BLOOD AND URINE CONCENTRATIONS OF FREE AND HYDROLYZED SULFONES IN LEPROSY PATIENTS UNDER TREATMENT

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For the past nine years interest has been centered on the sulfones, a group of chemicals whose effectiveness in the treatment of leprosy has been repeatedly demonstrated. The use of promin, diasone and sulphetrone has been extensively reported on in the literature. Most of the effective sulfones (promin, diasone, sulphetrone, and promacetin) possess a diaminodiphenyl sulfone nucleus with symmetrically placed amino groupings in the 4 and 4′ positions.

Whether or not diaminodiphenyl sulfone as such is the essential active substance in the sulfones has been the cause of considerable argument among research workers. This is mainly because all of the sulfones that have been studied extensively have been derivatives of that mother substance. It was the first sulfone compound synthesized, but although it has a high tuberculotherapeutic efficacy, its high toxicity has limited its clinical application. Promin, diasone and sulphetrone were made in order to reduce the toxic effects.

Because diaminodiphenyl sulfone *in vitro* is highly active, much more so than any of its derivatives, it is thought that all of the derivative compounds owe their activity to their slow hydrolysis and consequent release of the parent substance. Promacetin, one of the more recent sulfones used in the treatment of leprosy (4), is not a derivative of diaminodiphenyl sulfone, although that substance is its central nucleus. In the manufacture of promacetin, its molecule is built up by synthesis and not derived from the basic sulfone. According to the manufacturers (6) it does not hydrolyze, and therefore it does not depend upon diaminodiphenyl sulfone for its therapeutic value.

On the basis of toxicity studies, Brownlee and his associates (3) have assumed that sulphetrone is not hydrolyzed in the body to diaminodiphenyl sulfone. However, experimental work performed by Smith and associates (7) indicates that it does hydrolyze in the body in that manner.

METHODS USED

The work here reported was undertaken to determine the degree of break-down that occurs within the body of leprous patients during the course of sulfone treatment, and after rest periods when stored drug is being excreted.

This report is based upon 275 determinations of the amounts of free and hydrolyzed sulfone in the bloods and urines of 137 patients, made over a period of three months. The drugs used and the numbers of cases are as follows:

Promin, 43 patients taking the drug from 1 to 7 years.
Diasone, 55 patients taking the drug from 1 to 6 years.
Sulphetrone, 15 patients taking the drug from 4 to 21 months.
Promacetin, 24 patients taking the drug from 4 to 21 months.

The analytical procedure used was that of Bratton and Marshall (2) with modifications for sulphetrone as recommended by Brownlee, Green and Woodbine (3), and for promacetin as recommended by Bratton (1). All analyses were for free and for acetylated or conjugated sulfone. No attempt was made to identify the degraded product as diaminodiphenyl sulfone. According to Long and Bliss (5), that is excreted in the urine in unaltered as well as in conjugated form. The Klett-Summerson photoelectric colorimeter was used for the readings. The blood for the determinations was always drawn at the same hour of the day, between 8 and 9 a. m., and therefore in constant relation to the time of administration of the compound. The urine sulfone concentrations were based upon 24-hour specimens collected from 7 a. m. to 7 a. m. the next day. All determinations were made 24 hours after the last dose and after a rest period of 14 days, and all analyses were completed on the same day that the specimens were received.

RESULTS

The concentrations of the various sulfones in the bloods and urines of the 137 patients are presented in Tables 1 to 3, according to the particular drugs which the patients were receiving.

Promin.—The first section of Table 1 shows the minimum, maximum and average concentrations of free and hydrolyzed promin in the bloods and urines of 43 patients who had received single daily intravenous 5 gm. doses of the drug for from 1 to 7 years. In all cases promin was found to exist in the blood only in the free form, since the values for free and total promin by direct diazotization and by acid hydrolysis were identical. On

the other hand, in the urine the drug was found in both the free and conjugated forms on the 14th day of the rest period. The ratio between the free and conjugated forms varied con-

TABLE 1.—*Minimum, maximum and average concentrations of free and hydrolyzed sulfones in the blood and urine of patients receiving promin and diasone, 24 hours after the last dose and after a rest period of 14 days, according to the length of treatment; mgm. per 100 cc.*

Years of treatment	No. of cases	Range	Concentration 24 hours after last dose				Concentration after a 14-day rest period			
			Blood		Urine		Blood		Urine	
			Free	H/*a*	Free	H	Free	H	Free	H
Promin (43 cases)										
7	7	Min.	0.5	0	24.0	4.0	0	0	2.0	1.0
		Max.	3.4	0	204.0	56.0	0	0	6.0	4.0
		Ave.	1.1	0	97.0	26.0	0	0	4.0	1.8
6	6	Min.	0.8	0	21.0	5.0	0	0	1.0	0.8
		Max.	2.2	0	80.0	80.0	0	0	9.0	1.3
		Ave.	1.1	0	61.0	38.0	0	0	3.2	0.7
5	4	Min.	0.6	0	20.0	3.0	0	0	2.0	0.3
		Max.	2.3	0	60.0	52.0	0	0	4.2	1.2
		Ave.	1.0	0	31.0	20.0	0	0	3.2	0.6
4	4	Min.	0.5	0	10.0	3.0	0	0	1.0	1.0
		Max.	2.0	0	172.0	80.0	0	0	6.0	3.8
		Ave.	1.1	0	86.0	33.0	0	0	3.0	1.9
3	4	Min.	0.9	0	40.0	30.0	0	0	1.2	0.6
		Max.	2.0	0	180.0	60.0	0	0	10.0	10.0
		Ave.	1.3	0	120.0	42.0	0	0	3.8	3.4
2	12	Min.	0.9	0	80.0	10.0	0	0	1.0	0.8
		Max.	1.5	0	140.0	90.0	0	0	8.0	3.0
		Ave.	1.1	0	105.0	35.0	0	0	3.0	1.1
1	6	Min.	Tr.	0	12.0	6.0	0	0	1.5	0.2
		Max.	4.3	0	142.0	35.0	0	0	2.9	1.3
		Ave.	1.4	0	98.0	16.0	0	0	1.8	0.5

a Hydrolyzed sulfone.

TABLE 1.—Continued on next page.

TABLE 1.—Continued.

Years of treatment	No. of cases	Range	Concentration 24 hours after last dose				Concentration after 14-day rest period			
			Blood		Urine		Blood		Urine	
			Free	H/*a*	Free	H	Free	H	Free	H
Diasone (55 cases)										
6	11	Min.	0.5	0.1	10.0	0.2	0	0	0.9	0.3
		Max.	1.9	0.9	20.0	15.0	0	0	2.4	1.3
		Ave.	1.0	0.2	20.8	4.0	0	0	1.8	0.5
5	8	Min.	0.5	Tr.	6.0	0.4	0	0	1.3	0.3
		Max.	1.5	0.7	18.0	10.0	0	0	2.5	2.0
		Ave.	0.9	0.1	12.1	2.7	0	0	1.6	0.6
4	8	Min.	0.7	0.1	8.0	2.1	0	0	1.0	0.2
		Max.	1.9	0.9	30.0	15.0	0	0	2.8	1.1
		Ave.	1.1	0.3	16.0	5.0	0	0	1.8	0.5
3	19	Min.	Tr.	0.1	2.4	0.1	0	0	0.5	0.2
		Max.	2.1	0.8	21.0	5.2	0	0	3.9	1.0
		Ave.	1.0	0.2	11.0	2.0	0	0	1.6	0.3
2	5	Min.	0.6	0.1	8.4	0.3	0	0	1.1	0.4
		Max.	2.1	0.4	45.0	5.0	0	0	4.5	1.0
		Ave.	1.3	0.2	15.4	1.8	0	0	2.3	0.6
1	4	Min.	1.0	0.1	16.8	0.7	0	0	1.8	0.2
		Max.	1.5	0.8	74.0	26.0	0	0	2.6	0.9
		Ave.	1.3	0.4	44.0	9.5	0	0	2.1	0.6

a Hydrolyzed sulfone.

siderably in different individuals. In 4 of the patients the drug was excreted in the free and conjugated forms in equal amounts. Only one patient of the 43 showed free promin alone, with no conjugated product, in the urine 24 hours after the last injection. Only 8 cases showed conjugated promin in the urine after the rest period, although all of them eliminated free promin.

Diasone.—The second section of Table 1 shows in the same way the concentrations of free and hydrolyzed diasone in the bloods and urines of 55 patients who had received that drug for

from 1 to 6 years, with a daily oral dose of 1.0 gm. Of this group, 26 showed conjugation of the drug in the blood 24 hours after the last dose, while in 29 the conjugated form was absent. Neither form was present in the blood in any case after the 14-day rest period. Conjugation of diasone in the urine occurred in 52 of the cases, 24 hours after the last dose and after the rest

TABLE 2.—*Concentration of free and hydrolyzed sulphetrone in the blood and urine 24 hours after the last dose and after a rest period of 14 days; mgm. per 100 cc.*

Case Number	Dose, gm.	Concentration 24 hours after last dose				Concentration after a 14-day rest period			
		Blood		Urine		Blood		Urine	
		Free	H/*a*	Free	H	Free	H	Free	H
Cases with ordinary values (12)									
1996	1.0	4.0	0.3	70	20	0	0	8.0	0
1939	1.0	3.5	0.4	70	15	0	0	9.0	1.0
1911	1.5	3.0	1.5	70	70	0	0	15.0	2.0
1940	1.5	6.5	0.6	80	60	0	0	7.5	0.0
1936	2.0	5.5	0.5	120	120	0	0	2.5	0.5
795	2.0	6.3	2.7	110	30	0	0	5.0	.0.0
1984	2.5	1.2	0.3	90	0.5	0	0	10.0	0.3
1810	3.0	4.5	1.3	85	35	0	0	10.5	0.0
1720	3.0	4.0	0.3	80	40	0	0	15.0	2.0
1349	3.0	3.5	1.5	90	70	0	0	10.0	0.0
1975	3.0	2.1	1.4	24	26	0	0	9.0	0.0
1921	3.0	4.0	1.2	115	60	0	0	8.0	0.0
Averages		4.0	1.0	83.6	45.5	0	0	9.0	0.3
Cases with high values (3)									
1991	1.0	9.5	2.5	225	175	0	0	2.6	1.2
1997	2.0	10.0	1.5	500	150	0	0	4.0	11.0
1909	3.0	14.0	3.0	700	250	0	0	15.0	3.0
Averages		11.1	2.3	475	191	0	0	7.2	5.1

a Hydrolyzed sulfone.

period. The ratio between the free and conjugated drug in the blood and urine varied considerably in different individuals.

Sulphetrone.—Table 2 shows the concentrations of free and hydrolyzed sulphetrone in the bloods and urines of 15 patients who had received that drug for from 4 to 21 months, with daily oral doses of from 1.0 to 3.0 gm. Free and conjugated forms were found in both the blood and urine in all of the cases, 24 hours after the last dose. The drug was not present in the blood after the rest period. Conjugation of sulphetrone in the urine was detected in 8 of the 15 cases after the rest period, while free sulphetrone was eliminated in all of them. Especially high blood and urine concentrations were observed in 3 patients, with no apparent toxic manifestations. Marked variations occurred in the ability of the body to conjugate sulphetrone, as shown by the free and hydrolyzed contents of the urine.

Promacetin.—The results obtained in 24 patients who had been on promacetin therapy for from 4 to 21 months, with daily oral doses of from 1 to 3 gm., are shown in Table 3. The values for free and total promacetin were identical, which indicates that this drug is unaltered in the human body.

TABLE 3.—*Minimum, maximum and average concentrations of promacetin in the blood and urine of 24 patients 24 hours after the last dose and after a rest period of 14 days; mgm. per 100 cc./a*

Range	Concentration 24 hours after last dose		Concentration after a 14-day rest period	
	Blood	Urine	Blood	Urine
Minimum	0.50	20	Trace	1.1
Maximum	1.90	300	0.6	5.0
Average	1.15	84	0.4	3.5

a Determinations were made for free and hydrolyzed sulfone. Acetylization or conjugation was not found in this group.

SUMMARY

A total of 275 determinations of the concentrations of free and hydrolyzed sulfones in the blood and urine were made in 137 patients over a period of three months. Analyses were made 24 hours after the last dose, and after a rest period of 14 days.

In all of the 43 patients under promin treatment, the drug existed in the blood in the free form only, while in the urine it was found in both the free and conjugated forms after the 14-day

rest period. The ratio between the free and conjugated drug in the urine varied considerably in different individuals.

Of the 55 patients taking diasone, 26, or 47.3 per cent, showed conjugation of the drug in the blood 24 hours after the last dose, while in 29, or 52.7 per cent, there was no conjugation in the blood. No form of the drug was present in the blood of any of the patients after a rest period of 14 days. Conjugation in the urine occurred in 52 of the cases, 24 hours after the last oral dose and after the rest period.

Concentrations of free and conjugated sulphetrone were noted in the blood and urine in the 15 cases studied, 24 hours after the last dose. This drug was not present in the blood after the rest period. Conjugation of sulphetrone in the urine occurred in 8 of the 15 cases after the rest period, while free sulphetrone was eliminated in all of the cases.

When the bloods and urines of 24 patients receiving promacetin were analyzed by direct diazotization and by acid hydrolysis the values for free and total sulfone were identical, which indicates that this drug was unaltered in the body of the patients studied.

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RESUMEN EN ESPAÑOL

Un total de 275 determinaciones de concentración de sulfonas libres e hidrolizadas en la sangre y la orina, se hizo en 137 pacientes durante un período de 3 meses. Los analisis se practicaron 24 horas después de la última dosis y después de un período de reposo de 14 días.

En todos de 43 pacientes bajo tratamiento con promina, la droga fué hallada en la sangre en el estado libre solamente, mientras que en la orina se la encontró tanto libre como conjugada después de los 14 días de reposo. La proporción de droga libre a droga conjugada varió considerablemente entre los pacientes individuales.

De 55 pacientes bajo tratamiento con diasone, 26 (47.3%) demostraron conjugación de la droga en la sangre 24 horas después de la última dosis, mientras que en 29 (52.7%) no hubo conjugación en la sangre. No hubo droga en la sangre, en ninguna forma, después de los 14 días de reposo. Conjugación en la orina ocurrió en 52 de los casos 24 horas después de la última dosis y después del período de reposo.

Se notaron concentraciónes de sulfetrona libre y conjugada en la sangre y orina de 15 pacientes 24 horas después de la última dosis. No hubo droga en la sangre después del período de reposo.

Conjugación de sulfetrona en la orina ocurrió en 8 de los 15 casos después del período de reposo, mientras que sulfetrona libre fué eliminada por todos los pacientes.

Cuando las sangres y las orinas de 24 pacientes bajo tratamiento con promacetina, fueron analizadas, los valores para sulfonas libres y totales fueron idénticos, lo que indica que la droga no fué alterada por el sistema de los pacientes estudiados.

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