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The *N*-oxidation of trimethylamine in a Jordanian population

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The ability to oxidise trimethylamine (TMA) to trimethylamine *N*-oxide (TMAO) is distributed polymorphically within a British white population with the majority of individuals excreting greater than 90% of total urinary TMA as TMAO. The opposite extreme is characterised by a rare inborn error of TMA *N*-oxidation known as the fish-odour syndrome. However there is a lack of information regarding inter-individual variability in the *N*-oxidation of TMA in other ethnic groups. In this study the urinary excretion of TMA and TMAO was determined over a period of 24 h in 82 Jordanian subjects. A frequency distribution histogram of % of total urinary TMA excreted as TMAO revealed that the majority of subjects excreted greater than 80% of the total urinary TMA as TMAO, however eight subjects (9.7%) excreted less than 80% of the total TMA as TMAO. In a previous study of 169 white British subjects only one (0.6%) excreted less than 80% of the total TMA as TMAO. The results suggest that the prevalence of compromised ability to *N*-oxidise TMA may be higher in a Jordanian population than in a British population.

Keywords trimethylamine *N*-oxidation Jordanian

Introduction

Trimethylamine (TMA) is a volatile tertiary amine which is derived from the bacterial degradation of choline and carnitine, which are common food components, and trimethylamine *N*-oxide (TMAO), which is found in high levels in fresh marine fish [1, 2]. As a consequence of its dietary origin, individuals are exposed daily to significant amounts of TMA, the majority of which is normally oxidised to the odourless TMAO and as such TMA and TMAO are normal components of human urine [3].

The ability to *N*-oxidise TMA to TMAO has been shown previously to be polymorphically distributed within a British white population, with the majority of individuals excreting greater than 90% of the total TMA in the form of TMAO [4]. However a condition trimethylaminuria, known colloquially as the fish-odour syndrome, exists in which affected individuals have a compromised ability to *N*-oxidise TMA and as such excrete free TMA in their breath, sweat, urine and other bodily secretions which results in a characteristic fish-like odour [6, 7]. Such individuals appear to be homozygous for an allele which determines an impaired ability to perform this *N*-oxidation reaction [4, 8, 9]. Carriers (heterozygotes) of this defect can be identified on the basis of a compromised ability to *N*-oxidise TMA under normal dietary conditions and after oral challenge with a dose of TMA sufficient to saturate their *N*-oxidation capacity [9]. Using these methods the prevalence of heterozygotes in a British white population appears to be at least 1% [4].

The *N*-oxidation of TMA is mediated by the flavin-containing monooxygenase (FMO) system which is responsible for the oxidation of many nitrogen and sulphur-containing xenobiotics, e.g. imipramine and methimazole [10]. However unlike other pathways of drug metabolism which show polymorphic expression there is a lack of information regarding inter-individual variability in the *N*-oxidation of TMA in any ethnic group other than British. In this study we have investigated the *N*-oxidation of dietary derived TMA in a Jordanian population.

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Methods

The volunteers studied were 82 (24 female) unrelated, apparently healthy subjects of Jordanian ancestry aged 20–60 years (median = 23 years), attending Jordan University of Science and Technology as students or employees. All participants gave their written informed consent to the study which was approved by the local University Ethics Committee, and none was taking any medication at the time of the study.

The volunteers were asked not to eat fish for the 2 days prior to the test and on the day of the urine collection. Volunteers collected all the urine produced in a 24 h period into polypropylene bottles which contained hydrochloric acid (10 ml; 4 M). The urine volume was recorded and an aliquot (20 ml) stored frozen (–20° C) until assayed for TMA and TMAO by head-space gas chromatography [4]. Intra-assay variation was calculated by multiple analyses of a single spiked urine sample in a single assay and gave a coefficient of variation of 7.2% (n = 5) at a concentration of 10 μg ml^{-1}. Inter-assay variation was calculated by repeated analysis of a single spiked urine sample on sequential days and produced a coefficient of variation of 8.1% (n = 6). The lower limit of determination of TMA was 0.01 μg ml^{-1} and calibration curves were linear up to a concentration of 200 μg ml^{-1}.

All urine samples were collected during the period April to July 1992.

Results

The results obtained for the 82 subjects under investigation are shown in Table 1. Figure 1 illustrates the data presented in the form of a frequency distribution histogram of % of total urinary TMA excreted as TMAO. It can be seen that the majority of subjects (74 or 90.2%) excreted greater than 80% of the total TMA as TMAO. However five subjects excreted between 70 and 80%, two subjects excreted between 60 and 65% and one subject excreted 53.5% of the total TMA as TMAO.

Discussion

The results of this study indicate that within a population of Jordanians the ability to N-oxidise TMA is skewed. Although the majority of subjects investigated excreted greater than 80% of the total TMA in urine as TMAO, eight individuals excreted less than 80% of the total urinary TMA as TMAO. However none of the subjects studied appeared to be clinically trimethylaminuric. The predominance of urinary TMAO as a metabolite of TMA is consistent with observations made in a previous study in a British white population [4]. However in the British population only one out of 169 of the subjects excreted less than 80% of the total urinary TMA as TMAO under normal dietary conditions [4]. This subject was identified as a carrier of the N-oxidation defect on the basis of an impaired ability to convert TMA to TMAO after oral challenge with 600 mg TMA. Similarly a subject who excreted 84.1% of the total urinary TMA as TMAO under normal dietary conditions was also identified as a carrier on the basis of this test. Thus out of a population of 169 British white subjects only two were identified as probable heterozygotes for trimethylaminuria.

No data exist for intra-individual variability in the Jordanian subjects and although we recognise this as a deficiency in the present study, analysis of urine samples from British white subjects using the head-space gas chromatography method employed in the present study demonstrated that intra-individual variability in % of total urinary TMA excreted as TMAO was negligible (coefficient of variability < 1%). The mean values for absolute amounts of TMA and TMAO excreted in urine in the Jordanian population were approximately two-fold higher than those excreted by the British white population previously studied [4]. Although this increased load of TMA

Table 1 Urinary excretion of trimethylamine (TMA) and trimethylamine N-oxide (TMAO) during a 24 h period expressed as mg and μmol in 82 unrelated Jordanian subjects

	Range	*Median*	*Mean ± s.d.*
TMA (mg)	0.4–30.3	2.3	3.6 ± 4.6
TMA (μmol)	6.5–511.8	38.4	61.8 ± 78.6
TMAO (mg)	4.0–708.9	33.5	74.6 ± 117.9
TMAO (μmol)	53.6–9439.4	446.0	992.9 ± 174.5

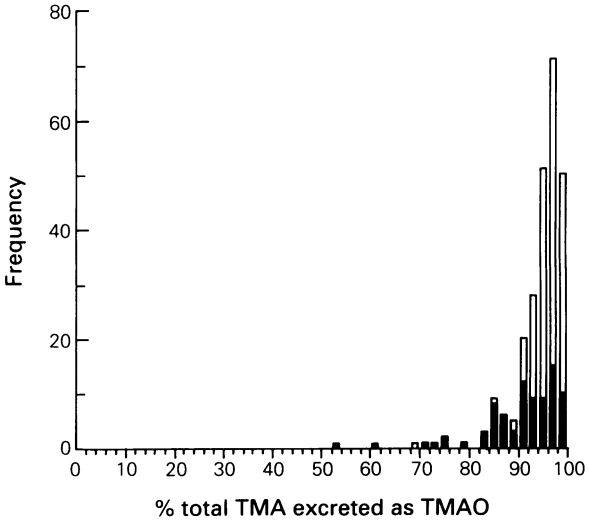

Figure 1 Frequency distribution histogram of percent of total trimethylamine (TMA) excreted in urine as trimethylamine N-oxide (TMAO) during a 24 h period in 82 unrelated Jordanian subjects (filled bars) and in 169 unrelated healthy British white volunteers (open bars) re-drawn from Al-Waiz *et al.*, 1987 [4].

available for *N*-oxidation might be expected to account for the apparently higher prevalence of compromised ability to produce TMAO in the Jordanian population, no relationship between % of total urinary TMA excreted as TMAO and absolute amounts of TMA + TMAO excreted was observed. The eight Jordanian subjects who excreted less than 80% of the total TMA as TMAO were not investigated after oral challenge with TMA and thus not positively identified as carriers of the *N*-oxidation defect by this method. As a consequence it is not possible to conclude that the inter-individual variability in capacity to *N*-oxidise TMA seen in this Jordanian population is due to genetic polymorphism in this *N*-oxidation pathway. Nevertheless since there is no evidence of misclassification of individuals who excrete less than 80% of the total TMA as TMAO from previous studies, the data suggest that these individuals may be heterozygous for the allele which determines an impaired ability to *N*-oxidise TMA.

Irrespective of the mechanism underlying this defect the results obtained in the present study contrast sharply with those obtained for the British white population and suggest that within this Jordanian population the prevalence of compromised ability to *N*-oxidise TMA is higher than in the British white population previously studied [4].

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