CLINICAL EVALUATION STUDIES IN LEPROMATOUS LEPROSY

FIFTH SERIES: DIETHYL DITHIOLISOPHTHALATE (ETISUL) AS A SUPPLEMENT TO 4-4′ DIAMINODIPHENYL SULFONE (DDS)[1]

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In 1959 Davey and Hogerzeil ([4]) reported early improvement in cases of lepromatous leprosy treated with diethyl dithiolisophthalate (Etisul), an ester formed from isophthalic acid and ethyl mercaptan. The drug was incorporated in a perfumed medium and given by inunction. After the second month of treatment progress was variable, and the authors concluded that the possible acquirement of drug resistance by the leprosy bacillus removed Etisul from the list of basic remedies, but that, if the development of resistance could be delayed by combination with other drugs, Etisul might have considerable value. Davey and Hogerzeil cited an earlier study by Bertaccini ([1]), who had obtained as good results in lepromatous leprosy with a compound similar to Etisul, sodium ethyl thiosulfate (ET), as with other drugs.

Because of these reports Etisul was selected for trial in the fifth series of our clinical evaluation studies as a supplement to 4-4′ diaminodiphenyl sulfone (DDS). Through the courtesy of Dr. J. M. Mun-

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gavin a supply of the cream preparation was obtained as well as an equal quantity of a second cream closely resembling the first in color, consistency and odor, but containing no Etisul. These creams were designated No. 1 and No. 2.

A number of earlier reports had shown that certain mercaptan compounds exhibit antituberculosis activity. Recent reviews ([3]) make it unnecessary to give details of these studies. Del Pianto ([12]) of the University of Rome reported in 1953 that a mixture of sodium 2-mercaptobenzathiazole-S sulfonate and sodium ethyl thiosulfate prevented the development of tuberculosis in guinea-pigs inoculated with *M. tuberculosis*. In a later report ([13]) he stated that sodium ethyl thiosulfate was highly active in experimental tuberculosis in mice when incorporated in the diet, but was inactive when administered by the subcutaneous or intraperitoneal route. It was inactive *in vitro* against *M. tuberculosis*.

Brown *et al.* ([2]) and Solotorvsky and associates ([15]) of the Merck Research Laboratories reported in 1954 that, of more than 350 ethyl mercaptans, over 50 showed antituberculosis effectiveness when given to infected mice at a level of 0.2 per cent or less in the diet. Among these samples, S-ethyl-L-cysteine was noteworthy on the basis of both efficacy and low acute and chronic toxicity. It was more active than p-aminosalicylic acid and pyrazinamide, but less active than either streptomycin or isoniazid (INH). It was inactive *in vitro*. Extension of these experiments ([14]) showed the interesting fact that S-ethyl-L-cysteine was inactive when the mice were kept in an open pan, but highly active when closed jars were used. Evidently it yielded a volatile component that was active against tubercle bacilli *in vitro*. This component was demonstrated to be ethyl mercaptan. Ethyl mercaptan, as well as methyl mercaptan, inhibited *M. tuberculosis* H37Rv *in vitro* in high concentrations.

Kushner *et al* ([11]) of the Lederle Research Laboratories in 1955 studied the antituberculosis activity of a series of mercaptan compounds, and found that the tuberculostatic action of these compounds, many of which were ethylthiol esters, resided in ethyl mercaptan itself. These compounds corresponded in activity to streptomycin given by the parenteral route and were more effective than pyrazinamide.

Davis *et al* ([6]) of the Imperial Chemical Industries Research Laboratories reported in 1956 the results of a systematic study of the activity of about 200 mercaptans and related compounds, in mice infected intravenously with *M. tuberculosis*, human type, strain 905. The drugs were given orally twice daily, or subcutaneously once daily, for two weeks to groups of 6 to 10 mice. It was concluded that ethyl mercaptan exerts a profound effect on the progress of tuberculosis in mice. All compounds that were expected to yield ethyl mercaptan, either before or after absorption, showed antituberculosis activity. Activity was specifically dependent upon the presence of the ethyl radical, even closely related thiols being completely inactive.

Because of the unpleasant odor of ethyl mercaptan, it was considered desirable that a therapeutically acceptable compound should release the thiol only after being absorbed. Thiol-esters were found to be suitable and were invariably more active when administered parenterally than when given orally. This work led to the discovery of diethyl dithiolisophthalate (ETIP or Etisul), the antituberculosis activity of which was reported by Davies and Driver ([6]) in 1957. When doses of 500 mgm. per kg. of Etisul were given subcutaneously to mice for 10 days, all survived 133 days; living tubercle bacilli could, nevertheless, be recovered. Untreated controls, on the other hand, had a mean survival time of only 17 days. Etisul was effective also in experimental tuberculosis of mice when applied as a paste to the shaved skin in a dosage of 10 mgm. per kg. Resistance to Etisul was produced rapidly.

METHODS AND MATERIALS

Organization.—As in the second, third and fourth series of these studies, volunteers were enlisted at the Central Luzon Sanitarium, situated about 20 miles north of Manila, and at the Eversley Childs Sanitarium, about 8 miles north of Cebu City. At each institution a leprologist, nurses, technicians and a clerk were assigned exclusively to the study.

Technical procedures were standardized. These included the dosages and methods of administration of DDS and of the two preparations to be given by inunction, as well as clinical and laboratory examinations, photography and record-keeping.

Physical examinations.—Regular dermatologic and neurologic examinations were made prior to therapy and at bimonthly intervals thereafter. Special examinations were made when indicated by worsening of signs or symtoms. The nose and throat examinations were made with the aid of a head mirror, and one of us (J.G.T.) was responsible for all such examinations at both institutions. In addition to being recorded on prescribed forms, the findings were depicted graphically on dermatologic and neurologic charts. One of the authors (J.G.T.), in the capacity of a nonresident consultant, examined all patients at the Central Luzon Sanitarium prior to therapy and at the end of 24 and 56 weeks; another (J.N.R.), in the same capacity, examined the patients at the Eversley Childs Sanitarium on identical occasions.

Bacteriologic examinations.—Separate smears were made from eight sites: right and left sides of the nasal septum, right and left earlobes, and four selected optional skin sites. These examinations were made on three occasions, during the preliminary period before therapy, at the end of 24 weeks of therapy with DDS (16 weeks after the inunctions were commenced) and at the end of 56 weeks of therapy with DDS (48 weeks after the start of the inunctions).

Photographic records.—Several black and white and color photographs were made of each patient prior to therapy and at the end of 56 weeks.

Other examinations.—Hemoglobin values, packed red-cell volume, erythrocyte counts, levels of sulfone in the blood, and presence or absence of albumin, sugar, casts, urobilinogen and bile in the urine, were determined prior to therapy at various regular intervals throughout the study. Lepromin and tuberculin (5 TU PPD) tests were made prior to and at the end of therapy. The blood protein pattern also was studied by the method of paper electrophoresis prior to and at the end of the study.

Selection of patients and assignment to groups.—At the Central Luzon Sanitarium 122 patients were selected for the experiment. At Eversley Childs 79 were selected in what was termed the "first batch" and 47 in the second. All patients were originally diagnosed as lepromatous in type, but later 15 were reconsidered as more correctly classifiable as borderline, or rather as being on the borderline side of lepromatous. All patients were bacteriologically positive and lepromin-negative at the outset.

As in the previous series of these studies, an index card was prepared for each patient, on which was entered a code name, date of birth, sex, previous therapy (if any), year of onset, date of admission, stage of disease, presence or absence of infiltration, nodules and certain other signs, height, weight and some other miscellaneous items. These cards were airmailed to the Medical Director of the Leonard Wood Memorial in Washington, D.C.

The plan adopted required assignment of the patients into two groups, "AA" and "BB." The method of assignment to groups was that used in the preceding four series. The index cards for each institution (and for each batch at Eversley Childs) were arranged in order of decreasing age. The two oldest males were then assigned either to group AA or group BB, using a table of random numbers, then the

TABLE 1.—*Pretherapy status of patients in the fifth series, with respect to certain characteristics possibly related to prognosis, by therapy group and institution.*

Characteristic of therapy group	Central Luzon			Eversley Childs		
	BB (DDS and Etisul)	AA (DDS and control ointment)	Both groups	BB (DDS and Etisul)	AA (DDS and control ointment)	Both groups
No. of patients	61	61	122	63	63	126
Average:						
Age (years)	27.4	26.1	26.8	28.1	27.1	27.6
Weight (lbs.)	98.7	97.8	98.2	100.0	99.0	99.5
Bact. score: nasal	6.4	6.9	6.6	9.2	8.8	9.0
skin	19.6	20.0	19.8	22.4	22.3	22.4
Percentage:						
Male sex	65.1	64.5	64.8	80.9	82.5	81.7
L1 or L2 class	52.4	61.3	56.8	76.2	69.8	73.0
Previously untreated[a]	100.0	100.0	100.0	95.2	96.8	96.0

[a] Includes all patients who had either received no DDS previously or a total of less than 5 gm.

second lot of two males, and so on through the rest of the males and females. Frequency distributions were prepared for the groups at each institution for stage of leprosy, preliminary bacteriologic score and a number of other items. After examination of these distributions, a few changes were made to achieve better balance. Certain characteristics of the groups are given for all entering patients in Table 1. The names of the patients selected for each group were then airmailed to each institution.

Plan of treatment and dosages.—It was decided to give all patients a maximum daily DDS dose of 2 mgm. per kg. of body weight, reached over an induction period of eight weeks. Beginning with the ninth week the inunctions were given three times weekly, the dosage on each occasion being 5 gm. for Etisul and the control. The rubbing was done on the back and maintained for a minimum of 20 minutes, patients rubbing one another under supervision. All of the patients at Central Luzon and the first batch at Eversley Childs commenced DDS therapy on March 14, 1960, and the inunctions on May 9. The second batch at Eversley Childs commenced DDS therapy on May 30 and the inunctions on July 15. Therapy was continued for 56 weeks from the commencement of DDS administration; i.e., the inunctions were given for 48 weeks.

It was determined by lot that the Etisul cream (in tubes labeled cream No. 1) would be given to patients of group BB, and the control ointment (in tubes labeled cream No. 2) to those of group AA. This fact, however, was not made known to any of the investigators at Central Luzon and Eversley Childs. All persons concerned with the treatment and evaluation of the results remained ignorant of the identity of the creams until the final conclusions of the 56th week examinations had been recorded.

TABLE 2. *Average amounts of DDS and inunctions per patient, prescribed and actually taken, for those completing 56 weeks of therapy (48 weeks for the inunctions), by institution and therapy group.*

Therapy group	Number of patients	DDS			Etisul and control inunction		
		Ave. amount per pt. (gm.)		Per cent taken	Ave. amount per pt. (gm.)		Per cent taken
		Prescribed	Taken		Prescribed	Taken	
Central Luzon Sanitarium:							
BB	53	26.4	18.8	71.4	700.0	451.8	64.5
AA	47	26.2	19.3	73.5	700.0	483.1	69.0
Total	100	26.3	19.0	72.3	700.0	466.5	66.5
Eversley Childs Sanitarium:							
BB	51	26.7	25.6	95.7	700.0	680.1	97.2
AA	51	25.8	24.4	94.7	700.0	660.4	94.3
Total	102	26.3	25.0	95.2	700.0	670.2	95.8

TABLE 3. *Numbers of patients selected at Central Luzon and Eversley Childs, classified according to treatment status at the end of 24 weeks and 56 weeks.*

	24 weeks DDS (16 weeks Etisul or control inunction)				56 weeks DDS (48 weeks Etisul or control inunction)			
	Central Luzon		Eversley Childs		Central Luzon		Eversley Childs	
Therapy status	BB	AA	BB	AA	BB	AA	BB	AA
Therapy complete	56	55	58	57	53	47	51	51
Therapy incomplete:								
Absconded	4	6	5	6	6	11	10	11
Died	1				2	2		1
Refused treatment							1	
Allergic to DDS							1	
Transferred (Culion)						1		
Original total	61	61	63	63	61	61	63	63

Deaths. Central Luzon: BB, #51-7-49, pulmonary tuberculosis, 18th week (7/12/60).
BB, #57-82-50, nephrosis, 47th week (2/12/61).
AA, #57-106-25, pneumonia, 40th week (12/12/60).
AA, #56-72-20, undetermined, 3/19/61. Patient left on pass 11/26/60 (40th week), died at home; cause apparently unrelated to leprosy.
Eversley Childs: AA, #60-102, gastric hemorrhage, 37th week (12/26/60).

Dosages prescribed and actually taken.—Chiefly because of reactions, the patients frequently asked to have therapy suspended temporarily or to have the quantity reduced. This resulted in a lowering of dosage, especially at the Central Luzon Sanitarium. At Eversley Childs the proportions taken of the total quantities prescribed were very high, considering the duration of the experiment. The record for each group and each institution is given in Table 2.

Dropped patients.—In all long term studies of any considerable number of patients in which losses are inevitable, one of the principal difficulties is to be certain that patients who are "dropped" do not affect the comparability of the groups in respect to any factor that may be related to prognosis. As in the previous four series, the principal cause of leaving the study was departure from the institution without permission or failure to return on time when permission for a short leave had been granted.

Four deaths occurred at Central Luzon, none of which was caused directly by leprosy. One was attributed to pulmonary tuberculosis, 1 to pneumonia and 1 to nephrosis. The cause was undetermined in the fourth, as the patient went out on pass and died at home. There was only one death at Eversley Childs; this was caused by hemorrhage from a gastric or duodenal ulcer. Autopsy was not performed in any of the above instances.

One patient refused to continue treatment and another was unable to continue on DDS because of a resulting severe allergic dermatitis. Both of these patients were at Eversley Childs.

The numbers of patients originally selected and completing 24 and 56 weeks of therapy are shown for each institution and each group in Table 3, those dropped being classified as to reasons for that action.

Comparison was made of the dropped patients with those completing the study with respect to those characteristics observed on preliminary examination that seemed probably related to prognosis. The average bacteriologic score, the percentage of cases classed as L2 and L3 and other characteristics were investigated. No significant differences were found in the frequency of any of these characteristics among patients, taken as a group, who were examined and those who were not examined (dropped) at 24 weeks and at 56 weeks, either at Central Luzon or at Eversley Childs.

Since one of the most important of these characteristics presumably is the "heaviness" of the bacterial load, the average preliminary bacteriologic scores for patients who completed and for those who did not complete 56 weeks of therapy (48 weeks of the inunctions) are given in Table 4. Obviously failure to be examined at the end of therapy was not related to the degree of bacteriologic positivity at the start of the study.

TABLE 4. *Average pretherapy bacteriologic scores (2 nasal and 6 skin sites) per patient, for those completing and not completing 56 weeks of therapy, by therapy group and institution.*

Therapy group	Sites	Central Luzon		Eversley Childs	
		Completing 56 weeks	Not completing	Completing 56 weeks	Not completing
BB (DDS plus Etisul)	Nasal	6.1	7.0	9.3	8.6
	Skin	19.8	21.2	22.2	20.2
AA (DDS plus control)	Nasal	6.6	8.5	8.9	8.3
	Skin	19.8	21.3	22.7	20.8
Both groups	Nasal	6.4	7.8	9.1	8.5
	Skin	19.8	21.3	22.5	20.5
	All sites	26.2	29.1	31.5	29.0

CLINICAL CHANGES

The difficulties of clinical appraisal of the value of drugs in leprosy have been discussed in previous reports. These difficulties notwithstanding, we feel that in comparison of carefully matched groups of patients by experienced and unbiased leprologists, it is unlikely that significant advantages of one therapy over another will escape detection. Essentially the same methods were used in this study as before. At each physical examination, the consultant was asked to give a numerical rating to the degree and extent of diffuse infiltration, nodulation and a number of other lesions for various regions of the body. If any lesion was not present, its absence was recorded. These ratings were added together for each type of lesion and used for comparison with results at later examinations. Neurologic and dermatolo-

gic charts of the patient's body, Kodachrome transparencies and black-white photographs made before therapy and again at the end of the study, were used by the consultant to supplement the written clinical records. On completion of each examination, the consultant summarized his opinion as to progress of the case, using the following grades: *improved* (marked, moderate or slight), *stationary* or *worse* (slight, moderate or marked). Examination of the detailed ratings of the more significant lesions shows support for the consultant's final opinion in nearly all instances. This is well illustrated by the records of patients regarded as having become worse.

After 24 weeks of therapy (16 weeks for the inunctions) at Central Luzon, the disease was recorded as worse in the following 4 patients of the Etisul group (BB) and 5 of the control group (AA):

BB, #55-89-12, female, 12 years of age. Appearance of ulceration of nasal septum (graded ++) and of infiltration of septal mucous membrane.

BB, #54-96-25, female, 25 years of age. New plaques on trunk (graded +++). Increased size of plaques on extremities and increase of infiltration of skin of buttocks.

BB, #56-104-22, female, 22 years. Increased infiltration of the skin of the ears, trunk and buttocks.

BB, #54-49-29, male, 29 years. New macules on buttocks; increase in infiltration and in ulceration of septum.

AA, #56-92-17, female, 17 years. Definite but slight increase in infiltration in several regions.

AA, #54-6-26, male, 26 years. Appearance of new plaques (graded +++).

AA, #59-52-15, male, 15 years. Definitely increased infiltration, trunk, buttocks, and extremities.

AA, #58-45-45, male, 45 years. Appearance of infiltration on lobes of ears and of new plaques on face; increased size of plaques on trunk and extremities.

AA, #59-116-21, female, 21 years. Increased infiltration of ears, buttocks and extremities; appearance of new plaques on trunk, buttocks and extremities (all graded ++).

At Eversley Childs, after 24 weeks, no patient of the Etisul group was recorded as worse. There were 2 such instances in the control group:

AA, #59-20, male, 14 years. New plaques on the buttocks and extremities. Development of kerato-conjunctivitis.

AA, #60-045, female, 20 years. Increased infiltration of face, buttocks and extremities.

At the Central Luzon Sanitarium, after 56 weeks of therapy (48 weeks of inunctions), 3 patients of the Etisul group and 1 of the control group were recorded as worse.

BB, #54-24-44, male, 44 years. New ulceration of nasal septum. Definite increase of infiltration, trunk, buttocks and extremities.

BB, #54-96-25, female, 25 years. Plaques recorded at 24 weeks still present; infiltration on face and trunk definitely greater than on preliminary examination.

BB, #55-89-12, female, 12 years. Nasal septum ulceration observed at 24 weeks still present. Infiltration of face, ears and extremities definitely more advanced than on preliminary examination.

AA, #56-92-17, female, 17 years. Definite increase of infiltration in several regions.

At Eversley Childs, after 56 weeks of therapy (48 weeks of inunctions), 1 patient of the Etisul group and none of the control were recorded as worse.

BB, #60-005, male, 27 years. Developed new lesions, irregularly shaped plaques above both knees and elbows.

A statistical summary of the clinical appraisals is given in Table 5, which shows the percentages of the patients of each therapy group rated as improved, stationary or worse after 24 weeks and 56 weeks of therapy at each institution.

The principal fact brought out in Table 5 is that at either institution, the proportion of patients showing clinical evidence of improvement was not higher for the Etisul group (BB) than the group receiving control inunctions (AA), either after 24 weeks (16 weeks of inunctions) or after 56 weeks (48 weeks of inunctions) of therapy. At both institutions, in fact, the trend at 56 weeks was in the other direction and in favor of the control group, but this is considered to be a chance variation.

Effect of treatment on specified lesions. As mentioned before, the consultants gave a numerical rating to the extent and degree of diffuse

TABLE 5.—*Percentages of patients classified by the consultants as improved, stationary or worse after completing 24 weeks and 56 weeks of treatment, by therapy group and institution.*

Therapy group	24 weeks of therapy				56 weeks of therapy			
	Number of patients	Per cent			Number of patients	Per cent		
		Improved	Stat.	Worse		Improved	Stat.	Worse
Central Luzon Sanitarium:								
BB (DDS + Etisul)	56	46.4	46.4	7.1	53	41.5	52.8	5.7
AA (DDS + control)	55	45.5	43.6	10.9	47	63.8	34.0	2.1
Total	111	45.9	45.0	9.0	100	52.0	44.0	4.0
Eversley Childs Sanitarium:								
BB (DDS + Etisul)	58	31.0	68.9	—	51	49.0	49.0	2.0
AA (DDS + control)	57	31.6	64.9	3.5	51	58.8	41.2	—
Total	115	31.3	67.0	1.7	102	53.9	45.1	1.0
Both institutions:								
BB (DDS + Etisul)	114	38.6	57.9	3.5	104	45.2	51.0	3.8
AA (DDS + control)	112	38.4	54.5	7.1	98	61.2	37.8	1.0
Total	226	38.5	56.2	5.3	202	53.0	44.5	2.5

infiltration, nodules, plaques and other lesions for different regions of the body (face, ears, trunk, buttocks and extremities). These ratings were added and the totals for successive examinations were compared in an attempt to measure the effect of therapy on each specified lesion.

Infiltration: as in the four previous series, diffuse infiltration was the one universal sign in the patients. Measurement of slight changes in degree or extent of infiltration is always more or less subjective. The recorded changes are shown in Table 6 for each therapy group at each institution.

Inspection of Table 6 reveals no evidence that supplementation of DDS therapy with Etisul was beneficial. The apparent higher rate of improvement in the control group at Central Luzon is obviously a chance finding, and illustrates the desirability of conducting a clinical evaluation at more than one institution. The greater improvement in both groups at Eversley Childs than at Central Luzon may have been due to the appreciably higher proportion of the prescribed dosage of DDS taken by the Eversley Childs patients.

TABLE 6.—*Numbers and percentages of patients of each therapy group recorded as improved with respect to diffuse infiltration after 56 weeks of treatment at each institution.*

Therapy group	Central Luzon			Eversley Childs		
	Number		Per cent improved	Number		Per cent improved
	Examined	Improved		Examined	Improved	
BB (DDS + Etisul)	53	25	47.2	51	39	76.5
AA (DDS + control)	47	29	61.7	51	38	74.5
Total	100	54	54.0	102	77	75.5

Nodules: nodular skin lesions were present at the outset in 25.2 percent of all patients in the study. Table 7 gives numbers and percentage of patients with and without nodules on preliminary examination, in particular relation to the outcome of such lesions after 56 weeks of treatment with DDS and 48 weeks of inunctions, by institution and therapy group. The figures clearly show no evidence that supplementing DDS with Etisul was beneficial with respect to the healing or disappearance of nodular skin lesions. As a matter of fact, they point in the opposite direction although this finding is probably again attributable to chance.

Lepromatous ulcers: as already mentioned, all nose and throat examinations were made by one of us (J.G.T.), using a head mirror and reflected light. As shown in Table 8, ulceration of the nasal septum was recorded on preliminary examination at Central Luzon in 37.7 per cent of the patients in the Etisul group and in 34.0 per cent of those in the control group; at the end of 56 weeks the percentage was 34.0 for each group. At Eversley Childs, nasal ulceration was recorded at the outset in 27.5 per cent of the Etisul group and in 15.7 per cent

TABLE 7.—*Numbers and percentages of patients with and without nodular skin lesions on preliminary examination in relation to status of such lesions after 56 weeks of therapy, by therapy group and institution.*

Therapy group	Number of patients	Nodules *present* on preliminary examination				Nodules *absent* on preliminary examination		
			Status at 56 weeks				Status at 50 weeks	
		Number	Absent	Improved	Stat. or worse	Number	Absent	Present (new)
Central Luzon Sanitarium:								
BB (DDS + Etisul)	53	14	2	5	7	39	39	—
AA (DDS + control)	47	6	1	4	1	41	41	—
Total	100	20	3	9	8	80	80	—
Eversley Childs Sanitarium:								
BB (DDS + Etisul)	51	17	6	7	4	34	33	1
AA (DDS + control)	51	14	4	7	3	37	35	2
Total	102	31	10	14	7	71	68	3
Both institutions:								
BB (DDS + Etisul)	104	31	8 (22.8%)	12 (38.7%)	11 (35.5%)	73	72 (98.6%)	1 (1.4%)
AA (DDS + control)	98	20	5 (25.0%)	11 (55.0%)	4 (20.0%)	78	76 (97.4%)	2 (2.6%)
Total	202	51 (25.2%)	13	23	15	151 (74.8%)	148	3

of the control; at the end of 56 weeks the respective percentages were 26.0 for the Etisul and 9.8 for the control. Obviously supplementation of DDS with Etisul was not beneficial in the healing of nasal ulcerations.

It has been noted in our previous reports ([12, 13, 14, 15]) that on the average, healing of nasal ulcerations takes much longer than is usually supposed and also that new ulceration may occur under continuous sulfone therapy. The same observations were made in the present series, as seen in Table 8. There was no evidence that the Etisul group fared any better than the control either in the healing of old ulcers or in the prevention of new ones.

Lepromatous ulceration of the skin was present at the beginning in only 10 patients at both institutions, 6 in the Etisul group and 4 in the control. Healing took place in all except one patient in the Etisul group. New ulceration, however, developed during therapy in 4 patients, 1 in the Etisul group and 3 in the group receiving the control inunctions.

Keratoconjunctivitis: this complication was found on preliminary examination in 18 patients at both institutions, 14 in the DDS-Etisul

group and 4 in the DDS-control group. The condition cleared up in 10 of the 14 in the former and in 3 of the 4 in the latter. It occurred during therapy in 1 patient of the DDS-Etisul group and in 3 patients of the DDS-control group, in all of whom this condition was absent at the outset.

Neurologic changes: no significant changes in extent or degree of anesthesia could be related to any of the two therapy groups in the study.

BACTERIOLOGY

The bacteriologic procedures adopted in the four previous series were adhered to in the fifth. As has been mentioned, smears were required from both sides of the nasal septum, both earlobes and 4 optional skin sites. The most marked or active lesions were selected as optional skin sites, and subsequent smears were made from approximately the same places. The present analysis compares the changes that took place in the two therapy groups in respect to nasal and skin sites, considered separately and together, between the preliminary examination and those made after 24 and 56 weeks of therapy (16 weeks and 48 weeks, respectively, for the injections).

There were no significant differences between Central Luzon and Eversley Childs with respect to the properties of patients of either

TABLE 8.—*Numbers and percentages of patients with ulceration of the nasal septum on preliminary examination and after 56 weeks of therapy (48 weeks for Etisul), by therapy group and institution.*

Therapy group	Number of patients	With nasal ulceration				
		Preliminary examination		56 weeks of therapy		
		Number	Per cent	Old	New	Per cent
Central Luzon Sanitarium:						
BB (DDS + Etisul)	53	20	37.7	12	6	34.0
AA (DDS + control)	47	16	34.0	13	3	34.0
Total	100	36	36.0	25	9	34.0
Eversley Childs Sanitarium:						
BB (DDS + Etisul)	51	14	27.5	9	4	26.0
AA (DDS + control)	51	8	15.7	4	1	9.8
Total	102	22	21.6	13	5	17.6
Both institutions:						
BB (DDS + Etisul)	104	34	32.7	21	10	32.7
AA (DDS + control)	98	24	24.5	17	4	20.2
Total	202	58	28.7	38	14	25.7

TABLE 9.—*Nasal and skin sites becoming bacteriologically negative after 24 weeks (16 weeks for Etisul) and 56 weeks (48 weeks for Etisul) of therapy, both institutions combined.*

Preliminary examination		24 weeks of therapy			56 weeks of therapy		
		Positive	Negative	Total	Positive	Negative	Total
2 nasal sites:							
BB (DDS + Etisul)	Positive	102	7	109	86	13	99
	Negative	—	5	5	1	4	5
	Total	102	12	114	87	17	104
AA (DDS + control)	Positive	99	6	105	79	15	94
	Negative	1	4	5	—	4	4
	Total	100	10	110	79	19	98
6 skin sites:							
BB (DDS + Etisul)	Positive	111	3	114	97	7	104
	Negative	—	—	—	—	—	—
	Total	111	3	114	97	7	104
AA (DDS + control)	Positive	105	5	110	88	10	98
	Negative	—	—	—	—	—	—
	Total	105	5	110	88	10	98

Note: *Two nasal sites.* In group BB (Etisul), 7/109 or 6.4% became bacteriologically negative at 24 weeks; and 13/99 or 13.1% at 56 weeks.
In group AA (control), 6/105 or 5.7% became bacteriologically negative at 24 weeks; and 15/94 or 15.8% at 56 weeks.
Six skin sites. In group BB, (Etisul), 3/114 or 2.6% became bacteriologically negative at 24 weeks; and 7/104 or 6.7% at 56 weeks.
In group AA, (control), 5/110 or 4.5% became bacteriologically negative at 24 weeks; and 10/98 or 10.2% at 56 weeks.

therapy group becoming *negative* after 24 weeks and 56 weeks of therapy in either nasal or skin sites. The results at both institutions have therefore been combined in Table 9.

As regards nasal sites, 7 of 109, or 6.4 per cent, of the patients of the Etisul group became negative after 24 weeks, and 13 of 99, or 13.1 per cent, became negative after 56 weeks. Of the patients receiving the control inunctions, 6 of 105, or 5.7 per cent, became negative after 24 weeks and 15 of 94, or 15.8 per cent, after 56 weeks. There was thus no significant difference between the groups with respect to the proportion of nasal sites becoming negative.

The findings for the skin are similar. Of the patients in the DDS-Etisul group, 3 of 114, or 2.6 per cent, became negative after 24 weeks and 7 of 104, or 6.7 per cent, were negative at the end of 56 weeks. In the DDS-control inunction group, 5 of 110, or 4.5 per cent, became negative at 24 weeks and 10 of 98, or 10.2 per cent, at 56 weeks.

Bacteriologic improvement short of negativity occurred in both groups at both institutions. In studying this aspect our usual practice of giving each smear an arbitrary numerical rating was followed. Smears marked v.s. (very scanty) were given a grade of 1, those marked + a grade of 2, ++ a grade of 3, +++ a grade of 4, and

TABLE 10.—*Average percentage reductions from pretherapy bacteriologic score, for nasal and skin sites, after 24 weeks (16 weeks for Etisul) and 56 weeks (48 weeks for Etisul) of therapy, by therapy group and institution.*

Therapy group	24 weeks of therapy (16 weeks for Etisul & control inunction)			56 weeks of therapy (48 weeks for Etisul & control inunction)		
	Number of patients	Average percentage reduction		Number of patients	Average percentage reduction	
		2 nasal sites	6 skin sites		2 nasal sites	6 skin sites
Central Luzon Sanitarium:						
BB(DDS + Etisul)	56	11.8	8.2	53	35.9	30.7
AA(DDS + control)	53	9.9	6.1	47	47.1	31.9
Total	109	10.8	7.1	100	41.3	31.3
Eversley Childs Sanitarium:						
BB(DDS + Etisul)	58	3.9	7.0	51	18.8	27.6
AA(DDS + control)	57	9.9	9.5	51	15.1	29.7
Total	115	6.3	8.6	102	17.1	28.7
Both institutions:						
BB(DDS + Etisul)	114	7.1	7.5	104	26 0	29.1
AA(DDS + control)	110	9.5	8.4	98	28.1	30.7
Total	224	8.7	8.9	202	27.0	29.9

++++ a grade of 5. The scores for the nasal sites and those for the skin were studied separately. It should be kept in mind that, as in the previous four series, our principal purpose was a comparison of groups of patients under different therapies. The practice of occasional blind exchanges of slides between technicians and institutions was continued. The results, as before, showed good agreement in the grading of smears; a difference of more than one step was very unusual. For both institutions separately and combined, the average percentage reductions in bacteriologic scores for nasal and skin sites after 24 weeks and 56 weeks of therapy between the DDS-Etisul group and the DDS-control inunction group are shown in Table 10.

The percentages of reduction in bacteriologic scores from the pretherapy level were very close for the two therapy groups at both institutions. For both institutions combined, the Etisul patients showed an average reduction in nasal smears of 7.1 per cent and 26.0 per cent after 24 weeks and 56 weeks, respectively. The corresponding average reductions in nasal smears of patients in the control group were 9.5 per cent and 28.1 per cent. For the skin sites, the Etisul patients showed an average reduction of 7.5 per cent after 24 weeks and 29.1 per cent after 56 weeks; the corresponding reductions in the control group were 8.4 per cent and 30.7 per cent. In view of the opinion held by some observers that Etisul is particularly effective in the early weeks of therapy, it is of interest that evidence of this did not appear

TABLE 11.—*Association between clinical and bacteriologic improvement at the end of 56 weeks, therapy groups combined, both institutions.*

Final clinical appraisal	Reduction in bacteriologic scores for 8 sites (6 skin & 2 nasal)			
	50% or more	Less than 50%	Total	Per cent reduced 50% or more
Improved	37	70	107	34.6
Stationary or worse	17	78	95	17.9
Total	54	148	202	26.7
Per cent improved	68.5	47.3	53.0	

TABLE 12.—*Relative frequency of the occurrence during therapy of various reactional manifestations, by therapy group, both institutions combined, for patients completing 56 weeks of treatment.*

Therapy group	Number of patients	Average number of positive weekly observations for various signs and symptoms of reaction					
		Erythema nodosum leprosum	Plaques	Acute infiltration	Fever	Neuritis	Conjunctivitis or iritis
BB(DDS + Etisul)	104	25.3	2.5	—	7.4	2.9	1.0
AA(DDS + control)	98	20.7	3.6	—	6.1	3.1	2.1
Total	202	23.2	3.1	—	6.7	3.0	1.5

TABLE 13.—*Clinical and bacteriologic improvement after 56 weeks, in relation to sex, age-group and stage of the disease, for all therapy groups at both institutions.*

	Number of patients	Per cent clinically improved	Per cent reduction in total bacteriologic scores for 8 sites
Sex:			
Male	142	54.9	27.5
Female	60	48.3	33.1
Age-group:			
10-19 yrs.	73	57.5	31.5
20-29 yrs.	66	50.0	23.4
30 yrs. & over	63	50.8	32.5
Stage of the disease:			
L1	32	53.1	47.4
L2	101	50.5	29.1
L3	54	53.7	20.9
Borderline	15	66.7	27.2
Total	202	53.0	29.1

in this study. Granular bacilli were present both in the preliminary and posttherapy smears but no differences were observed between the therapy groups in this respect, as far as we were able to determine.

Association between clinical and bacteriologic improvement. A positive correlation is to be expected between the clinical and bacteriologic appraisals at the end of 56 weeks and this is evident in Table 11. It was of a somewhat lower degree than anticipated, however, as shown by the fact that 47.3 per cent of the patients with a reduction of less than 50 per cent in bacteriologic score were still rated as clinically improved, as compared to 68.5 per cent of those with a bacteriologic reduction of 50 per cent or greater. Similarly, 17.9 per cent of the patients regarded as clinically unchanged or worse had a reduction of 50 per cent or more in bacteriologic score, as compared to 34.6 per cent of those showing clinical improvement.

ERYTHROCYTE SEDIMENTATION AND GAMMA GLOBULIN LEVELS BEFORE AND AFTER THERAPY

Red-cell sedimentation rates by the Westergren method and serum gamma globulin measurements by paper electrophoresis were made on all the patients prior to therapy and at the end of 56 weeks to determine whether a trend towards normality would be more evident in one therapy group than in the other. For both procedures the average reductions from pretherapy levels were small and there were no appreciable differences between the Etisul and control groups. Extremely wide variations in sedimentation rate and relative gamma globulin percentage were observed in lepromatous patients of the same clinical or bacteriologic status. Furthermore, sedimentation rates and gamma globulin values were affected much more markedly by the occurrence of lepra reaction in the patients than by the effect of therapy on the underlying lepromatous condition.

REACTIONS

A special study of reactions was made during this series. All patients were examined once a week on the same day for signs and symptoms of lepra reaction. The absence or presence and the relative severity or extent of each manifestation were entered as —, or +, ++ and +++ in a weekly chart designed for this purpose. Table 12 summarizes the findings in terms of average "person-weeks" of each reactional sign or symptom in patients completing 56 weeks of treatment in each therapy group, both institutions combined.

Erythema nodosum leprosum was by far the most commonly observed reactional condition. It was present in 47.6 per cent of the total weekly observations made on all patients, the averages being 25.3 person-weeks of ENL for the Etisul group and 20.7 for the control group. Plaques of possible reactional nature, fever, neuritis and conjunctivitis or iritis, occurred much less frequently, as shown by the average number of weeks during which these manifestations were

present in the patients. There was no significant difference between the two therapy groups in respect to the occurrence of various reactional manifestations, although the patients receiving Etisul inunctions in addition to DDS developed somewhat more ENL than the controls.

LEPROMIN TESTS

All patients were negative to Mitsuda-Hayashi lepromin when tested at the start of the Fifth Series. At Central Luzon 100 patients were retested with the same lepromin at the end of 56 weeks; only 1 developed a Mitsuda reaction larger than 4 mm. in diameter. At Eversley Childs 98 patients were similarly retested; 2 had late reactions measuring somewhat more than 4 mm. in diameter. Thus, of 198 patients who were retested only 3, or 1.5 per cent, developed significant reactivity of the Mitsuda type at the end of 56 weeks. Two of these 3 patients were regarded as moderately improved, 1 as only slightly clinically improved; 2 became completely bacteriologically negative and 1 showed a 70 per cent reduction in total bacteriologic score.

VARIOUS BACKGROUND FACTORS

In the previous four series ([7, 8, 9, 10]), as in this one, such factors as the sex and age of the patients, stage or advancement of the disease, and previous sulfone therapy, were always taken carefully into account in matching the therapy groups because of their possible influence on the outcome of therapy. No consistent or significant relationship was found in the four previous series, however, among any of these factors and either clinical or bacteriologic changes occurring during therapy.

In the present series, as seen in Table 13, there was again no evidence of any significant relationship between prognosis and the age or sex of the patients and the stage of the disease, except for a slightly greater bacteriologic improvement in 32 patients classed as L1 than in the more advanced L2 and L3 patients, which was probably a chance occurrence. Among the 15 basically lepromatous patients (all had diffuse infiltration) with borderline lesions (well-defined plaques) who were included in the study, clinical improvement (66.7%) was somewhat higher than among the purely lepromatous cases, but the bacteriologic improvement in these borderline cases was not better than that observed in all patients in the experiment.

Although the findings in previous series never supported the opinion that prior sulfone therapy is an important factor in determining the outcome of treatment, an effort was made in this series to limit the patients largely to new and untreated cases. As seen in Table 1, only 5 patients had received between 5 and 7 gm. of total prior DDS therapy. Of these 5 patients, only 1 was considered clinically improved at the end of 56 weeks and their average reduction in total bacteriologic score was 24.9 per cent compared to 29.1 per cent for all patients in the study.

SUMMARY

In a duplicate "double-blind" therapeutic study on lepromatous leprosy carried out at two Philippine leprosaria, two major groups of patients were treated respectively with DDS plus Etisul inunctions, and with DDS plus inunctions with a control ointment similar to the Etisul cream in color, consistency and odor. Treatment was continued for 56 weeks, the maximum daily DDS dose of 2 mgm. per kgm. body weight being reached over an induction period of eight weeks. The Etisul and control inunctions (one 5-gm. tube three times a week) were started on the ninth week and continued for 48 weeks until the end of the experiment.

No evidence was found that supplementary therapy with Etisul was of added advantage to the patients. Patients of both therapy groups showed definite clinical and bacteriologic improvement, but those treated with Etisul inunctions in addition to DDS showed no better progress than those treated only with DDS.

Determination of erythrocyte sedimentation rates and of the serum gamma globulin fraction did not prove helpful in measuring the efficacy of the therapies.

Reactional manifestations were approximately equally frequent in the two therapy groups.

As in previous series, there was no evidence that sex, age or the stage of the disease was unduly associated with either clinical or bacteriologic improvement.

Only about 1.5 per cent of the patients developed significant lepromin reactivity of the Mitsuda type at the end of 56 weeks, and all the reactions were small.

RESUMEN

En un estudio terapéutico duplicado "ciego" sobre lepra lepromatosa llevado a cabo en dos leprosarios filipinos, dos grandes grupos de pacientes fueron tratados respectivamente con DDS mas unciones de Etisul, y con DDS mas unciones con un unguento similar a la crema de Etusil en el color, consistencia y olor. El tratamiento fué contínuo durante 56 semanas, la dosis máxima de DDS fué de 2 mgm. por kilo de peso del cuerpo, que fué alcanzado en un período de inducción de 8 semanas. Las unciones de Etisul y del control (un tubo de 5 gm. tres veces a la semana) fueron comenzadas en la novena semana y continuaron durante 48 semanas hasta el final del experimento.

No fué encontrada evidencia de que la terapia suplementaria con Etisul tuviera alguna ventaja para el paciente. Pacientes de ambos grupos terapéuticos mostraron mejoría definida clínica y bacteriológica, pero aquellos tratados con unciones de Etisul en adición al DDS, no mostraron mejores progresos que aquellos tratados solamente con DDS.

Las determinaciones de los niveles de eritrosedimentación y de las fracciones de gamma globulina sérica no probaron ser una ayuda en la medición de las terapias.

Las manifestaciones reaccionales fueron aproximadamente iguales en ambos grupos de terapia.

Como en series previas, no hubo evidencias de que el sexo, la edad o el estadio de la enfermedad, estuvieran excesivamente asociadas con la mejoría clínica o bacteriológica.

Solamente alrededor del 1.5% de los pacientes desarrollaron reacción significativa lepromina del tipo Mitsuda al final de 56 semanas, y todas las reacciones fueron pequeñas.

RESUMÉ

Une étude thérapeutique double conduite selon la technique du double incognito chez des malades atteints de lépre lépromateuse a été menée dans deux léproseries des Philippines. Deux groupes principaux de malades ont été traités respectivement par la DDS avec frictions d'Etisul et par la DDS accompagnée de la friction d'une pommade témoin ressemblant à la crème d'Etisul quant à la couleur, la consistance et l'odeur. Le traitement a été poursuivi durant 56 semaines, la dose quotidienne maxima d'Etisul, soit 2 mgm. par kilogramme de poids corporel, ayant étè atteinte progressivement au bout de 8 semaines de posologie graduellement augmentée. L'Etisul et les frictions de controle (un tube de 5 gm. à raison de trois applications par semaine) ont été commencées à la neuvième semaine et poursuivies durant 48 semaines jusqu'à la fin de l'expérience.

Rien n'a permis de mettre en évidence le fait qu'un avantage complémentaire serait fourni aux malades par le thérapeutique d'appoint à l'Etisul. Les malades répartis dans les deux groupes thérapeutiques ont témoigné d'une amélioration nette, tant clinique que bactériologique, mais ceux traités par les frictions d'Etisul en plus du traitement par la DDS n'ont pas manifesté de progrès supérieurs à ceux des malades traités par la DDS seulement.

Les déterminations du taux de sédimentation des érythrocytes et de la fraction gamma des globulines sériques ne se sont pas révelées utiles pour mesurer l'efficacité des traitements.

Des manifestations réactionnelles ont été notées approximativement avec la même fréquence dans les deux groupes thérapeutiques.

Comme dans les séries précedentes, il n'y a pas eu d'evidence que le sexe, l'âge ou le stade de la maladie soient spécialement associés avec l'amélioration clinique ou bactériologique.

Environ 1.5% des malades seulement ont developpé aprèe 56 semaines une capacité de réponse du type Mitsuda à la lépromine, et toutes les réactions étaient de petites dimensions.

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REFERENCES

1. Bertaccini, G. (1957). Atti S. I. D. E. S. Minerva Dermatol. 32. 1. 22-24
2. Brown, H. D., Matzuk, A. R., Becker, H. J., Conbere, J. P., Constantin, J. M., Solotorovsky, M., Winsten, S., Ironson, E., and Quastel, J. H. The antituberculosis activity of some ethylmercapto compounds. J. American Chem. Soc. **76** (1954) 3860.
3. Chang, Y. T., and Doull, J. A. Mercaptan compounds in tuberculosis and leprosy. Leprosy Briefs **10** (1959) 41.43.

4. Davey, R. F. and Hogerzeil, L. M. Diethyl dithiolisophthalate in the treatment of leprosy (ETIP or "Etisul"); A progress report. Leprosy Rev. **30** (1959) 61-72.
5. Davies, G. E. and Driver, G. W. The antituberculous activity of ethyl thioesters, with particular reference to diethyl dithiolisophthalate. British J. Pharmacol. **12** (1957) 434-437.
6. Davies, G. E., Driver, G. W., Hoggarth, E., Margin, A. R., Paige, M. F. C., Rose, F. L., and Wilson, B. R. Studies in the chemotherapy of tuberculosis: Ethyl mercaptan and related compounds. British J. Pharmacol. **11** (1956) 351-356.
7. Doull, J. A. Clinical evaluation studies in lepromatous leprosy. First series: Diasone (Diamidin), 4,4′—diaminodiphenyl sulfone, dihydrostreptomycin. Internat J. Leprosy **22** (1954) 377-402.
8. Doull, J. A., Rodriguez, J. N., Davison, A. R., Tolentino, J. G., and Fernandez, J. V. Clinical evaluation studies in lepromatous leprosy. Second series: Isoniazid and diasone (Diamidin), isoniazid and dihydrostreptomycin. Also a pilot study with streptohydrazid. Internat. J. Leprosy **25** (1957) 173-192.
9. Doull, J. A., Rodriguez, J. N., Davison, A. R., Tolentino, J. G., and Fernandez, J. V. Clinical evaluation studies in lepromatous leprosy. Third series: Nicotinamide, and BCG vaccination as supplements to diaminodiphenyl sulfone (DDS). Internat. J. Leprosy **26** (1958) 219-235.
10. Doull, J. A., Rodriguez, J. N., Tolentino, J. G., Fernandez, J. V., Guinto, R. S., Rivera, J. N., and Mabalay, M. C. Clinical evaluation studies in lepromatous leprosy. Fourth series: 4,butoxy-4′dimethylaminodiphenyl thiourea (DPT), amodiaquin, and 4,4′-diaminodiphenyl sulfone (DDS) 2.5 mgm. and 4 mgm. per kg. of body weight. Internat. J. Leprosy **29** (1961) 291-317.
11. Kushner, S., Dalalian, H., Back, F. L., Jr., Centola, D., Sanjurjo, J. L., and Williams, J. H. Experimental chemotherapy of tuberculosis. III. Ethyl mercaptan and related compounds in tuberculosis. J. American Chem. Soc. **77** (1955) 1152-1155.
12. del Pianto, E. Antituberculosis activity of sodium ethylthiosulfate. Ricerca Sc. **23** (1953) 1785-1793. (Cited in Chem. Abstr. **48** (1954) 13989.)
13. del Pianto, E. Thioethyl compounds in therapy of leprosy. Leprosy Rev. **30** (1959) 23-24.
14. Solotorovsky, M. Effect of ventilation on antituberculosis activity of S-ethyl-L-cysteine and related compounds. American Rev. Tubere. **74** (1956) 68-71.
15. Solotorovsky, M., Winsten, S., Ironson, E. J., Brown, H. D., and Becker, H. J. S-ethyl-L-cysteine, a member of a new group of antituberculous compounds. American Rev. Tubere. **70** (1954) 806-811.