The effect of thyrotoxicosis on isoniazid acetylation

M. D. LITTLE¹, A. HUTCHINGS¹, B. P. SPRAGG², P. A. ROUTLEDGE³ & J. H. LAZARUS¹

Departments of Medicine¹, Toxicology², and Clinical Pharmacology³, Llandough Hospital, Penarth, South Glamorgan

Isoniazid acetylation was assessed in 10 thyrotoxic patients before and after standard anti-thyroid therapy. The group contained five fast and five slow acetylators and all remained within the same phenotypic classification when rendered euthyroid. There was no significant change in the elimination half-life of isoniazid between thyrotoxicosis and euthyroidism for the group as a whole or for fast and slow acetylators considered separately. Thyrotoxicosis does not appear to be an important determinant of isoniazid acetylation in man.

Keywords isoniazid acetylation thyrotoxicosis

Introduction

Genetic polymorphism of drug acetylation in man is well recognised (Price Evans *et al.*, 1960). The autosomal single gene inheritance produces three sub-populations representing homozygous fast, heterozygous fast and homozygous slow. The former two are not clearly distinguished by most studies and for practical use the population is divided into fast and slow only. In the UK, 45% of the population are fast acetylators, but the proportion varies widely in different parts of the world (Evans, 1969; Jeanes *et al.*, 1972; Hashem *et al.*, 1969).

More recently, the effects of some non-genetic factors on acetylator function have been studied. Ethanol loading (Olsen & Morland, 1978) and glucose loading (Thom *et al.*, 1981) in man have both been shown to enhance acetylation. In contrast, reduced acetylation has been demonstrated in renal failure (Gold *et al.*, 1976) and hepatic disease (Levi *et al.*, 1968). However, the effects in these studies have been quantitatively less than those produced by the genetic influences.

Acetylation occurs primarily in the liver, with a limited amount in the wall of the small intestine (Hearse & Weber, 1970). Alterations in hepatic metabolism might be important in determining acetylator function and it is clear that the pharmacokinetics of some drugs are very different in

states of thyroid dysfunction (Shenfield, 1981). There appears however to be no published data regarding thyrotoxicosis and acetylation.

Oral isoniazid has been widely used for the determination of acetylator function. It is rapidly and almost completely absorbed in the fasting state. New assay methods allow the use of a lower dose, and therefore a lower risk of adverse effects (Hutchings & Routledge, 1983a). We therefore assessed isoniazid acetylation in 10 thyrotoxic subjects, before and after antithyroid treatment to euthyroidism.

Methods

Subjects

Ten thyrotoxic patients were interviewed at the time of first outpatient visit. Five male and five female patients volunteered (age 21–66 years, mean 45 ± 4.5) and gave informed, written consent to the study which had been approved by the local Medical Ethics Committee. No patient had any other significant illness and all had normal renal and hepatic function. Thyrotoxicosis was confirmed by the finding of elevated free thyroxine (FT₄) and free triiodothyronine

(FT₃) in serum. Isoniazid acetylation was assessed twice in each patient – when thyrotoxic and again when euthyroid.

The study was performed following a 9 h fast. Blood samples were drawn from a cannula in an antecubital vein at 15, 30, 45, 60, 90, 120, 180, 240, 300 and 360 min. Isoniazid (200 mg) was given orally as 2 × 100 mg tablets with 50 ml water at time zero.

Because of instability of isoniazid in blood, (Hutchings *et al.*, 1983b) all specimens were centrifuged and separated immediately, then the plasma stored at –20°C till the end of the 6 h study period. All specimens were then transferred to –70°C storage pending isoniazid analysis.

Following the first study, antithyroid treatment was commenced and the patients were reviewed at intervals until clinically and biochemically euthyroid. The study as above was then repeated. Antithyroid therapy with carbimazole, propylthiouracil or propranolol was discontinued where necessary for 72 h before repeat testing.

Five of the 10 patients studied were treated with a single dose of radioactive sodium ¹³¹I iodide orally followed by carbimazole and the remainder received carbimazole only. Short term therapy with propranolol was also used in some patients. One patient developed a macular rash on carbimazole which resolved when he was changed to propylthiouracil. There were no other adverse drug reactions, in particular none related to isoniazid.

Analytical methods

Free thyroxine (FT₄) and triiodothyronine (FT₃) were measured by Amerlex Kit (Amersham International plc) in baseline samples from the study before and after antithyroid therapy. TSH was measured by a sensitive two site immunoradiometric assay (John & Jones, 1984). Isoniazid and acetyl isoniazid were assayed by high performance liquid chromatography (Hutchings *et al.*, 1983b). Samples stored at –70°C have been shown to retain full isoniazid activity for 5 weeks and all were analysed within this period (Hutchings *et al.*, 1983b).

Statistical tests

The elimination half-life of isoniazid was calculated by regression analysis of log isoniazid concentrations over the 6 h study period. Results were compared by Wilcoxon test or Student's *t*-test when appropriate.

Results

Thyrotoxicosis was confirmed by the finding of elevated FT₄ levels (mean ± s.d. 69.6 ± 33.6, range 22.7–139, normal 8–26 pmol l⁻¹) and/or FT₃ levels (mean 26.1 ± 11.5, range 11.6–43.8, normal 3–9 pmol l⁻¹). This was further confirmed by suppressed TSH levels. Following antithyroid

Table 1 Elimination half-life and acetyl isoniazid/isoniazid ratio at 180 min (A/I 180) of 10 patients when thyrotoxic and when euthyroid

	Patient	Treatment	Pre-treatment		Euthyroid	
			t _{1/2} (min)	A/I 180	t _{1/2} (min)	A/I 180
Slow	1	¹³¹ I	134	0.70	156	0.89
	2	¹³¹ I	276	0.26	264	0.28
	3	carb	172	0.26	226	0.34
	4	carb	147	0.38	202	– *
	5	¹³¹ I	230	0.41	240	0.44
	Mean		191.8	0.40	217.6	0.49
	Median		172	0.38	226	0.39
	Range		134–276	0.26–0.7	156–264	0.28–0.89
Fast	6	carb	72	3.55	60	3.21
	7	¹³¹ I	78	3.27	94	5.15
	8	carb	92	2.77	77	6.41
	9	¹³¹ I	122	3.24	114	2.51
	10	carb	93	2.59	94	3.12
	Mean		91.4	3.08	87.8	4.08
	Median		92	3.24	94	3.21
	Range		72–122	2.59–3.55	60–144	2.51–6.41

carb = carbimazole; ¹³¹I = radioiodine therapy. For details see text. Patients treated with carb stopped therapy 72 h before test. * 180 min specimen lost.

treatment, all patients were clinically euthyroid and all but one had thyroid hormone levels within the normal range. This patient had received radioiodine therapy and became biochemically hypothyroid between his outpatient visit and being able to return for repeat study.

Repeat acetylator function was performed 97 to 263 days after the first study (mean 181 days), patients being assessed after all medication had been discontinued for 72 h. Isoniazid half-lives ranged from 76 to 276 min when thyrotoxic and 60 to 264 min following therapy (Table 1). There was no significant change in half-life for the group as a whole ($P > 0.05$ Wilcoxon test) between thyrotoxicosis and euthyroidism. In addition the mean half-life of slow and fast acetylators did not differ significantly before and after therapy when considered separately (see Figure 1). Similarly, the acetyl isoniazid/isoniazid (AcINH/INH) ratio in the 3 h sample did not differ significantly between the hyperthyroid and euthyroid states. All patients remained within the same phenotypic classification before and after treatment (five fast, five slow) whether this was assessed by half-life or AcINH/INH ratio (Hutchings & Routledge, 1986).

Discussion

Thyrotoxicosis is well known to affect metabolism and more specific pharmacokinetic studies have shown enhancement of microsomal drug oxidation (Eichelbaum, 1976). An extensive review of drug disposition in relation to thyroid disease (Shenfield, 1981) provides further evidence from a number of authors but only microsomal oxidation appears to have been studied in detail. In the intact cell, oxidation occurs in the smooth endoplasmic reticulum whereas the enzyme *N*-acetyl transferase is found in the cytosol. This difference in location may explain the apparent differences between the two metabolic pathways for oxidation and acetylation. Although the activity and reaction rate of malic enzyme, located in the cytosol, is dependent on the intracellular con-

Figure 1 Elimination half-lives (min) of isoniazid before and after treatment of thyrotoxicosis.

centration of triiodothyronine (Goodridge, 1983), there is no evidence that other cytosolic enzymes are under thyroid hormone control.

It should be noted that the elimination half-life of isoniazid increased in four of the five slow acetylators in this study and because of the numbers involved it is not possible to exclude the possibility that a real effect may occur in this group (i.e. a type II error has been made). Nevertheless, no subject changed his or her phenotype, determined either by isoniazid half-life of elimination or by the ratio of acetyl isoniazid to isoniazid in the 3 h sample (Hutchings & Routledge, 1986). It is therefore unlikely that any small effect missed in this study will be clinically relevant in view of the marked differences attributable to genetic influences of isoniazid metabolism.

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