References

CHRISTIANSEN, J. & SQUIRES, R.F. (1974). Antagonistic effects of apomorphine and haloperidol on rat striatal synaptosomal tyrosine hydroxylase. *J. Pharm. Pharmac.*, **26**, 367-369.

MILLER, R.J., HORN, A.S. & IVERSEN, L.L. (1974). The action of neuroleptic drugs on dopamine stimulated adenosine cyclic 3', 5'-monophosphate production in rat neostriatum and limbic forebrain. *Molec. Pharmac.*, **10**, 759-766.

SEEMAN, P. & LEE, (1975). Anti-psychotic drugs: direct correlation between clinical potency and a presynaptic action on dopamine neurones. *Science*, **188**, 1217-1219.

Monoamine oxidase activity in distinct populations of rat brain mitochondria

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Lai, Walsh, Dennis & Clark (1975) separated three distinct populations of mitochondria from rat brain by discontinuous Ficoll gradient. The mitochondria were metabolically active and relatively free of non-mitrochondrial material. Two of the mitochondrial populations (SM and SM2) were derived from synaptosomes and the remainder were 'free' mitochondria (M).

Johnston (1968) described two forms of monoamine-oxidase (MAO) in rat brain—type A and type B—on the basis of inhibition studies. Later work has shown that serotonin (5-HT) and phenylethylamine are preferentially deaminated by type A and type B MAO respectively, whereas tyramine is a substrate for both types of the enzyme. Type A MAO is relatively sensitive to clorgyline and type B to deprenyl.

Three populations of mitochondria were prepared by a modification of the procedure of Lai, Walsh, Dennis & Clark (1975). The three populations exhibited distinctly different MAO activities when assayed with 5-HT and phenylethylamine as substrates in a radiometric technique similar to that described by Robinson, Lovenberg, Keiser & Sjoerdsma (1968).

Table 1 MAO activity in mitochondrial populations

n	*Mitochondria*	*5-HT*	*Phenylethylamine*
8	M	51.6 ± 16,4	15.6 ± 7.5
8	SM	117.5 ± 16.6	31.7 ± 5.8
8	SM2	106.6 ± 3.0	51.8 ± 4.6

Results (mean ± s.d.) expressed as nanomoles product formed/mg protein per 30 min (analysis of a contingency table gave $\chi^2 = 48$ $P > 0.0001$).

Inhibition studies with clorgyline and deprenyl revealed that MAO of the lighter synaptosomal mitochondria (SM) was considerably more sensitive to inhibition by clorgyline than the M or SM2 mitochondria or a crude mitochondrial preparation. SM mitochondrial MAO was inhibited approximately 50% by 10^{-12} M clorgyline. The same population of mitochondria (SM) was the least sensitive to deprenyl although the differences in inhibition of MAO of the three sets of mitochondria were less pronounced with deprenyl than with clorgyline. When percentage inhibition of MAO activity was plotted against concentration of clorgyline or deprenyl double sigmoid curves resulted for all three mitochondrial populations with the plateaus suggesting A:B MAO ratios of about 4:1 for SM and 1.5:1 for M and SM2.

In agreement with previous findings (Youdim & Sourkes, 1965) heat inactivation at 50°C for 1 h resulted in the loss of about 15% of MAO activity of a crude mitochondrial preparation when tyramine was used as substrate. In contrast the purified mitochondria M, SM and SM2 lost approximately 75% of enzyme activity when similarly treated.

Attempts to fractionate further the SM mitochondria will be discussed.

References

JOHNSTON, J.P. (1968). Some observations upon a new inhibitor of monoamine oxidase in brain tissue. *Biochem. Pharmacol.*, **18**, 1447-1454.

LAI, J.C.K., WALSH, J.M., DENNIS, S.C. & CLARK, J.B. (1975). Compartmentation of citric acid cycle and related enzymes in distinct populations of rat brain mitochondria. Proceedings of the NATO Advanced Study Institute on Metabolic Compartmentation in the Brain (in press).

ROBINSON, D.S., LOVENBERG, W., KEISER, H. & SJOERDSMA, A. (1968). Effects of drugs on human blood platelet and plasma amine oxidase activity in vitro and in vivo. *Biochem. Pharmacol.*, **17**, 109-119.

YOUDIM, M.B.H. & SOURKES, T.L. (1965). The effect of heat, inhibitors and riboflavin deficiency on monoamine oxidase. *Can. J. Biochem.*, **43**, 1305-1318.