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LETTERS TO THE EDITOR

Kinetics of some pyrazole derivatives in the rat

The inhibitory effect of pyrazole and derivatives of pyrazole on the activity of alcohol dehydrogenases, of the microsomal ethanol-oxidizing system and of other microsomal enzymes has been extensively studied, both *in vitro* and *in vivo* (for reviews see Reynier 1969; Lieber, Rubin & others, 1970; Rydberg, 1972). However, little is known about the distribution and elimination of these compounds from the body. When high doses of pyrazole were given together with ethanol to rats, an inhibitory effect on ethanol oxidation could be detected for 2–3 days (Goldberg & Rydberg 1969). Using a spectrophotometric method to determine pyrazole, Lester & Benson (1970) estimated its half-life to be about 14 h in the rat. From the inhibitory effect on ethanol elimination, Goldstein & Pal (1971) estimated the half-life of pyrazole in mice to be about 10 h, and for 4-bromopyrazole less than 3 h. Blomstrand & Theorell (1970) reported that the inhibitory effect of 4-methylpyrazole lasted for 36 h in the rat.

Since methods for the determination of pyrazole in blood have not been available, measurements of the half-life have been based on indirect estimations. We have used a recently introduced g.l.c. method (Rydberg & Buijten 1972) applied to determine pyrazole and 4-methylpyrazole in blood.

Male rats of the Sprague-Dawley strain received pyrazole (1·5 mmol/kg, 2% in saline), without and with ethanol (32·6 mmol/kg, 14% in saline); or 4-methylpyrazole hydrochloride (0·84 mmol/kg, 2% in saline), without and with ethanol (32·6 mmol/kg, 14% in saline) by intraperitoneal injection. Blood samples (50 μl) were withdrawn from the tip of the tail and were diluted with 1 ml of 0·9% (w/v) sodium fluoride solution. Pyrazole and 4-methylpyrazole were determined by g.l.c. using a column of 5% Carbowax 20 M on acid-washed Chromosorb W at 120°. An F & M gas chromatograph model 609 was used. For further details, see Rydberg & Buijten

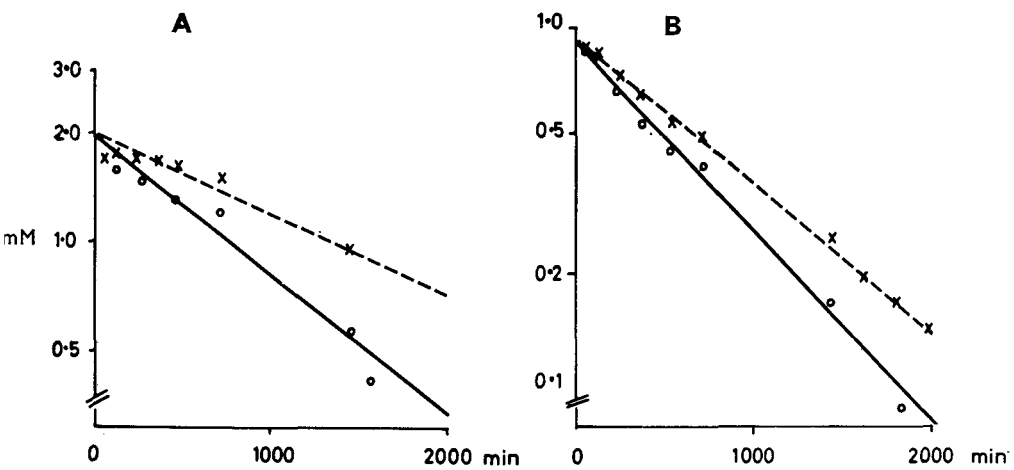

FIG. 1.A. The concentration of pyrazole (log scale) in rat blood after i.p. injection of pyrazole (1·5 mmol/kg or 100 mg/kg) without (o) and together with (x) ethanol (32·6 mmol/kg; 1·5 g/kg).

B. The concentration of 4-methylpyrazole (log scale) in rat blood after i.p. injection of 4-methylpyrazole (0·84 mmol/kg; 100 mg/kg) without (o) and together with (x) ethanol (32·6 mmol/kg. 1·5 g/kg).

(1972). Ethanol was determined in 10 μl blood samples according to the ultramicro distillation method (Buijten, unpublished.)

After doses of pyrazole or 4-methylpyrazole to rats substances with the gas chromatographic properties of pyrazole and 4-methylpyrazole were present in the blood. No other peaks were seen. The mean maximal concentration of pyrazole was 1·75 mM (Fig. 1A), and of 4-methylpyrazole 0·84 mM (Fig. 1B). No difference in maximal concentration was seen when ethanol was given.

The half-life of pyrazole in blood was 796 $\pm$ 53 min (mean $\pm$ s.e.), Fig. 1A. When ethanol was given, this increased to 1285 $\pm$ 107 min. The results were submitted to a two-tailed *t*-test. The difference was significant (d.f. = 7, $P < 0{\cdot}01$). The half-life of 4-methylpyrazole in blood was 544 $\pm$ 28 min (Fig. 1B). When ethanol was administered, this time increased to 731 $\pm$ 51 min. The difference was statistically significant (d.f. = 8, $P < 0{\cdot}02$). When ethanol was given together with pyrazole, the ethanol concentration was zero after 770 min, when given with 4-methylpyrazole the ethanol concentration was zero after 1790 min.

The results indicate that the half-life of pyrazole is longer than that of 4-methylpyrazole. The elimination of both substances was inhibited by ethanol. The mechanism for the elimination of pyrazole and 4-methylpyrazole by excretion and metabolism was not further investigated. Pyrazole is known to inhibit hepatic microsomal drug metabolizing enzymes, and binds to the haemoprotein resulting in a "type 2 spectrum" (Rubin, Gang & Lieber, 1971). Since ethanol is a potent inhibitor of compounds producing a type 2 spectrum (Rubin, Misra & others, 1970), it might inhibit a microsomal mechanism for elimination of pyrazole and derivatives.

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