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Experimental gastric ulcers and stomach tissue pepsin activity in the rat

Recently we demonstrated a relation between uropepsinogen excretion and experimental gastric ulcers in the rat (Coppi, Bonardi & Gaetani, 1971). We now report the pepsin activity of the stomach tissue of normal rats, of rats with reserpine or phenylbutazone-induced ulcers and in ulcerated rats also treated with atropine or oxyphencyclimine.

Male or female Sprague-Dawley rats, 160–170 g, had gastric ulcers produced by reserpine or phenylbutazone (Coppi & others, 1971). Groups of normal rats or with reserpine- or phenylbutazone-induced ulcers were treated orally with atropine sulphate or oxyphencyclimine (hydrochloride) both suspended in 5% acacia gum. The stomachs were removed 18 h after the reserpine or the last phenylbutazone dose, washed in running water, dried between filter paper, weighed, homogenized (1%) in 0·06N HCl by a blender (Sorvall Omni-Mixer) and used for pepsin assay (Anson, 1963). A portion of this homogenate was centrifuged at 3000 rev/min for 20 min

Table 1. *Relation between gastric ulcers and stomach tissue pepsin activity in the rat.* The pepsin activity and the morphological examination was made on rats 18 h after the reserpine or the second phenylbutazone dose. Atropine and oxyphencyclimine were given simultaneously with the reserpine or the second phenylbutazone dose.

Treatment	Pepsin activity (mg tyrosine/g N) mean ± s.e.		Morphological examination	
	1% Homogenate	Supernatant	Ulceration index mean ± s.e.	% Ulcerated animals
5% Acacia gum	13·6 ± 0·8 (20)	27·5 ± 1·7	0 (40)	0
Reserpine (5 mg/kg, i.p.)	5·9 ± 0·5 (20)†	9·0 ± 0·8†	2·0 ± 0·2 (39)†	93
Reserpine (5 mg/kg, i.p.) + atropine (10 mg/kg, orally)	5·6 ± 0·7 (19)†	10·7 ± 1·1†	1·5 ± 0·2 (40)†	75
Reserpine (5 mg/kg, i.p.) + oxyphencyclimine (10 mg/kg, orally)	9·7 ± 1·0 (20)†*	18·4 ± 2·2†*	0·5 ± 0·1 (40)†*	38
5% Acacia gum	17·4 ± 0·9 (27)	34·0 ± 1·9	0·2 ± 0·1 (30)	17
Phenylbutazone (100 mg/kg, orally twice in 8 h)	13·0 ± 1·0 (26)†	26·1 ± 2·1†	3·1 ± 0·2 (28)†	100
Phenylbutazone (100 mg/kg, orally twice in 8 h) + atropine (10 mg/kg orally)	17·3 ± 1·3 (26)*	36·4 ± 2·8*	1·8 ± 0·3 (29)†*	77
Phenylbutazone (100 mg/kg, orally twice in 8 h) + oxyphencyclimine (10 mg/kg, orally)	21·3 ± 1·1 (27)†*	43·6 ± 2·4†*	1·5 ± 0·3 (30)†*	60
5% Acacia gum	15·8 ± 1·3 (15)	31·7 ± 2·7	0·5 ± 0·2 (31)	36
Atropine (10 mg/kg, orally)	15·4 ± 1·5 (15)	29·1 ± 2·8	0·3 ± 0·1 (30)	23
Oxyphencyclimine (10 mg/kg, orally)	21·7 ± 1·2 (19)†	44·0 ± 3·0†	0 (30)†	0

* Significance relative corresponding control group; † significance relative to reserpine or phenylbutazone-treated group: *t*-test, ($P \leqslant 0{\cdot}01$).

Numbers of animals are in parentheses.

and the supernatant also assayed for pepsin. Portions of homogenate and of supernatant were also assayed for nitrogen (Kjeldhal). Further groups of rats were similarly treated and the number of animals with gastric lesions and the severity of these graded by an arbitrary scale from 0 to 4+ was recorded.

The pepsin activity of stomach tissue is always lower in the ulcerated animals (Table 1). Oxyphencyclimine and, to a lesser extent, atropine reduced the ulceration index and simultaneously increased the stomach tissue pepsin activity of the animals with reserpine- and phenylbutazone-induced ulcers.

In these groups there was a good relation between the enzyme activity and the severity of gastric lesions although the two parameters were not always proportionally related.

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Protection against phenylisothiocyanate by various steroids, phenobarbitone and diphenylhydantoin

Various arylisothiocyanates, including α-naphthylisothiocyanate and phenylisothiocyanate, cause serious morphologic and functional alterations in the liver (Becker & Plaa, 1965; Plaa, Rogers & Fouts, 1965). The fact that certain isothiocyanates exhibit strong antimicrobial and carcinostatic activities (Horáková, Drobnica & others, 1968a,b) raised interest in this group of compounds. A short time ago, we were able to demonstrate that a series of catatoxic steroids (known to induce hepatic microsomal drug-metabolizing enzyme synthesis), like phenobarbitone (a non-steroidal enzyme inducer), protect the rat against otherwise lethal doses of the α-naphthyl compound (Selye, 1971). Hence, it seemed of interest to establish whether such compounds would also be effective against phenylisothiocyanate.

The techniques were those previously described (Selye, 1971). Female Sprague-Dawley rats, 100 g, were divided into 16 groups and treated as outlined in Table 1. The steroids were administered at 10 mg in 1 ml water (homogenized with a trace of Tween 80), and phenobarbitone sodium and diphenylhydantoin were given at the dose of 6 mg in 1 ml water, by stomach tube, twice daily. L-Thyroxine, as its sodium salt, was injected subcutaneously, once daily, at 200 μg in 0·2 ml water. On the fourth day of this treatment, all animals received 25 mg/100 g of phenylisothiocyanate in 1 ml corn oil orally. The degree of prostration was estimated as previously described (Selye, 1971) and the mortality rate registered on the 9th day after initiation of the experiment. The apparent differences between the control and pretreated groups were computed by the "Exact Probability Test" of Fisher and Yates (Finney, 1948; Siegel, 1956).

Phenylisothiocyanate intoxication is completely prevented by the most active catatoxic steroid known up to date, namely, pregnenolone-16α-carbonitrile (Table 1); however, considerable protection has also been obtained by steroids (previously shown to possess strong catatoxic activity against other substrates) such as 9α-fluoro-