and liquid BCG Copenhagen were used on Noemfoor Island. The initial campaign was done in 1959, the resurvey approximately one year later, in 1960. Both surveys on Noemfoor Island were part of a more detailed study of the local picture of tuberculosis and leprosy and the effect of BCG vaccination thereon. In the present communication, however, we report only on the findings regarding the relation between pre- and post-vaccination tuberculin sensitivity.

OBSERVATIONS

Relation between percentage of population vaccinated during initial campaign and sensitivity measured one year later

As mentioned at the beginning of this paper, a relation between the size of the post-vaccination

reactions and the percentage of the population vaccinated initially had been observed. Post-vaccination reactions tended to be larger in districts where relatively few people were vaccinated and smaller in districts where many people were vaccinated.

In order to eliminate any unintentional variation in the conduct of these local campaigns that might have contributed to the difference in vaccination results, it seemed desirable to use a single survey, break it down into subgroups, and then make comparisons between them. As the 1957 Mass Revaccination Campaign in the Sentanie district was conducted by a single team from the Division for Tuberculosis Control and was performed as a single large project, its subjects seemed to form an appropriate group on which to study the interrelation between the percentage of persons vaccinated with BCG and their sensitivity to tuberculin, one year later.

To this end, the villages were first arrayed in order of the percentage of the population that had been vaccinated initially. Subsequently, the total series was broken down into four subgroups of approximately equal size and with decreasing percentages of vaccinated people. Table 1 summarizes this breakdown.

Distributions of reactions measured among the vaccinated inhabitants of these 4 groups of villages are given in the upper four segments of Fig. 1. The lower segment of this figure shows, by way of comparison, the distribution of reactions among persons who had been excluded from vaccination or who had not been tested in 1956.

In examining these distributions, one should realize that the mean ages of these various population subgroups are different. The smaller the percentage of vaccinated persons, the lower their mean age; the higher the percentage of vaccinated persons, the higher their mean age (see also Table 1).

Although higher age is, in our experience, associated with larger reactions to tuberculin after vaccination with BCG, it is apparent from Fig. 1 that the smaller the percentage of persons vaccinated initially (low mean age), the larger their mean post-vaccination reactions.

It is not likely that this increase in the mean size of the reactions is caused primarily by superinfections with human tubercle bacilli. The large number of

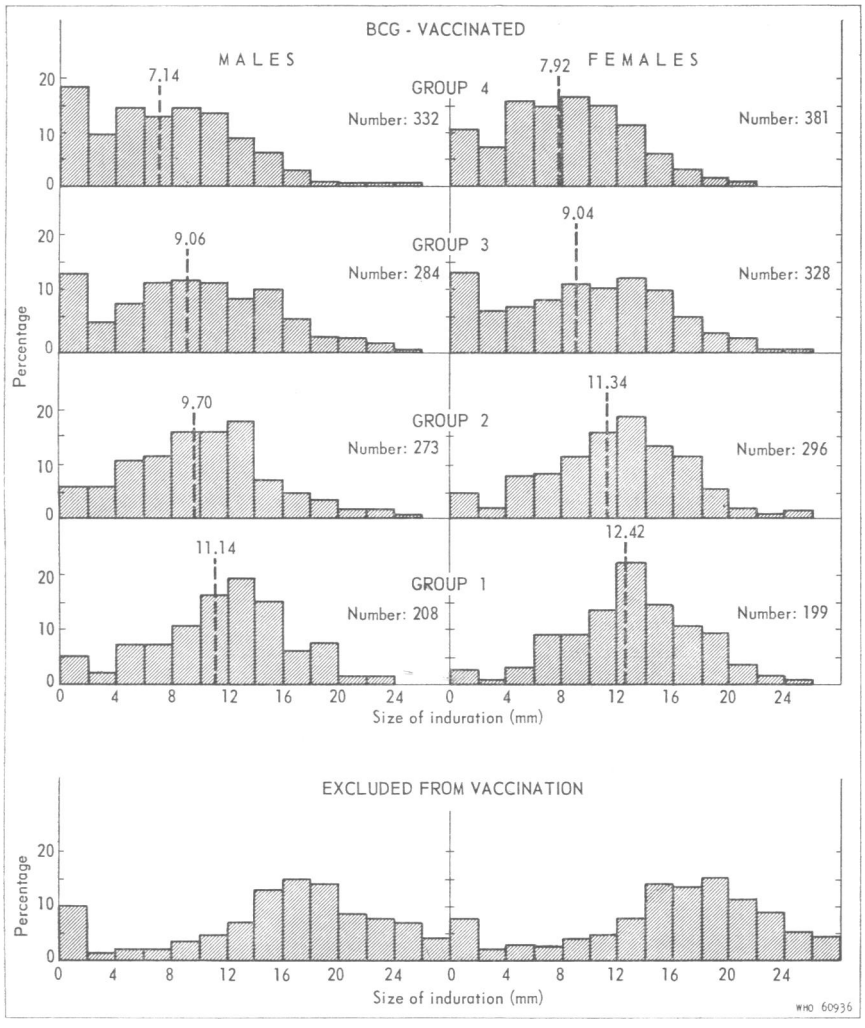
TABLE 1

POPULATION COVERED DURING THE 1957 MASS REVACCINATION CAMPAIGN, ACCORDING TO THE PERCENTAGE OF THE VILLAGE POPULATIONS VACCINATED IN 1956

Age (years)	Group 1			Group 2			Group 3			Group 4		
	No. vaccinated	Total	% vaccinated	No. vaccinated	Total	% vaccinated	No. vaccinated	Total	% vaccinated	No. vaccinated	Total	% vaccinated
Males												
2-5	75	111	68	88	104	85	84	96	88	113	123	92
6-11	80	138	58	87	117	74	80	98	82	86	96	90
12-17	12	61	20	28	55	51	36	54	67	35	54	65
18-24	21	251	8	44	235	19	57	229	25	69	200	35
45+	2	34	6	9	68	13	4	40	10	9	41	22
Total	190	595	32	256	579	44	261	517	50	312	514	61
Females												
2-5	72	105	69	83	102	81	99	123	80	100	106	94
6-11	69	128	54	84	125	67	76	89	85	77	89	87
12-17	12	55	22	42	86	49	36	67	54	32	47	68
18-44	28	275	10	57	295	19	87	269	32	132	256	52
45+	5	47	11	12	64	19	18	55	33	21	39	54
Total	186	610	30	278	672	41	316	603	52	362	527	69

FIG. 1

DISTRIBUTION OF POST-VACCINATION REACTIONS TO 5 TU OF RT 22 IN 0.1 ml (WITHOUT TWEEN 80) AMONG THE PEOPLE IN FOUR GROUPS OF VILLAGES WHERE DIFFERENT PERCENTAGES OF THE POPULATION HAD BEEN VACCINATED AT THE INITIAL CAMPAIGN



persons that could be vaccinated during the initial campaign indicates that the infection rate must be too low to explain differences in distribution by this mechanism alone. Furthermore, if superinfections with tubercle bacilli were the only cause, one would expect a selective increase in the frequency of the very large reactions. Such an increase in frequency is not observed, however. The increase in mean size is due, rather, to a gradual shift of the entire distribution towards a higher mode, suggesting that some factor other than infection with human tubercle bacilli must be responsible. Nor does the apparent shift in these distributions towards a higher mode seem to be fortuitous. An analysis of variance shows that the probability that such a series of distributions would arise by chance alone is less than 1 in 1000.

In a further breakdown of this analysis, the difference between each distribution and that of the

TABLE 2

DISTRIBUTIONS OF POST-VACCINATION REACTIONS TO 5 TU OF RT 22 IN 0.1 ml OF PHOSPHATE BUFFER (WITHOUT TWEEN 80) AMONG THE TOTAL POPULATIONS OF FOUR GROUPS OF VILLAGES [a]

Reaction to 5 TU (mm induration)	Village group (males)				Village group (females)			
	1	2	3	4	1	2	3	4
0-	11	15	40	59	5	13	46	39
2-	5	15	15	30	2	5	25	28
4-	15	29	25	46	6	23	29	58
6-	15	31	36	42	18	24	30	54
8-	22	42	37	47	18	33	40	62
10-	34	42	35	44	27	45	37	55
12-	39	48	27	29	44	54	44	42
14-	32	19	32	20	29	39	36	23
16-	13	13	17	9	21	33	20	12
18-	16	9	8	3	18	16	10	5
20-	3	5	7	1	7	5	8	3
22-	3	4	4	1	3	2	1	—
24-	—	1	1	1	1	4	1	—
26-	—	—	—	—	—	—	1	—
28-	—	—	—	—	—	—	—	—
Total	208	273	284	332	199	296	328	381
Mean	11.14	9.70	9.06	7.14	12.42	11.34	9.04	7.92

[a] A summary of the computations for analysis of variance of these distributions is presented in Table 3.

remaining groups, which included larger percentages of vaccinated persons, was estimated. Group 1 (less than 40% vaccinated) was thus compared with the total of groups 2, 3 and 4; group 2 (40-45% vaccinated initially) with the total of groups 3 and 4, and finally group 3 (46-55% vaccinated) with group 4 (more than 55% vaccinated). This subsidiary analysis shows that the difference between one distribution and the others is significant at the 0.001 level in all cases except for one in which a P value of 0.01 was calculated. It is therefore concluded that there exists a rather gradual shift in the distributions of groups 1-4 towards a smaller mean value, although the difference between each set of distributions may not always be of the same magnitude. The actual distributions and their mean values are shown in Table 2, and a summary of the computations for the analysis of variance is presented in Table 3. As all the data were collected among the population of one district by one team in one single survey, using the same tuberculin dilution, it is felt that the relation between pre- and post-vaccination reactions that has been described is a real relation and not a fortuitous observation that could be attributed to chance.

The positive correlation between the size of the post-vaccination reactions and the percentage of the population vaccinated initially does not seem to be confined to any one age-group. Fig. 2 portrays age and sex specific mean indurations among the population of these four groups of villages.[1] Although the lines in Fig. 2 show some irregularity, they seem to indicate that:

(*a*) post-vaccination tuberculin sensitivity tends to increase with age until it attains a certain maximum value; and

[1] Correlation tables showing the frequency distributions on which the calculations of the mean values were based have been deposited in the WHO Library. Copies may be obtained on request.

TABLE 3

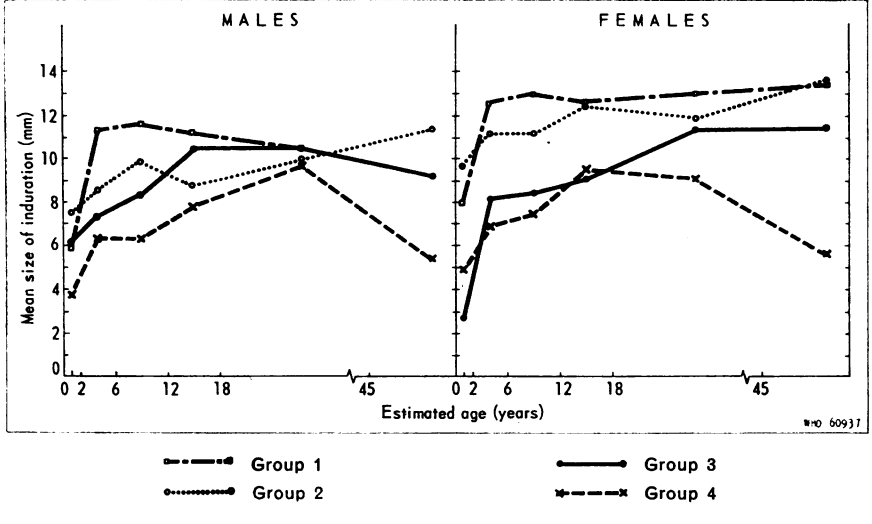
SUMMARY OF COMPUTATIONS FOR ANALYSIS OF VARIANCE FOR THE DISTRIBUTIONS SHOWN IN TABLE 2 [a]

Source of variation	Degrees of freedom	Sum of squares	Mean square	Variance ratio (*F*)	P value
		Males			
Between columns:	(3)	(562)	(187)	(28.1557)	< 0.001
Subsidiary:					
1 *versus* 2, 3 and 4	1	286	286	42.9926	< 0.001
2 *versus* 3 and 4	1	134	134	20.1434	< 0.001
3 *versus* 4	1	142	142	21.3460	< 0.001
Within columns:	1 093	7 271	6.6523		
Total	1 096	7 833			
		Females			
Between columns:	(3)	(906)	(302)	(47.7727)	< 0.001
Subsidiary:					
1 *versus* 2, 3 and 4	1	409	409	64.6988	< 0.001
2 *versus* 3 and 4	1	442	442	69.9190	< 0.001
3 *versus* 4	1	55	55	8.7003	< 0.01
Within columns:	1 200	7 586	6.3216		
Total	1 203	8 492			

[a] Calculations based on 2-mm groupings.

FIG. 2

MEAN SIZES OF POST-VACCINATION REACTIONS TO 5 TU OF RT 22 IN 0.1 ml (WITHOUT TWEEN 80), BY AGE AND SEX, AMONG THE PEOPLE IN FOUR GROUPS OF VILLAGES WHERE DIFFERENT PERCENTAGES OF THE POPULATION HAD BEEN VACCINATED AT THE INITIAL CAMPAIGN



(*b*) the mean size of the reactions tends to be larger among the populations of those groups of villages where a larger proportion has been excluded from vaccination.

Correlation between size of reaction to tuberculin before and after vaccination among groups of individuals

Pre- and post-vaccination reactions were tabulated by size of induration in tables from which correlation coefficients (*r* values) and regression lines have been calculated using the method of least squares.

For the calculation of the regression lines, the size of the pre-vaccination reaction has been taken as the independent variable, while post-vaccination sensitivity has been treated as the dependent variable. This was done because selection for vaccination is based on the size of the reaction to the screening test, regardless of the errors inherent in the testing procedure. Any person with a reaction that exceeds the vaccination criterion is excluded, again regardless of the possibility of misclassification. Furthermore, it was the pre-vaccination reaction as measured that appeared to have a relation to the post-vaccination reaction as determined some time later.

Calculations for the Sentanie district are based on tables presented earlier, in an abbreviated form, in unpublished government reports; the calculations for Noemfoor Island were made from unpublished and unabbreviated tables showing the exact measurements of the pre- and the post-vaccination reactions. The latter group is of particular interest because, on Noemfoor Island, only 50% of those eligible were vaccinated, while the other 50%, selected at random,[1] received an intradermal injection of 0.1 ml of saline instead of BCG. Consequently, for each age and sex group of the population of Noemfoor Island, two regression lines could be calculated, one for the relation between pre- and post-vaccination reactions, and the other for the relation between the first and the second test results among the non-vaccinated controls.[2] The difference between these two lines must represent the total increment in sensitivity resulting from testing and vaccination as compared to testing alone. The significance of any difference in sensitivity between the two groups has been determined by co-variant analysis.

Sentanie district studies

As mentioned earlier, all the Sentanie district studies reported here made use of a standard test with 5 TU of RT 22 in 0.1 ml of phosphate buffer (without Tween 80). Liquid BCG Manila (Alabang) had been used in the initial campaign.

(*a*) *Correlation between size of reaction to tuberculin before and after vaccination in the 1956 BCG Assessment Survey.* The coefficients of correlation between the two tests and their corresponding regression lines were calculated for six age-groups of males and females. These age-groups are based on estimates, since the people from the district usually do not know their exact age. While such an age estimation is certainly not the most accurate, it is believed that the degree of inaccuracy is irrelevant to the present considerations.

Fig. 3 shows the regression lines calculated for these 12 groups of people.[3]

The horizontal axis of Fig. 3 represents the pre-vaccination reaction to 5 TU in millimetres of induration; the vertical axis represents the reaction to the same test after vaccination with BCG. Where the correlation coefficient was significant at the *0.05 level* or less, the regression line has been drawn as a heavy line; where the coefficient was not significant at this level, a light line has been used.

The values for *N* (number examined in each group), *r* (correlation coefficient) and P (probability that this correlation could be explained by chance alone) are also given in Fig. 3.

It is interesting to note that significant correlations were more frequently encountered among children than among adults. There may be a tendency for the coefficient to increase with increasing age and, having reached a maximum value, to decrease or to become non-significant.

(*b*) *Correlation between size of reaction to tuberculin before and after vaccination in the 1957 BCG Assessment Survey.* The correlation between pre- and post-vaccination reactions measured among a sample of the population of the Sentanie district during the 1957 BCG Assessment Survey is presented in Fig. 4 in the same way as was done in Fig. 3 for

[1] Fisher's tables were used. Random numbers were assigned to the people in the order in which they presented themselves for test reading. Odd-numbered people received the placebo, even-numbered ones received BCG.

[2] Although the pre- and post-vaccination reactions among the vaccinated, as well as the first and second test results among the controls, are subject to the same inherent error of determination, and hence correlation analysis is appropriate, it is necessary, as explained earlier, that the pre-vaccination reaction be considered the independent variable. Regression as well as correlation analysis is therefore used.

[3] Tables showing the data on which the calculations were based have been deposited in the WHO Library. Copies may be obtained on request.

FIG. 3. RELATION BETWEEN SIZE OF REACTIONS TO 5 TU OF RT 22 IN 0.1 ml (WITHOUT TWEEN 80) BEFORE VACCINATION AND 8-10 WEEKS AFTER VACCINATION, BY AGE AND SEX: 1956 BCG ASSESSMENT SURVEY

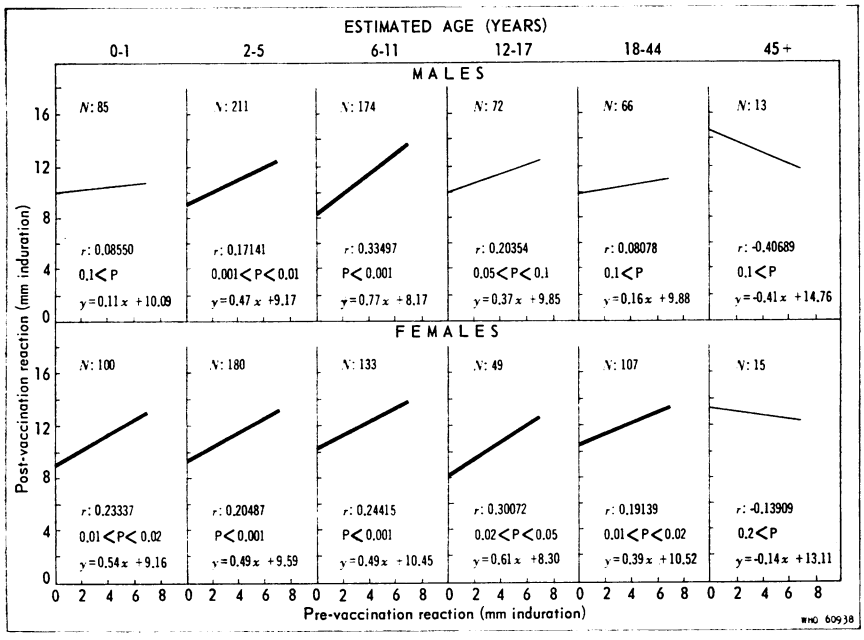


FIG. 4. RELATION BETWEEN SIZE OF REACTIONS TO 5 TU OF RT 22 IN 0.1 ml (WITHOUT TWEEN 80) BEFORE VACCINATION AND ONE YEAR AFTER VACCINATION, BY AGE AND SEX: 1957 BCG ASSESSMENT SURVEY

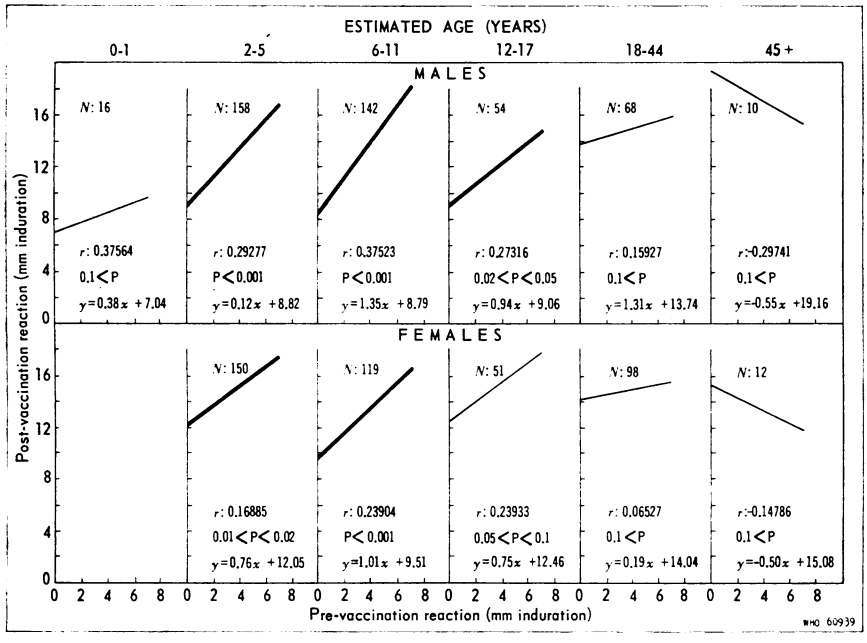
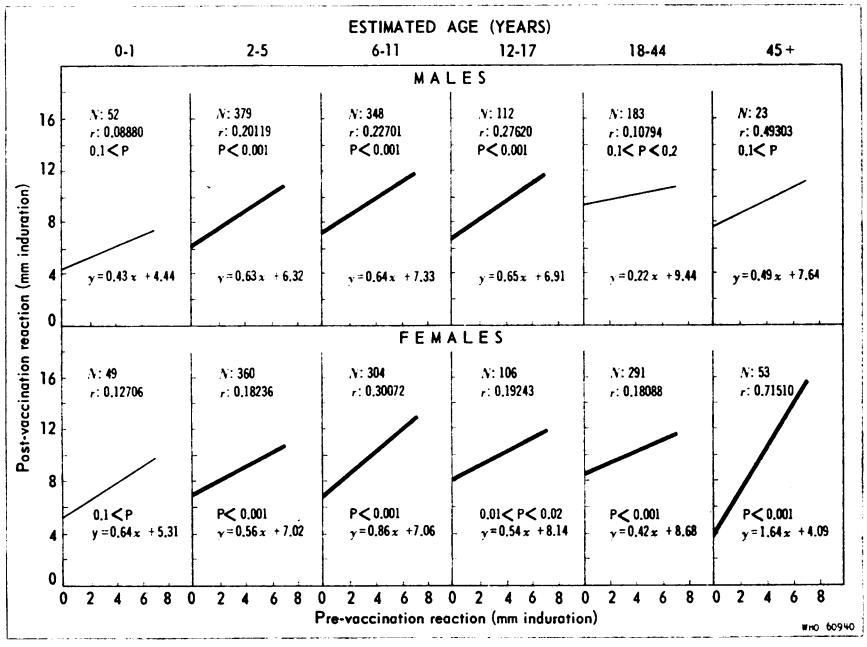


FIG. 5

RELATION BETWEEN SIZE OF REACTIONS TO 5 TU OF RT 22 IN 0.1 ml (WITHOUT TWEEN 80) BEFORE VACCINATION AND ONE YEAR AFTER VACCINATION, BY AGE AND SEX: 1957 MASS REVACCINATION CAMPAIGN



the 1956 survey.[1] It may be recalled here that approximately 80% of this sample was also contained in the 1956 BCG Assessment Survey.

As not one girl aged one year or less had a pre-vaccination reaction exceeding 0 mm, it was not possible to calculate a correlation coefficient for this group.

Fig. 4 is similar to Fig. 3. The correlation between pre- and post-vaccination tuberculin sensitivity is again significant among children aged 2-11 years. Above that age range, however, the correlation coefficient and the significance thereof tend to decrease.

Finally, it may be pointed out that the reactions in 1957 were considerably larger than those measured in 1956. In addition, the regression lines calculated for 1957 tended to be steeper. Both observations are compatible with earlier reports regarding an enhancing effect of tuberculin testing on tuberculin sensitivity among the BCG-vaccinated (Magnus, 1957; Magnus & Edwards, 1955).

(c) *Correlation between size of reaction to tuberculin before and after vaccination in the 1957 Mass Revaccination Campaign.* The findings in the 1957 Mass Revaccination Campaign are presented in Fig. 5.[2] Again, a significantly positive correlation between pre- and post-vaccination sensitivity was found among children aged 2-11 years and even in the next group (those aged 12-17 years).

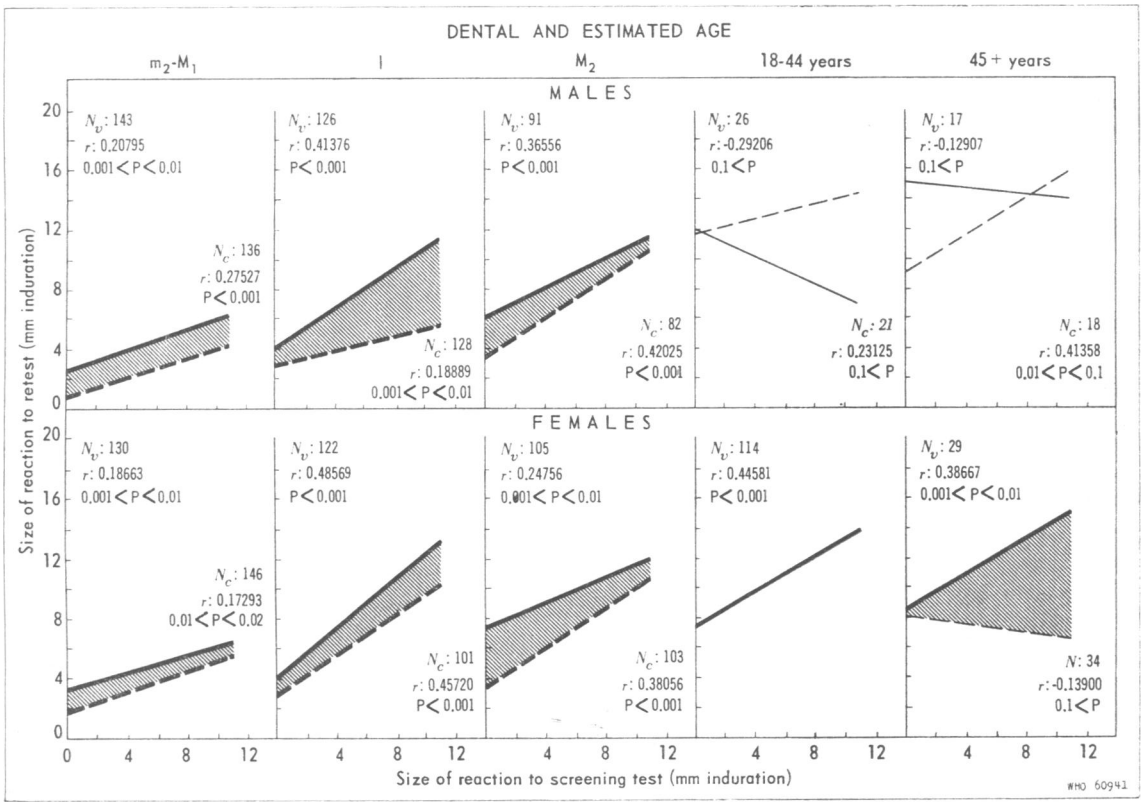
It will be seen that the post-vaccination reactions measured in the 1957 Mass Revaccination Campaign were considerably smaller than those measured in the 1957 BCG Assessment Survey. This is not surprising. The reaction results in an assessment survey always tend to be somewhat larger than those measured in a mass campaign. Furthermore, it may be recalled that a large percentage of the persons tested in the 1957 BCG Assessment Survey had also been examined one year earlier. This additional testing may have enhanced their reactions (Magnus, 1957; Magnus & Edwards, 1955). As the examinations were not performed by the same team under the same

[1] Tables showing the data on which the calculations were based have been deposited in the WHO Library. Copies may be obtained on request.

[2] Tables showing the data on which the analysis was based have been deposited in the WHO Library. Copies may be obtained on request.

FIG. 6

COMPARISON OF RELATION BETWEEN SIZE OF REACTIONS TO 1 TU OF RT 23 IN 0.1 ml (WITH TWEEN 80) AT INITIAL TEST AND AT A SECOND TEST ONE YEAR LATER AMONG THE VACCINATED AND THE CONTROLS, BY AGE AND SEX



circumstances, we have not attempted to test the significance of the observed differences.

Noemfoor Island study [1]

As mentioned earlier, the vaccination campaign on Noemfoor Island offered BCG vaccination to a 50% sample of those eligible for vaccination, while the remaining 50% were injected with 0.1 ml of normal saline in order to serve as controls. 1 TU of RT 23 in 0.1 ml of diluent (with Tween 80) was used for the tuberculin test and liquid BCG Copenhagen as the vaccine.

Pre- and post-vaccination reactions were compared in the same way as has been described for the three Sentanie district studies. In addition, the first and second test results among the controls were similarly compared. Regression lines for both sets of data are presented in Fig. 6. The findings among the vaccinated are portrayed as continuous lines, those among the controls as broken lines. Where the correlation

[1] It should be noted that age distributions in this study population are given in terms of dental age and not in terms of estimated age. At the time of this study, the practice of assessing age on the basis of dental eruption had been adopted. Accordingly, the left upper jaw was inspected for dental elements and the last erupted element recorded. The symbols, given in the order of eruption, indicate:

0: no element visible.
m1: milk molar 1 visible.
m2: milk molar 2 visible.
M1: permanent molar 1 visible.
I: permanent incisor visible.
M2: permanent molar 2 visible.
M3: permanent molar 3 visible.

The following conversion table can be applied:

0-ml: 0-1 years of age
m2: 2-5 years of age
Ml-I: 6-11 years of age
M2: 12-17 years of age
M3: 18 years and older

For further information the reader is referred to the original publication on the subject by Voors & Metselaar (1958).

coefficients were significant at the *0.05 level* or less, heavy lines are used. Where the findings were not significant at this level, light lines are used.

No correlation coefficient was calculated for the very young (age 0-ml) as there was hardly any measurable pre-vaccination reaction in this age-group. It is therefore not represented in Fig. 6.[1]

The relation between pre- and post-vaccination reactions is again very similar to what has been described in the Sentanie district studies. Positive and significant correlations between the two tests were found among the younger age-groups. In adults, however, a positive and significant correlation was only found among the females.

Unfortunately, the data collected for females aged 18-44 years were lost owing to some administrative error and cannot be presented. In the remaining 9 groups of controls, however, the regression lines describe a relation not unlike that found among the BCG-vaccinated. This seems to confirm what is suggested by the relation between pre- and post-vaccination sensitivity found in the Sentanie studies —namely, that not all the change in sensitivity is the result of vaccination, but that part of it must be explained differently. As the vaccinated and the controls differed only in the fact that one received BCG and the other received saline, the area between the two regression lines must represent that increment in tuberculin sensitivity which was induced by the vaccination procedure.

To determine whether the difference between the vaccinated and the controls was significant or whether such a difference could be explained by chance alone, correlation tables of reactions among the two groups were compared in a co-variant analysis. Where the difference was found to be significant, it is indicated in Fig. 6 by a shading of the area between the two lines.

Fig. 6 demonstrates that, although the increment in tuberculin sensitivity due to vaccination was not impressive, it was none the less significant at the 0.05 level or less among the younger age-groups and also among the adult females for which data were available. Non-significant differences were encountered among adult males only.

Table 4 shows the regression equations, the standard deviations for the scatter of the observations around these regression lines, the standard deviations for the slopes and the intercepts of the lines and, in comparing the results among vaccinated and controls, a summary of the co-variant analysis.

It is interesting to note that, in females and in the youngest group of males, no significant deviation from parallelism between the two regression lines could be calculated; they differed merely in intercepts, suggesting that the difference in reaction size between the vaccinated and the controls was much the same for any level of pre-vaccination tuberculin sensitivity. In the remaining groups of males, however, the lines were not parallel but differed in slope, suggesting a change in increment of tuberculin sensitivity related to changes in the degree of pre-vaccination tuberculin sensitivity.

The fact that 50% of those eligible for vaccination at the initial campaign were left unvaccinated as controls permits a comparison to be made between the size of the reactions to the first test among the non-vaccinated controls and that of the reactions to a second test given one year later under similar circumstances. Any demonstrable difference between the two observations must be due to differences in the techniques used (mode of injection, tuberculin dilution, reading, etc.) or in changes in the tuberculin reactor status of the individuals (enhancing effect of the first injection; super-infection with atypical and/or human tubercle bacilli; etc.).

In comparing first and second test results among the vaccinated and the controls (Fig. 6), it was necessary to take the first test as the independent variable, since its size determined the selection of subjects for vaccination. If, however, we wish to measure the difference between the first and the second test result among non-vaccinated persons only (that is, the controls as well as those not eligible for vaccination), we should not take the first reaction as the independent variable. In fact, it would be very inappropriate to do so, since there is no reason to consider one test result more reliable than another. Equal weight must therefore be attached to each of them and hence the mean k value $\left(\sqrt{\frac{\Sigma y^2}{\Sigma x^2}}\right)$ should be used for the calculation of the regression lines describing the relation between first and second test results.

If everybody had been tested twice and nobody had been vaccinated, it would have been simple to compare the two test results. One would have calculated the sum of each set of reactions (first plus

[1] Tables showing the data used for the calculation of the correlation coefficients and the regression lines, respectively, for the BCG-vaccinated and for the controls have been deposited in the WHO Library. Copies may be obtained on request. (See also footnote on page 464.)

TABLE 4

COMPARISON OF LINEAR REGRESSION IN REACTION SIZE BETWEEN FIRST AND SECOND TESTS WITH 1 TU OF RT 23 IN 0.1 ml (WITH TWEEN 80) AMONG THE VACCINATED AND THE CONTROLS, BY AGE AND SEX

Age	Regression equation		Standard deviation		F ratio	P value
			Vaccinated	Controls		
Males						
m²-MI	$y = 0.32x + 2.51$ Vaccinated Control $y = 0.31x + 0.87$	Scatter	3.38	1.97	$F\ {141 \atop 134}\ 2.92291$	< 0.001
		Slope	0.13	0.09	$F\ {1 \atop 275}\ 0.00131$	> 0.2
		Intercept	0.33	0.20	$F\ {1 \atop 276}\ 24.48646$	< 0.001
I	$y = 0.67x + 3.92$ Vaccinated Control $y = 0.23x + 2.91$	Scatter	4.41	3.81	$F\ {124 \atop 126}\ 1.34254$	$0.01 < P < 0.05$
		Slope	0.13	0.11	$F\ {1 \atop 250}\ 6.69487$	$0.01 < P < 0.05$
		Intercept	0.47	0.48		
M2	$y = 0.50x + 5.79$ Vaccinated Control $y = 0.65x + 3.37$	Scatter	4.53	4.41	$F\ {89 \atop 80}\ 1.05581$	> 0.2
		Slope	0.14	0.15	$F\ {1 \atop 169}\ 10.76368$	$0.001 < P < 0.01$
		Intercept	0.57	0.62		
18-44	$y = 0.46x + 11.82$ Vaccinated Control $y = 0.24x + 11.64$	Scatter	5.94	5.04	$F\ {24 \atop 19}\ 1.38738$	> 0.2
		Slope	0.31	0.24	$F\ {1 \atop 43}\ 3.29250$	$0.01 < P < 0.05$
		Intercept	1.57	1.56		
45 +	$y = -0.11x + 15.03$ Vaccinated Control $y = 0.63x + 8.87$	Scatter	4.32	6.37	$F\ {16 \atop 15}\ 2.17737$	$0.05 < P < 0.1$
		Slope	0.23	0.35	$F\ {1 \atop 31}\ 3.18522$	$0.05 > P > 0.1$
		Intercept	1.41	1.67	$F\ {1 \atop 32}\ 2.51172$	$0.1 > P > 0.2$
Females						
m2-MI	$y = 0.28x + 3.13$ Vaccinated Control $y = 0.33x + 1.78$	Scatter	3.47	2.08	$F\ {128 \atop 144}\ 2.80236$	> 0.001
		Slope	0.13	0.10	$F\ {1 \atop 272}\ 0.68454$	> 0.2
		Intercept	0.15	0.20	$F\ {1 \atop 273}\ 37.51493$	< 0.001
I	$y = 0.83x + 3.91$ Vaccinated Control $y = 0.67x + 2.97$	Scatter	3.97	3.59	$F\ {120 \atop 99}\ 1.22092$	$0.2 > P > 0.1$
		Slope	0.14	0.13	$F\ {1 \atop 219}\ 1.50403$	< 0.2
		Intercept	0.43	0.44	$F\ {1 \atop 220}\ 22.43152$	< 0.001
M2	$y = 0.40x + 7.33$ Vaccinated Control $y = 0.66x + 3.23$	Scatter	5.05	4.95	$F\ {103 \atop 101}\ 1.04017$	> 0.2
		Slope	0.16	0.15	$F\ {1 \atop 204}\ 0.98321$	> 0.2
		Intercept	0.64	0.62	$F\ {1 \atop 205}\ 27.75955$	< 0.001
18-44	$y = 0.60x + 7.18$ Vaccinated Control Data lost	Scatter	4.85			
		Slope	0.11			
		Intercept	0.62			
45 +	$y = 0.57x + 8.57$ Vaccinated Control $y = -0.14x + 7.91$	Scatter	5.53	7.00	$F\ {32 \atop 27}\ 1.60072$	> 0.2
		Slope	0.26	0.34	$F\ {1 \atop 59}\ 2.72688$	$0.2 > P > 0.1$
		Intercept	1.61	1.65	$F\ {1 \atop 60}\ 4.49790$	$0.01 < P < 0.05$

second test result) as well as their difference (second test result minus first) and made a distribution of differences for each sum of test results (Guld, 1953). The mean difference and its significance would then have been calculated for different levels of tuberculin sensitivity. Such levels are expressed better by the sum of the two reactions than by the reaction to either the first or the second tuberculin test.

On Noemfoor Island, 50% of people with reactions of 0-11 mm to the first tuberculin test were BCG-vaccinated. In comparing the first and second test results among the non-vaccinated (those not eligible for vaccination as well as the controls), we have to take this selective removal of a part of the distribution into account. If we were to ignore it, we should be attaching unequal, rather than equal, weight to the two test results.

As mentioned earlier, the persons to be vaccinated were randomly selected from among those who were eligible. It is therefore reasonable to assume that the vaccinated individuals would have reacted to a second test in much the same way as did the present controls if BCG had been withheld. On this assumption we have "reconstructed" the table of correlation between the first and second test results among the non-vaccinated people (including the controls), by multiplying the observations made within the range of the vaccination criterion by the weighting factor 2. An alternative procedure, leading to the same result, would have been to divide the observations made among those that were earlier excluded from vaccination by the same weighting factor of 2.

This reconstructed table has then been divided into two halves, one consisting of observations among persons with relatively strong sensitivity, the other among persons with relatively weak sensitivity to the two tests combined. People who had a combined test result of 27 mm or more were considered strong reactors, and those who had a combined result of 26 mm or less were considered weak reactors. This division point was arbitrarily chosen on the basis of a probit analysis of distributions of first, second and combined test results in a number of age-groups. Reconstructed tables (not presented here) were used for this analysis. According to these tables this division point seems to agree fairly well with the selected vaccination criterion of a reaction of 11 mm or less induration to the first test.

The selected division point is not more accurate in its separation between strong and weak reactors than any conventional division between these two based on the configuration of a frequency distribution of reactions according to size of induration. It would, however, seem to be the best criterion by which to divide the group as a whole into one class of persons who react strongly to tuberculin on two consecutive occasions and another of persons who respond differently. The class of strong reactors would seem to contain most of the individuals who possessed high-grade tuberculin sensitivity, presumably caused by infection with virulent tubercle bacilli, *prior to the initial campaign*. The class of weak reactors, on the other hand, would seem to contain most of the individuals who had weak or unstable sensitivity at the time of the initial campaign and were, to all intents and purposes, *not yet infected* at the time of the initial campaign. It seemed desirable to determine the possible difference between the results of the first and the second tests separately for the two groups.

Distributions of differences between the first and the second test results have been made separately for the two groups on the basis of the reconstructed tables. The mean difference has been subjected to a t test for significance, again separately for the weak and the strong reactors to tuberculin. In applying this t test, the number of degrees of freedom has been calculated on the basis of the *actual* number of observations, not on the numbers appearing in the reconstructed tables. The results of this calculation are presented in Table 5.

With the exception of one subgroup (males, dental age M2, difference between the two tests significant at the 0.02 level), no significant differences could be calculated for the strong reactors. Within the limits of normal error they reacted similarly. Among the weak reactors, however, the picture was different. The reaction to the second test was larger than that to the first in all groups of dental age I and above, except for the older males (45 years and over).

Fig. 7 portrays the regression equations for the controls with weak sensitivity. The mean k value $\left(\sqrt{\frac{\Sigma y^2}{\Sigma x^2}}\right)$ has been used for the calculation of the slopes of these lines. The diagonal of the correlation diagram ($y = x$) is indicated by a broken line. Whenever a significant increase in sensitivity was calculated by the t-test procedure, it has been indicated by a shading of the area enclosed between the diagonal and the regression line.

It is not possible to determine what causes these differences in testing results. They could reflect an unintentional variation in the testing technique (Wijsmuller, 1963). They might, howerer, reflect a

TABLE 5

DIFFERENCE BETWEEN FIRST AND SECOND TESTS AMONG NON-VACCINATED WEAK AND STRONG REACTORS TO TUBERCULIN

Dental age	Males				Females			
	Degrees of freedom (*n*-l)	Mean difference	SE [a]	P value	Degrees of freedom (*n*-l)	Mean difference	SE	P value
	Weak reactors							
m2-M1	139	0.07	0.17	NS [b]	148	0.23	0.16	NS
I	138	1.00	0.28	<0.001	104	1.14	0.25	<0.001
M2	89	2.32	0.39	<0.001	111	1.91	0.36	<0.001
18-44	31	4.84	1.13	<0.001	← Data lost →			
45+	30	1.28	1.43	NS	46	2.38	0.97	<0.02
	Strong reactors							
m2-M1	5	0.44	0.37	NS	4	1.40	1.58	NS
I	35	0.25	0.60	NS	27	0.14	0.59	NS
M2	34	1.37	0.55	<0.02	44	0.33	0.41	NS
18-44	267	0.52	0.68	NS	← Data lost →			
45+	197	0.28	0.29	NS	148	0.56	0.32	NS

[a] SE = standard error.
[b] NS = non-significant.

real change in the tuberculin-reactor status of the tested individuals, caused by the first test (enhancing effect) or by exposure to the factors that cause tuberculin sensitivity to develop naturally. Finally, a combination of these and/or other factors may be postulated.

The fact, however, that a change in tuberculin sensitivity could not be demonstrated among the very young (who do not yet possess a great deal of naturally acquired low-grade tuberculin sensitivity) or among the oldest males (who presumably possess whatever low-grade tuberculin sensitivity can be acquired naturally) leads us to suspect the possibility of an enhancing effect.

Such an enhancing effect of tuberculin testing on BCG-induced tuberculin sensitivity has been reported in the past (Magnus, 1957; Magnus & Edwards, 1955). Recently, it has been reported for high-grade tuberculin sensitivity (Ferebee & Mount, in press). The present data, showing a relation between pre- and post-vaccination tuberculin sensitivity as well as a difference between the results of the first and second tests among non-vaccinated persons with low-grade tuberculin sensitivity, suggest also that naturally acquired low-grade tuberculin sensitivity can be enhanced by tuberculin testing.

DISCUSSION

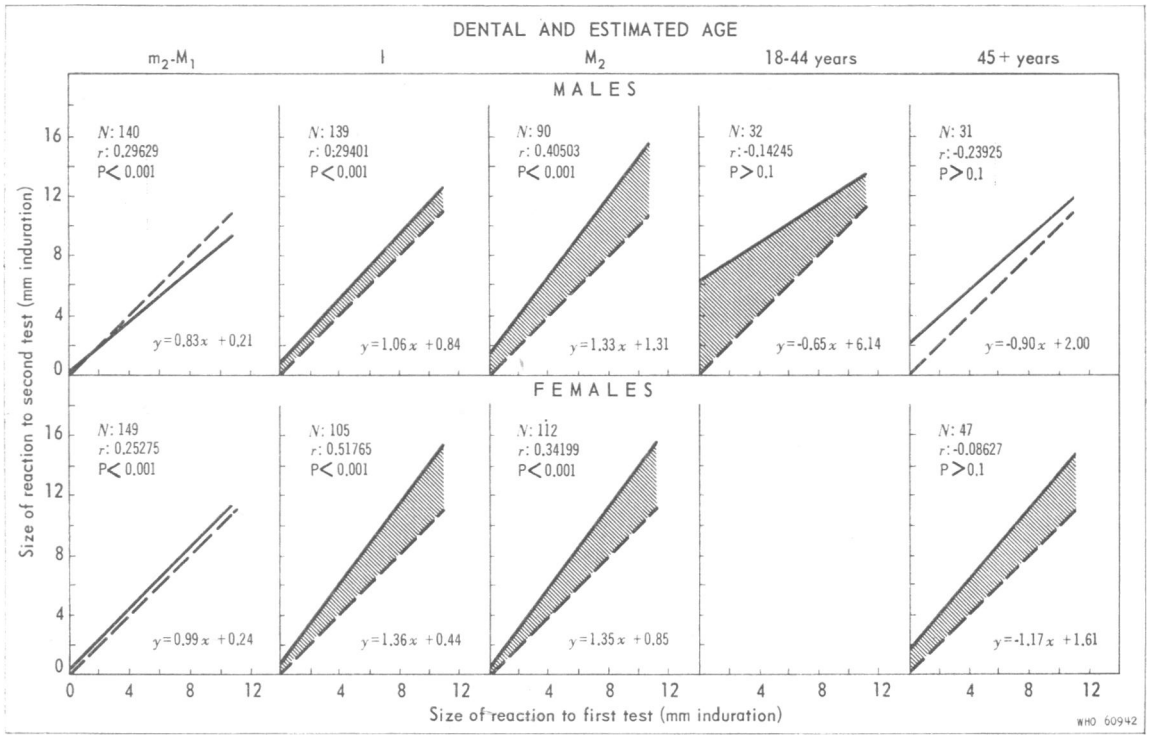
In many developing countries, tuberculosis is a major health problem; the extent of this problem is usually not known in precise terms. Facilities for diagnosis, treatment and follow-up are usually lacking, and the funds to create such facilities are not readily available. In an effort to make at least a start with a nation-wide antituberculosis drive, mass BCG vaccination has been adopted in many of these countries. Indeed, this approach is often regarded as the only one that is feasible, in terms of both cost and personnel.

A difficulty with programmes of this nature is that they cannot be evaluated satisfactorily because there are no non-vaccinated controls. Even if such controls were available, however, an evaluation of the programme would be very difficult because of the lack of diagnostic and other facilities.

While it has been impossible to assess BCG-induced protection in any mass campaign, it is often possible

FIG. 7

RELATION BETWEEN SIZE OF REACTIONS TO 1 TU OF RT 23 IN 0.1 ml (WITH TWEEN 80) AT INITIAL TEST AND AT A SECOND TEST ONE YEAR LATER AMONG NON-VACCINATED INDIVIDUALS WITH WEAK TUBERCULIN SENSITIVITY, BY AGE AND SEX



to measure the reaction to tuberculin some time after vaccination, if only on a selected sample of the population. This is commonly done, and it is assumed that the presence of a satisfactory level of tuberculin sensitivity some time after vaccination indicates that the campaign is achieving its objective—namely, to increase the resistance of the population against infections with virulent tubercle bacilli (Great Britain, Medical Research Council, 1960).

A positive correlation between BCG-induced tuberculin sensitivity and protection against challenge infections with virulent tubercle bacilli has been demonstrated in the guinea-pig (Tuberculosis Program, Public Health Service, USA, 1955a-1955d; 1957). Animals that developed more tuberculin sensitivity after vaccination survived the challenge infection longer than did animals that developed less sensitivity after vaccination with the same batch of BCG. Similarly, groups of animals vaccinated with a vaccine that induced strong sensitivity were better protected than animals that had been vaccinated with a vaccine that induced weak sensitivity.

Unfortunately, the relation between tuberculin sensitivity and immunity is not as clear in man as in animals. Earlier papers reporting the usefulness of BCG as a prophylactic measure in the control of tuberculosis have all been conducted in countries with temperate climates in which the prevalence of low-grade tuberculin sensitivity is low. In such countries, BCG vaccination is generally followed by the development of rather strong tuberculin sensitivity, which, in the absence of naturally acquired low-grade tuberculin sensitivity, must have been induced by the vaccination procedure. Unfortunately, most of these studies report on protection induced by BCG vaccination rather than on a possible relation between protection and tuberculin sensitivity. The degree of post-vaccination tuberculin sensitivity in a particular study is therefore not always known.

Recently, two large-scale studies have provided more precise information on this point. One of these

was performed in Britain (Great Britain, Medical Research Council, 1956, 1959, 1963), the other one in two different parts of the USA—namely, the Muscogee-Russell area (two adjacent south-eastern counties of Muscogee, Ga. and Russell, Ala.) and the island of Puerto Rico (Palmer, Shaw & Comstock, 1958). While naturally acquired low-grade tuberculin sensitivity is considered uncommon in Britain, it is highly prevalent in the two areas of the USA that were chosen for the BCG trial.

In the British study, post-vaccination tuberculin sensitivity was measured approximately 3-5 months after vaccination. At that time, 76-96% of the vaccinees reacted with an induration of 5 mm or more to 3 TU of Old Tuberculin (OT). Non-reactors to this test were further examined with 100 TU of OT. Essentially all the vaccinees who did not show a reaction to the low dose of 3 TU, reacted with an induration of 5 mm or more to the second dose of 100 TU. As only the non-reactors to 100 TU had been selected for vaccination initially, it is reasonable to assume that the vaccinees acquired their sensitivity through the vaccination procedure.

Vole vaccine was used in addition to BCG in this British study. Some of the earlier batches of vole vaccine contained substandard numbers of viable bacilli, and children vaccinated with such batches had substantially lower " conversion " rates when retested some time later than children who were vaccinated with the later batches of vole vaccine that contained a much higher number of viable bacilli. It was concluded that " these early batches, despite low conversion rates, thus afforded considerable protection against disease " (Great Britain, Medical Research Council, 1956).

In the US Public Health Service studies, two different criteria for vaccination were used. In Puerto Rico, the vaccination criterion was set at a reaction of 5 mm or less to 10 TU of PPD RT 19-21; in the Muscogee-Russell area, it was set at 4 mm or less to 5 TU of the same tuberculin. For practical purposes, these two criteria can be considered to be the same.

Both in Puerto Rico and in the Muscogee-Russell area, low-grade tuberculin sensitivity is highly prevalent (Palmer, Shaw & Comstock, 1958). The actual degree of prevalence was determined by giving potential vaccinees a final test with 100 TU of PPD. The selection for vaccination, however, was not based upon the reaction to this final test, but on the reaction to 5 or 10 TU. Samples of the population were retested for post-vaccination tuberculin sensitivity some time later. It is reported that 89% of the people vaccinated in Puerto Rico reacted with an induration of 6 mm or more to 10 TU 1-2 years after vaccination. In the Muscogee-Russell area, 54% of the vaccinees had reactions of 5 mm or more to 5 TU 2 years after vaccination.

It may be recalled, however, that reactors to the high dose of tuberculin who were non-reactors to the intermediate dose of 5 or 10 TU were considered eligible for vaccination. As low-grade tuberculin sensitivity is highly prevalent in the areas chosen for the US studies, substantial numbers of vaccinees must have been moderately sensitive to tuberculin before they were vaccinated. Consequently, we do not know what part of the observed post-vaccination tuberculin sensitivity is due to vaccination and what part can be explained by the degree of tuberculin sensitivity present before giving BCG. What we do know is that, despite the rather high degree of post-vaccination tuberculin sensitivity, the measured protection was only small; it remained far below what was found in the British studies. It would be extremely interesting to know whether this finding of an apparent lack of protection in the presence of a reasonable level of post-vaccination sensitivity is a function of the vaccine that was used or of the population that was treated. Surely, the British and US results lead us to suspect that tuberculin sensitivity and protection after BCG vaccination are not related in as simple a way as has been found in guinea-pig experiments.

In discussing the discrepancies between these two major studies (Great Britain, Medical Research Council, 1959), it has been suggested that the controls in the US study may have possessed some immunity against infections with virulent tubercle bacilli, because many of them possessed low-grade tuberculin sensitivity. It would be equally possible that pre-vaccination low-grade tuberculin sensitivity has diminished the effect of vaccination because of an associated specific immunity directed against the vaccine (Wijsmuller, 1959b). According to this second concept, the live vaccine would be inactivated, and a situation would develop as if the vaccinees had been injected with dead bacilli rather than with living ones. Either of these two mechanisms or a combination of them could explain the discrepancies in result between the British and the US studies. Such an interpretation would actually imply that the effect of BCG vaccination may depend upon geographic factors such as the presence or absence of low-grade tuberculin sensitivity. If this were the case, mass

vaccination programmes, using living vaccines, might not be very effective in reducing the incidence of tuberculous disease in areas where low-grade tuberculin sensitivity is highly prevalent.

To return to our own observations, a positive correlation between pre- and post-vaccination tuberculin sensitivity was always observed in children aged 2-11 years. This observation indicates that post-vaccination tuberculin sensitivity is only partially induced by the vaccination procedure. This inference is supported by observations made on Noemfoor Island, where non-vaccinated controls were available and the increment in tuberculin sensitivity due to vaccination and to other causes could be measured separately. A co-variant analysis of these data revealed a small but significant difference in sensitivity between vaccinees and controls in most age and sex groups. However, in the older males, the analysis failed to show any difference in sensitivity between the vaccinated and the controls, despite the fact that this age-group had exhibited the largest post-vaccination reactions.

It will be clear that, under such conditions, the degree of tuberculin sensitivity measured an appreciable time after vaccination cannot be a useful guide in assessing the conduct of the campaign. Post-vaccination tuberculin sensitivity becomes, at least partly, a function of pre-vaccination tuberculin sensitivity; hence the same vaccine, used among populations of the same ethnic and sociological background, can be expected to induce dissimilar levels of post-vaccination tuberculin sensitivity. Within the same population, post-vaccination tuberculin sensitivity will depend upon the age and sex composition of the vaccinated group.

The present data further suggest that, while post-vaccination tuberculin sensitivity as measured in the Sentanie district and on Noemfoor Island was generally low, it would have been lower still had the population possessed no pre-vaccination tuberculin sensitivity. It is possible that the apparent inability of the New Guinean to develop strong sensitivity after vaccination with BCG could be explained by deficiencies in the vaccination techniques that have been employed. Another possibility would be that the tuberculin dilutions were not of appropriate strength.

The latter possibility can be readily rejected, because the same tuberculin dilution elicited large reactions in persons who were proved to have a tuberculous infection (Wijsmuller, 1959a, 1959b, 1961, 1963). The reactions among tuberculous individuals were not smaller but rather larger than those normally found in comparable groups in Europe.

The first possibility, namely, that the vaccination technique is to be blamed, has also been scrutinized. It was demonstrated that the vaccine, which was produced in Europe, taken into the field half a world away and sent back to Europe under most difficult conditions of transportation and refrigeration control, had not changed in viability count as compared to control ampoules of the same batch stored under refrigeration at the point of origin (Wijsmuller & Schneider, 1962). In our attempts to induce tuberculin sensitivity through BCG vaccination, we have also, at annual intervals, retested and, if need be, revaccinated, entire populations (Wijsmuller, 1959b). We have failed, however, to induce in New Guineans tuberculin sensitivity of the same degree as can be produced in Danish schoolchildren by a single vaccination with the same BCG vaccine (Guld et al., 1958). As both the vaccine and the vaccination technique were of good quality, we must conclude that New Guineans differ from Danish schoolchildren in their ability to develop strong sensitivity after BCG vaccination. We do not know the reason for this difference between these two populations. Is it racial? Is it determined by nutritional factors? Has it to do with chronic infections or with a high prevalence of acute infections? At the moment, there is no way of knowing. However, two points seem to be clear, namely:

(1) New Guineans differ from Danes in their ability to develop strong sensitivity after vaccination; and

(2) post-vaccination tuberculin sensitivity in New Guinea (and presumably also in other countries where low-grade tuberculin sensitivity is prevalent) is a function of pre-vaccination tuberculin sensitivity. Consequently, a better conversion rate may merely indicate a difference in the prevalence of low-grade tuberculin sensitivity.

In the British study on BCG and vole vaccination, a high degree of protection was reported even in the absence of measurable post-vaccination tuberculin sensitivity (vole-vaccinated group). This seems to indicate that, while a high degree of post-vaccination tuberculin sensitivity in Britain may be taken as evidence of a successful infection with BCG bacilli, the absence of post-vaccination tuberculin sensitivity does not exclude the presence of BCG-induced immunity.

In the US Public Health Service studies, however, which were conducted among a population with a high prevalence of low-grade tuberculin sensitivity

the degree of post-vaccination tuberculin sensitivity was not greatly different from that measured in Britain. This observation applies particularly to the study in Puerto Rico, in which 89% of the vaccinees reacted with an induration of 6 mm or more to 10 TU of PPD 1-2 years after BCG vaccination. The degree of protection afforded by BCG vaccination, however, was substantially lower in Puerto Rico than in Britain. It thus appears that the relation between post-vaccination tuberculin sensitivity and immunity differs in these two populations.

In the present study, an apparent inability to develop strong sensitivity after vaccination was associated with a positive correlation between pre- and post-vaccination tuberculin sensitivity. This finding seems to suggest that BCG vaccination has enhanced an existing pattern of low-grade tuberculin sensitivity instead of inducing a pattern of its own. If this were the case, it is tempting to assume that the BCG bacilli might not have had a chance to multiply within the host after they were introduced, and as a result failed to mobilize the immune mechanism of the host. Could it be that such a mechanism has been operative in the US Public Health Service studies as a consequence of the high prevalence of low-grade tuberculin sensitivity among their vaccinees?

We do not know what level of protection, if any, has been induced in the people of New Guinea through the mass BCG-vaccination programme. What seems to be clear, however, both from the literature and from our own observations, is that the protection induced by a mass vaccination programme cannot be estimated through a study of post-vaccination tuberculin sensitivity. This observation would seem to apply particularly to countries where low-grade tuberculin sensitivity is highly prevalent. In view of this fact, and in recognition of the evidence that the presence of low-grade tuberculin sensitivity may be associated with partial immunity against infections with virulent tubercle bacilli (and possibly also against BCG), a further evaluation of BCG protection in areas where low-grade tuberculin sensitivity is highly prevalent is much needed.

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RÉSUMÉ

La présente enquête a été menée dans deux districts de l'ancienne Nouvelle-Guinée néerlandaise (actuellement Province d'Irian occidental de la République d'Indonésie). Des groupes importants de population ont été vaccinés à l'aide de deux préparations de BCG; les résultats des épreuves tuberculiniques pratiquées, avant et après administration du vaccin, avec de faibles doses de tuberculine ont été comparés. Un groupe témoin non vacciné a été soumis à des tests tuberculiniques identiques.

Une corrélation franche entre les valeurs de la sensibilité tuberculinique relevées avant et après vaccination a été observée chez les sujets âgés de 2 à 11 ans, indiquant que la sensibilité postvaccinale est fonction de l'allergie existant avant l'administration du BCG. L'accroissement de sensibilité apporté par la vaccination a été évalué en comparant les résultats des tests pré- et postvaccinaux chez les sujets vaccinés et dans le groupe témoin. Son importance est apparue significative chez les sujets jeunes des deux sexes ainsi que chez les femmes adultes, mais un effet identique n'a pu être démontré chez les hommes adultes. L'analyse des résultats des épreuves successives dans le groupe témoin a mis en évidence une augmentation de la sensibilité tuberculinique chez les sujets qui, initialement, présentaient de faibles réactions. L'auteur attribue ce phénomène à la répétition des tests et exclut une variation éventuelle dans les techniques utilisées ou l'influence de facteurs extérieurs capables d'accroître la sensibilité à la tuberculine dans une collectivité.

L'auteur analyse ensuite les résultats discordants observés lors d'études consacrées à la vaccination par le BCG aux Etats-Unis d'Amérique et en Grande-Bretagne. Il suggère que, au cours de certaines d'entre elles, intéressant des populations où un faible degré initial de sensibilité tuberculinique est fréquent, la vaccination par le BCG a renforcé la sensibilité préexistante plutôt que suscité par elle-même l'allergie. Dans de semblables collectivités, la

multiplication du BCG chez l'hôte est peut-être moins active que dans les populations où l'allergie est inexistante, et le vaccin ne peut assurer un degré de protection équivalent à celui qu'il apporte aux sujets exempts de toute allergie prévaccinale.

Selon l'auteur, on ne peut apprécier le succès d'un programme de vaccination antituberculeuse en se basant sur l'étude de la sensibilité tuberculinique postvaccinale; ceci est vrai notamment pour les pays où une fraction importante de la population témoigne d'une allergie de faible degré. Pratiquée dans des conditions identiques, la vaccination par le BCG donnera naissance à des niveaux de sensibilité tuberculinique variables suivant la fréquence de l'allergie prévaccinale au sein d'une population, d'où la nécessité de procéder à de nouvelles recherches sur la protection conférée par la vaccination antituberculeuse dans les pays où ce type d'allergie est particulièrement répandu.

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