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Drug interaction during multidrug regimens for treatment of leprosy

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The influence of concurrently administered rifampicin and clofazimine on the metabolism of 4, 4′-diaminodiphenyl sulfone (dapsone, DDS) has been studied in 30 subjects on multidrug regimens for treatment of leprosy. Plasma and urinary levels of drugs were determined on days 2, 8 and 15 after administration of the drug, while creatinine levels in urine were also determined to overcome the effect of diuresis. During concurrent administration of rifampicin the plasma levels of DDS gradually fell from day 2 to day 15 of rifampicin administration and the decrease was most significant on day 15. Clofazimine did not exert any such influence on DDS metabolism. A comparison of urine and plasma levels of DDS showed an inverse relationship between the plasma levels and urinary excretion of DDS during the course of treatment. The findings suggest that during concurrent administration of DDS and rifampicin, the intake of DDS should be regular and uninterrupted.

The study of interactions of the drugs used in the treatment of leprosy is of topical importance in the context of the multidrug regimens widely employed. The first report on the possible interaction of rifampicin and DDS was made by Gelber *et al*[1] who observed that concurrent administration of the two drugs increased the rate of plasma DDS clearance. Peters *et al*[2] reported that concurrent administration of rifampicin decreased the blood and tissue levels of DDS. These findings were substantiated by Balakrishnan and Seshadri[3,4] and Venkatesan *et al*[5]. We undertook a study of the interaction between rifampicin and DDS as also between clofazimine and DDS, and the findings obtained are reported here.

Material & Methods

Thirty clinically active patients of lepromatous leprosy were included in this study. Untreated patients or those receiving dapsone monotherapy were given preference. None of the patients had received rifampicin or clofazimine for the previous one year. The patients were admitted and kept under supervision during the period of investigation.

During the first phase, the interaction between DDS and rifampicin were studied. The basal blood and urine specimens were collected and a dose of 100 mg DDS was administered. Blood and urine specimens were then collected at 3 h, 6 h and 24 h after administration of the drug. No drug was administered over the next two days. On the fourth day, a dose of 500 mg sulphadimidine (Elkosin) was administered and blood and urine specimens were collected at 3 h and 6 h. The patient was put back on DDS (100 mg) daily for about a week. Subsiquently 600 mg rifampicin along with 100 mg DDS was administered on an empty stomach daily for 15 consecutive days. During this period, blood and urine specimens were collected on days 2, 8 and 15 at 3 h, 6 h and 24 h after drug administration.

In the second phase, the interaction between DDS and clofazimine was studied. For this the patient was given 100 mg DDS daily for a week, followed by 100 mg clofazimine along with 100 mg DDS for the next 15 days. Blood and urine specimens were collected on days 2, 8 and 15.

In all patients blood specimens were collected in oxalated tubes and plasma was separated immediately. Drug assays were either done immediately or the plasma aliquots were frozen at -20°C till the assay was done. The urine samples were also kept in the refrigerator if assays were not carried out immediately.

The plasma DDS levels were determined by the spectrophoto-fluorometric technique[6]. An Aminco-Bowman spectrophotofluorometer (American Instrument Company, Silver Spring, Maryland, USA) was used throughout the experiment. The urinary DDS levels (total acid labile metabolites) were measured by the colorimetric technique of Ellard *et al*[7]. Rifampicin levels in plasma were determined by microbiological assay[8] with *Staphylococcus aureus* (NCTC 10702). Rifampicin assays in urine were carried out by chloroform extraction method[9]. Creatinine levels in urine were determined by alkaline picrate method[10]. Clofazimine levels in plasma and urine were not estimated due to the lack of required equipment (HPLC). The colorimetric technique of Barry *et al*[11] for estimation of clofazimine is not sensitive enough to detect clofazimine in plasma or urine after a daily dose of 100 mg.

In order to find out the acetylator phenotypes, sulphadimidine levels in plasma were assayed by the method of Bratton and Marshall[12]. The standard DDS powder was obtained from Wellcome Research Laboratories, Beckenham, England. Purified rifampicin powder was procured from Lepetit, Milan, Itlay. Arithmetical mean and standard error were calculated for the data.

Results

During rifampicin administration the plasma levels of DDS gradually fell from days 2 to 15 and the decrease was marked on day 15. This decrease was reflected at all time intervals. During clofazimine administration the plasma levels of DDS did not seen to be altered (Table I).

In order to overcome the problem of diuresis the creatinine levels in urine were also estimated and the DDS levels are expressed as μg/mg creatinine. A comparison of urine and plasma levels of DDS (Tables I and II) shows an inverse relationship between the plasma levels and urinary excretion of DDS during concurrent rifampicin administration. No such relationship

Table I. Plasma DDS levels (μg/ml) in leprosy patients (30) on multidrug therapy (Data are mean $\pm$SE)

Drug administered	Day of examination	Time interval			
		Basal	3 h	6 h	24 h
DDS only		1.18±0.19	2.23±0.22	2.28±0.18	1.48±0.16
DDS+RIF	2	—	2.87±0.17	2.45±0.14	1.43±0.12
	8	—	1.74±0.12	1.44±0.11	0.75±0.08
	15	—	1.46±0.13	1.15±0.10	0.44±0.05
DDS+CLF	2	—	2.26+0.16	1.82+0.13	1.07+0.12
	8	—	2.17±0.14	1.97±0.13	1.41±0.14
	15	—	2.24±0.15	2.20±0.13	1.39±0.12

RIF, rifampicin; CLF, clofazimine

Table II. Urinary DDS level (μg/mg creatinine) in leprosy patients (30) on multidrug therapy (Data are mean±SE)

Drug administered	Day of examination	Time interval			
		Basal	3h	6 h	24 h
DDS only		34.94±4.90	60.52±6.13	72.02±8.31	55.15±4.46
DDS+RIF	2	—	76.20±6.60	80.79±5.96	69.71±5.06
	8	—	80.19±5.63	88.05±7.17	58.11±5.79
	15	—	83.55±6.19	102.55±8.03	53.04±3.99
DDS+CLF	2	—	88.17±8.76	95.83±6.40	53.77±3.67
	8	—	81.81±5.00	88.07±6.31	57.51±3.62
	15	—	80.80±7.27	82.95±5.35	57.75±4.28

was noticed between plasma and urinary levels during clofazimine treatment.

The rifampicin levels in plasma and urine in the same subjects (Table III) clearly show that the drug is cleared from the body within 24 h.

Acetylation rates in the plasma samples drawn at 6 h were studied and the acetylator phenotypes were classified according to Balakrishnan and Ramu[13]. Among the 30 patients studied, 3 were rapid, 8 were intermediate and 19 were slow acetylators. Table IV shows DDS levels in plasma, classified according to acetylator phenotypes during concurrent administration of rifampicin. The results clearly indicate that the acetylator phenotype has no effect on the DDS clearance phenomenon by rifampicin.

Table III. Plasma and urinary rifampicin levels in leprosy patients on multidrug therapy, DDS+RIF

Day of examination	Time interval		
	3 h	6 h	24 h
Plasma :			
2	12.30±0.91	10.65±1.07	0.82±0.26
8	11.13±1.14	9.63±1.17	0.64±0.46
15	10.36±0.65	8.29±0.74	0.52±0.36
Urinary :			
2	260.22±27.01	265.07±17.47	21.49±4.74
8	156.47±16.58	178.19±17.49	28.54±6.28
15	185.46±16.97	234.70±20.55	20.04±3.81

Table IV. DDS levels in plasma (μg/ml) classified according to acetylator phenotypes during concurrent rifampicin administration

(Data are mean ± SE)

Acetylator phenotypes	Day of examination	Time interval		
		3 h	6 h	24 h
Rapid (n=3)	2	2.84±0.07	2.44±0.04	1.41±0.04
(61—80%)	8	1.77±0.05	1.44±0.06	0.74±0.02
	15	1.43±0.08	1.16±0.04	0.46±0.02
Intermediate (n=8)	2	2.90±0.18	2.43±0.14	1.42±0.13
(36—60%)	8	1.76±0.13	1.43±0.12	0.75±0.07
	15	1.45±0.08	1.12±0.05	0.47±0.06
Slow (n=19)	2	2.89±0.14	2.49±0.14	1.47±0.11
(10—35%)	8	1.71±0.10	1.46±0.10	0.75±0.08
	15	1.49±0.12	1.17±0.10	0.41±0.04

Discussion

Drug interaction is a common phenomenon during multidrug regimens. Balakrishnan and Seshadri[3,4] confirmed earlier observations[1,2] on the interaction of DDS and rifampicin and also suggested that rifampicin had a transient mobilizing effect on DDS depot in the body, resulting in an increased excretion of DDS in urine. In the present study, it was observed that during rifampicin administration the plasma level of DDS gradually fell from day 2 to 15 and the decrease was most marked on day 15.

A few authors have reported about the possible induction of liver microsomal enzymes by rifampicin[1,14–16] which is the cause for the rapid clearance of DDS from plasma as seen in the present studies as well.

Rifampicin induces its own metabolism in addition to that of many other drugs, as

is evident from our data (Table III). It was reported that during continuous administration of rifampicin, its half-life gradually decreases as a result of microsomal enzyme induction until a steady state is reached[17]. It has been observed that the rifampicin/creatinine ratios were initially high and this is followed by a rapid fall to a steady level in the later stages[4]. In the present study we observed that more than 95 per cent of the drug was cleared from the body within 24 h and its level in plasma gradually decreased and reached the minimum on day 15.

The lowest DDS level encountered in the present study during concurrent administration of rifampicin was 0.44 μg/ml in the 24 h blood sample on day 15. However, it may be mentioned that the level of drug was very much above the minimal inhibitory concentration of DDS against *Mycobacterium leprae* which is about 3 ng/ml[18]. Hence, the 14 days intensive rifampicin therapy as used by the programmes under Government of India will have no adverse effect despite the lower plasma level of DDS, even after 15 days of combined therapy.

In the present study, it was observed that clofazimine has no effect on plasma DDS clearance rate. A similar observation was made by Venkatesan *et al*[19] during concurrent administration of DDS, rifampicin and clofazimine. Clofazimine has no effect on the urinary excretion of DDS also. The studies conducted by Venkatesan *et al*[5] and Balakrishnan and Seshadri[4] also did not show any influence on urinary excretion of DDS by clofazimine. However, Grabosz and Wheate[20] reported that clofazimine administration enhanced the urinary excretion of DDS in 8 of 17 leprosy patients studied by them.

It has been reported that the DDS clearing phenomenon by rifampicin was not significantly affected by acetylator phenotypes[5]. In the present study also, similar observations have been made in respect of plasma DDS clearance rate and acetylator phenotypes.

The clinical implication of the present findings is that during concurrent administration of rifampicin with DDS, the intake of DDS needs to be regular and uninterrupted. The possible mechanism of rifampicin induction over the metabolism of dapsone is yet to be investigated.

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