Br. J. clin. Pharmac. (1987), **23**, 482–485

Evidence for polymorphic oxidation of sparteine in Japanese subjects

T. ISHIZAKI[1], M. EICHELBAUM[2]*, Y. HORAI[1], K. HASHIMOTO[3], K. CHIBA[1] & H. J. DENGLER[2]
[1]Division of Clinical Pharmacology, Clinical Research Institute, National Medical Center, Tokyo, Japan, [2]Department of Medicine, University of Bonn, Bonn, FRG, and [3]Department of Pharmacology, Yamanashi Medical College, Yamanashi, Koufu City, Japan

The metabolism of sparteine which exhibits a genetic polymorphism in Caucasians was studied in 84 unrelated Japanese subjects. In contrast to a recent study where debrisoquine was used as a probe and no poor metabolizers could be observed in Japanese involving 100 subjects, two subjects had a urinary metabolic ratio of sparteine > 20 and thus were poor metabolizers of sparteine. The incidence of poor metabolizer phenotype of sparteine oxidation of 2% seems to be lower in Japanese as compared with various Caucasian populations where 5 to 10% are poor metabolizers of sparteine. However, this is not conclusive, because the 95% confidence interval of the observed frequency, 0.6 to 8%, covers the range reported in the literature for Caucasians.

Keywords genetic polymorphism sparteine oxidation Japanese

Introduction

A close relationship between polymorphic oxidation of sparteine and debrisoquine has been observed in German, Swedish (Bertilsson *et al.*, 1980; Eichelbaum *et al.*, 1982), British (Price-Evans *et al.*, 1983) and Canadian populations (Inaba *et al.*, 1980), suggesting that the oxidative metabolism of these two drugs is under common genetic control. However, Woolhouse *et al.* (1985) have shown that the close correlation between sparteine and debrisoquine metabolism that exists among Caucasians is lacking among Ghanaians, providing evidence of a dissociation of co-regulatory control of debrisoquine–sparteine oxidation among different ethnic groups. Recently, Nakamura *et al.* (1985) have reported pronounced interethnic differences in the oxidative metabolism of debrisoquine and mephenytoin between white American and Japanese subjects. In 183 white Americans the frequency of poor metabolizers (PM) for 4-hydroxylation of debrisoquine was 8.7% and for 4-hydroxylation of mephenytoin was 2.7%, respectively. In contrast, in 100 Japanese subjects, no PMs of debrisoquine could be identified, whereas the incidence of PMs of mephenytoin was 18%. These substantial differences in oxidative metabolism may have implications for the therapeutic efficacy and toxicity of drugs that are metabolized by the cytochrome P-450 isozymes involved (Eichelbaum, 1982, 1984; Idle & Smith, 1984).

Due to the lack of data concerning polymorphic oxidation of sparteine in Japanese and in view of the above findings the metabolism of sparteine was studied in a Japanese population.

Methods

Eighty-four Japanese subjects (50 medical students, 34 members of laboratory and hospital staff; 10 females, 74 males; aged 21 to 43 years)

*Present address: Dr Margarete Fischer-Bosch-Institut für Klinische Pharmakologie, 7000 Stuttgart 50, FRG
Correspondence: Dr Takashi Ishizaki, Division of Clinical Pharmacology, Clinical Research Institute, National Medical Center, Toyama 1-21-2, Shinjuku-ku, Tokyo 162, Japan

participated in the study. After emptying their bladder, each took by mouth 100 mg of sparteine sulphate (Depasan® tablet, Giulini Pharma, Hannover, West Germany) following an overnight fast. They had breakfast 2 h later. During the following 6 h urine was collected, the volume measured, and an aliquot was stored at −20° C until analyzed. Since in contrast to our previous phenotyping procedure where urine was collected for a 12 h period, the sampling period in this study was only 6 h, a sample of 52 German subjects was included. In this group of German subjects urine was collected from 0–6 and 6–12 h in order to evaluate whether the metabolic ratio might be different with different sampling periods.

Sparteine and its 2- and 5-dehydrometabolite were analyzed in urine by the method of Eichelbaum *et al.* (1979), with a described modification (Eichelbaum *et al.*, 1982). PMs of sparteine were defined as subjects with a urinary metabolic ratio (MR) greater than 20 (Eichelbaum *et al.*, 1982, 1986) which is calculated from: (amount of sparteine/(amount of 2- plus 5-dehydrosparteine).

Results

Comparison of the MR obtained in the 52 German subjects with urine collection periods of 0–6 and 0–12 h showed a highly significant correlation between the two collection periods ($P < 0.001$, $r^2 = 0.98$, $y = 1.04\,x + 0.03$, where y = MRs from 0–12 h samples and x = MRs from 0–6 h samples). Almost identical MRs were obtained when the MRs of the two collection periods were compared. Thus, an influence of collection periods on the phenotype assignment can be excluded. Three of the 52 German subjects had a MR > 20 and, therefore, were PMs. This corresponds to frequency of the PM phenotype of 6% (the 95% confidence interval, 2 to 16%).

The frequency distribution histogram of $\log_{10}$ MR of sparteine in the 84 Japanese subjects is shown in Figure 1. Two (both males, 24 and 28 years old) of the 84 subjects had a MR > 20 (30.0 and 83.9) and thus were PMs. This corresponds to a frequency of the PM phenotype of 2% with a confidence interval of 0.6 to 8% at the 95% level. Based upon the data of this sample size, the frequency of the allele controlling deficient sparteine metabolism is estimated to be 0.154 and for the allele controlling metabolism to be 0.846. The expected genotype frequencies for homozygous PMs are 0.024, heterozygous metabolizers 0.261 and homozygous metabolizers 0.715.

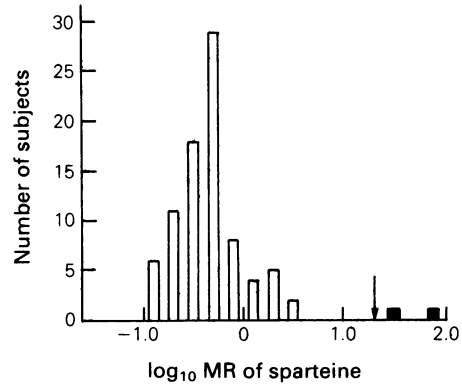

Figure 1 Frequency distribution histogram of $\log_{10}$ metabolic ratio (MR) of sparteine in 84 unrelated Japanese subjects.

The arrow indicates the boundary value ($\log_{10}$ MR = 1.3) to discriminate metabolizers according to the criteria (Eichelbaum *et al.*, 1982, 1986).

The mean cumulative urinary excretion of sparteine and its dehydrometabolites for the 82 EMs and the 2 PMs are listed in Table 1. The urinary recovery (0–6 h) of sparteine and 2- and 5-dehydrosparteine in 82 Japanese was almost identical to the data in 49 German EMs. Both the two Japanese and three German PMs excreted less than 1.5% of the dose as dehydrosparteines and more than 41% as unchanged drug within 6 h (Table 1).

Discussion

The Japanese population appears to be unique in terms of not only polymorphic *N*-acetylation but also polymorphic drug oxidation. Compared with Caucasians where 40 to 50% are rapid acetylators, as much as 90% of the Japanese belong to the rapid acetylator phenotype (Sunahara *et al.*, 1961). The frequency of PMs of mephenytoin was reported to be 18% (Nakamura *et al.*, 1985) and 22.6% (Jurima *et al.*, 1985) in a Japanese population, whereas it was 2.7% in a white American (Nakamura *et al.*, 1985) and 4.2% (Jurima *et al.*, 1985) in a Canadian population. However, no PMs of debrisoquine were observed among the 100 Japanese subjects studied by Nakamura *et al.* (1985). Since a close relationship has been observed between polymorphic debrisoquine and sparteine oxidation in various Caucasian populations, one would expect in view of the findings by Nakamura *et al.* (1985) that no PMs of sparteine might be present in Japanese.

Table 1 Cumulative urinary excretion of sparteine (S) and 2- and 5-dehydrosparteine (2-DH and 5-DH) in EM and PM subjects

	Phenotype	*S*	*5-DH*	*2-DH*	*S + 2-DH and 5-DH*
Japanese (n = 82)	EM	15.9 ± 7.5	5.9 ± 1.9	28.2 ± 10.2	50.0 ± 10.5
German (n = 49)	EM	16.5 ± 9.4	5.7 ± 2.3	30.4 ± 13.2	52.5 ± 13.2
Japanese	PM				
	1	43.9	0.29	1.18	45.4
	2	41.9	0.03	0.47	42.4
German	PM				
	1	47.9	0.15	1.15	48.9
	2	44.9	0.1	0.7	45.7
	3	44.1	0.1	0.8	45.0

Values for EMs are presented as means ± s.d. For PMs the individual values are given.

Contrary to the above assumption, the metabolism of sparteine in Japanese seems to be polymorphic (Figure 1) as in various Caucasian populations. However, the identification of the PMs in this study is based upon the criteria of the MR derived from German population (Eichelbaum *et al.*, 1982, 1986). Whether extrapolating the antimode from one population to another is valid remains uncertain. In spite of this limitation the observed frequency of 2% seems to indicate a lower frequency of the deficiency allele in Japanese as compared with various Caucasian populations, where the frequency of the PM phenotype ranges from 5 to 10% (Vinks *et al.*, 1982; Eichelbaum & Woolhouse, 1985; Clark, 1985; Eichelbaum *et al.*, 1986). Nonetheless, the rather small sample population investigated so far does not allow any firm conclusion to be drawn with regard to the allele frequency. The apparent difference in the incidence of the PM phenotype for debrisoquine and sparteine may have resulted from either the very low incidence and consequently small sample sizes studied or dissociation of co-regulatory control in Japanese subjects. However, by considering the present observation coupled with that of Nakamura *et al.* (1985), the possibility that Japanese might be a population showing a dissociation in the genetic control of debrisoquine/sparteine oxidation as indicated in a Ghanaian population study by Woolhouse *et al.* (1985) cannot be denied. Obviously, a crossover study on debrisoquine and sparteine metabolism in an extended Japanese population is required in order to prove or reject this hypothesis and such a study is currently underway.

This study was supported by a grant from the Sandoz Stiftung für Therapeutische Forschung. The skillful technical assistance by Mrs J. Caspar and S. Hoffmeister and the secretarial assistance by Miss K. Narita are gratefully appreciated.

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(Received 3 September 1986, accepted 14 November 1986)