## Studies on monoamine oxidase in rat spinal cord

## A.J. GEORGE & G.T. JONES

Pharmacology Department, School of Pharmacy, Liverpool Polytechnic, Liverpool, UK.

Monoamine oxidase (MAO) exists in at least two forms ('A' & 'B') which differ in their affinities for different amine substrates (Johnston, 1968). In this study, rat spinal cord and brain MAO activities were compared. Male rats (200-250 g) were killed and the

clorgyline and pargyline were investigated, with regard to enzyme and substrate specificity.

When tyramine is the substrate, clorgyline  $(1\times10^{-7}\,\mathrm{M})$ , a specific inhibitor of MAO 'A' (Johnston, 1968) causes a greater inhibition of MAO activity in spinal cord (80%) than in brain (41%). Pargyline  $(1\times10^{-7}\,\mathrm{M})$ , a specific MAO 'B' inhibitor (Squires, 1972), inhibited brain MAO activity by 63% and spinal cord MAO activity by 24% using tyramine as substrate. Since tyramine is a substrate for both MAO types, it appears that type A activity is proportionately greater in rat spinal cord than in rat brain.

Mean MAO activity as n moles product formed per mg protein h-1

	5-HT			Tyramine			Benzylamine		
	Control	С	P	Control	С	P	Control	С	P
Brain	20.62	2.31	17.97	9.64	3.70	6.10	7.21	6.98	1.05
	+ 1.98	+0.24	+ 1.83	+ 0.89	+ 0.29	+ 0.58	+ 0.78	+ 0.67	+ 0.09
Spinal cord	16.51	1.62	14.96	7.56	1.40	5.92	3.27	3.21	0.25
	<u>+</u> 1.67	± 0.18	± 1.32	± 0.71	± 0.13	± 0.60	± 0.36	± 0.34	± 0.23

C = clorgyline  $1 \times 10^{-7}$  M, P = pargyline  $1 \times 10^{-7}$  M Each result is the mean of 7 experiments.

spinal cord was removed, blotted, weighed, and then homogenized in ice-cold sucrose (0.25 M). Each homogenate was centrifuged at  $8 \times 10^2$  for 5 min and the precipitate retained and resuspended in phosphate buffer (pH 7.2). Brain MAO was prepared as described by Johnston (1968). To assess MAO activity, 0.1 ml of suspension was incubated at 37°C lumol of [14C]-tyramine 1 h with (0.3 μ Ci/μmol) and phosphate buffer pH 7.2. A blank was prepared by boiling a 0.1 ml sample of the suspension. The reaction was terminated with HCl and [14C]metabolite was extracted with ethyl acetate: benzene (1:1 v/v). 1 ml of extract was assayed for radio-activity (George & Leach, 1975) and the protein concentration of a 0.1 ml sample of suspension was determined as described by Lowry, Rosebrough, Farr & Randall (1951). The experiments were repeated using [14C]-5-HT and [14C]-benzylamine as substrates. The effects of the MAO inhibitors This work was supported by a grant from W. Warner and Co.

## References

GEORGE, A.J. & LEACH, G.D.H. (1975). The involvement of Ca<sup>2+</sup> and Mg<sup>2+</sup> in the spontaneous and drug induced release of <sup>3</sup>H-noradrenaline from mesenteric arteries. *Biochem. Pharmac.*, 24, 737-741.

JOHNSTON, J.P. (1968). Some observations upon a new inhibitor of monoamine oxidase in brain tissue. *Biochem. Pharmac.*, 17, 1285–1297.

LOWRY, O.H., ROSEBROUGH, N.J., FARR, A.L. & RANDALL, R.L. (1951). Protein measurement with the Folin phenol reagent. J. Biol. Chem., 193, 265-275.

SQUIRES, R.F. (1972). Multiple forms of monamine oxidase in intact mitochronia. In: *Monoamine Oxidases—New Vistas Biochemical Psychopharmacology*, Vol. 5, eds. Costa, E. & Sandler, M., pp. 355-371. Raven Press: New York.