Biochemical Pharmacology, Vol. 31, No. 24, pp. 4061–4066, 1982.
Printed in Great Britain.

0006-2952/82/244061-06 $03.00/0
Pergamon Press Ltd.

MONOAMINE OXIDASE TYPE A: DIFFERENCES IN SELECTIVITY TOWARDS *l*-NOREPINEPHRINE COMPARED TO SEROTONIN

NANCY A. GARRICK*
Clinical Neuropharmacology Branch, NIMH, Bethesda, MD 20205, U.S.A.; and the Physiology Division, University of Maryland Department of Zoology, College Park, MD 20742, U.S.A.
and
DENNIS L. MURPHY
Clinical Neuropharmacology Branch, NIMH, Bethesda, MD 20205, U.S.A.

(*Accepted* 6 *May* 1982)

Abstract—*l*-Norepinephrine and serotonin have been regarded as preferential substrates for monoamine oxidase (MAO) type A. A close comparative examination of a number of tissues from different species, however, indicated the following differences. Serotonin was a more selective substrate for MAO-A, being inhibited by low concentrations ($< 10^{-7}$ M) of the irreversible MAO-A inhibitor, clorgyline, more consistently and to a greater extent (80–100%) than was *l*-norepinephrine (30–85%). These serotonin–norepinephrine differences were greater in humans and other primates than in rodents. Serotonin also had a 2- to 4-fold smaller apparent K_m for MAO-A than *l*-norepinephrine and was deaminated 2- to 5-fold more readily by MAO *in vitro* in most tissues. In contrast, the MAO-B in human platelets deaminated *l*-norepinephrine more readily than serotonin. Thus, *l*-norepinephrine, like dopamine, should be regarded as a substrate for both MAO-A and MAO-B *in vitro*. The prominent role of MAO-B in norepinephrine degradation in primates may need to be considered in interpreting laboratory and clinical studies of clorgyline and other selective MAO-inhibiting drugs.

l-Norepinephrine and serotonin are generally regarded as the two substrates for monoamine oxidase (MAO) which are selectively degraded by the MAO type A enzyme on the basis of (a) their sensitivities to inhibition by low concentrations of the specific MAO-A inhibitor, clorgyline, and (b) the localization of MAO-A in norepinephrine-rich areas (e.g. sympathetic neurons and locus coeruleus) [1–4]. Dopamine is also principally deaminated by MAO-A in rodent brain, although not in primate brain [5–8]. In contrast, most other monoamines, such as *p*-tyramine, tryptamine and kynuramine, are deaminated by both MAO-A and MAO-B, while a few trace amines, including β-phenylethylamine, phenylethanolamine, *o*-tyramine and tele-methylhistamine, are good substrates for MAO-B but poorer substrates for MAO-A [4, 9–12].

Most studies comparing MAO-A and MAO-B have used serotonin as the prototypical MAO-A substrate, and only a few investigations have directly evaluated the deamination of norepinephrine by MAO-A. Goridis and Neff [2] originally reported that *l*-norepinephrine deamination in rat superior cervical ganglia and brain was inhibited approximately 90% by low concentrations (10^{-7} M) of clorgyline. The simple sigmoidal clorgyline inhibition curves they obtained in these same tissues with serotonin as substrate were interpreted as indicating that serotonin and *l*-norepinephrine were deaminated 100 and 90%, respectively, by MAO-A [3]. A similar curve for norepinephrine was obtained with rat liver mitochondria [4], but discrepant results were obtained in another study which demonstrated that only 50% of *l*-norepinephrine deamination in human brain was sensitive to low (10^{-8} to 10^{-6} M) clorgyline concentrations [13]. Numerous investigations have demonstrated that serotonin acts generally as a highly preferential substrate for MAO-A, and that it is only weakly deaminated by MAO-B in human platelets (which contain only this form of the enzyme) and other tissues in a few species when high substrate concentrations are used [11, 14, 15]. In the present study, we have measured the deamination of *l*-norepinephrine and compared it to that of serotonin and of the MAO-B selective substrate, β-phenylethylamine, in several MAO-A rich tissues (superior cervical ganglia, placenta and the N1E-115 neuroblastoma cell line), an MAO-B rich tissue (the human platelet), and in brain tissues from several species containing differing proportions of both MAO-A and MAO-B.

MATERIALS AND METHODS

Tissue preparation. Sprague–Dawley male rats (Taconic Farms, Germantown, NY), Swiss Webster male mice (NIH), female golden hamsters, *Mesocricetus auratus* (Lakeview Hamster Colony, Newfield, NJ), and male dwarf hamsters, *Phodopus sungorus* (NIH), were decapitated, and the brains and peripheral tissues were removed rapidly. Adult male and female St. Kitts vervets, *Cercopithecus aethiops sabeus*, and male rhesus monkeys, *Macaca mulatta*,

* Address all correspondence to: Nancy A. Garrick, Clinical Neuropharmacology Branch, National Institute of Mental Health, NIH Clinical Center, 10/3D41, Bethesda, MD 20205, U.S.A.

were killed as part of other studies and dissected as described previously [16]. Human brains were obtained at autopsy from adult males within 24 hr of death. Deaths in the individuals chosen for this study were primarily the result of cardiovascular disorders without direct cerebrovascular involvement, although the death of one individual (whose MAO activities were comparable to the others) was complicated by pneumonia and sepsis. Human placenta was obtained after normal delivery and drained of blood; trophoblast tissue was dissected free from the placental vessels and washed with 0.9% NaCl solution. Mouse neuroblastoma N1E-115 cells and human platelets were prepared as described previously [14, 17].

All tissues were stored at −80° until homogenization in 10% (w/v) 0.08 M phosphate buffer (Na_2HPO_4-KH_2PO_4), pH 7.2. The homogenate was centrifuged at 900 *g* for 10 min at 4°; the supernatant fraction was carefully removed and stored at −80° until assayed. In the case of human placenta, the supernatant fraction was centrifuged at 16,000 *g* for 30 min to obtain a crude mitochondrial pellet. Tissues were sonicated on ice for 15 sec (Sonifer Cell Disruptor, model W 140, Heat Systems-Ultrasonics, Inc., Plainview, Long Island, NY) prior to each experiment to disrupt large membraneous fragments and to prepare an even suspension of enzyme. Prior studies using rat brain homogenates and human platelet homogenates demonstrated no effects of brief sonication on the deamination of either an MAO-A substrate (serotonin) or MAO-B substrates (phenylethylamine and benzylamine) [C. H. Donnelly, Ph.D. Thesis, University of Maryland (1977)].

MAO assay. Monoamine oxidase activity was determined by a modification of the method used by Robinson *et al.* [18]. Sonicated enzyme preparations were assayed in triplicate by incubating 50 μl aliquots in 500 μl of 0.08 M phosphate buffer (Na_2HPO_4-KH_2PO_4), pH 7.2, containing 10^{-4} M EDTA and 10^{-4} M ascorbate with 25 μl of radioactively labeled substrate for 20 min with shaking at 37°. Blank values, a measure of non-enzymatic degradation of substrate, were established by incubating enzyme samples with 10^{-3} M clorgyline (May & Baker Ltd., Essex, England) and 10^{-3} M pargyline (Abbott Laboratories, North Chicago, IL) to inhibit completely MAO-A and MAO-B and subtracting this value from that of the active enzyme preparation. Blank values obtained in this manner were similar to heat-inactivated samples. The reaction was stopped by immersion in an ice-water bath and cooling for 5 min. Samples were transferred to Pasteur pipettes containing 0.5 × 2.5 cm Amberlite resin (CG-50, 100–200 mesh, Mallinckrodt Chemical Works, St. Louis, MI). The columns were washed twice with 1.0 ml of distilled water and the entire 2.5 ml was collected in glass vials containing 17.5 ml Aquasol (New England Nuclear Corp., Boston, MA). The radioactive products of the reaction were counted by liquid scintillation spectrometry.

In some studies, inhibition curves were established by preincubating the enzyme preparations with clorgyline in final concentrations of 10^{-3} to 10^{-11} M for 30 min at 25° prior to the addition of radioactive substrate. At the chosen substrate concentrations, the percent inhibition at the plateau regions from 10^{-7} M to 10^{-8} M clorgyline was used to establish the relative proportions of MAO-A versus MAO-B activity, on the basis of data from many other studies indicating the validity of this approach [8, 17, 19]. Apparent K_m and V_{max} values were calculated by linear regression analysis from plots obtained from Lineweaver–Burk and Hanes (s vs s/v) transformations of the Michaelis–Menten equation; substrates were varied over a 10-fold concentration range which in final determinations approximated 0.5 to 5 times the K_m. In some studies to evaluate the separate K_m values for MAO-A and MAO-B in human and rat cortex, the tissue homogenates were preincubated for 30 min at 25° with 10^{-7} M clorgyline or 10^{-6} M deprenyl to inactivate one or the other form of the enzyme. These inhibitor concentrations were chosen on the basis of previous data [8, 13, 14, 16] and the present results establishing these concentrations as yielding mid-plateau points on inhibition curves for these tissues.

[^{14}C]β-Phenylethylamine was purchased from the New England Nuclear Corp.; [^{14}C]serotonin and [^{14}C]*l*-norepinephrine were purchased from the Amersham/Searle Corp., Arlington Heights, IL. Non-radiolabeled amines (Calbiochem, San Diego, CA) were added when required to obtain the desired final concentrations. Protein values were 0.01 to 0.02 mg/aliquot when β-phenylethylamine was used as substrate and 0.2 to 2.8 mg/aliquot for all other substrates. These enzyme concentrations were chosen to ensure that substrate deamination was less than 10% over the reaction period. Monoamine oxidase activity was linear with respect to protein concentration and time of incubation. Protein was determined by a modification of the method of Lowry *et al.* [20], using bovine serum albumin as standard. It should be noted that MAO activity determined with both an MAO-A substrate (serotonin) and an MAO-B substrate (phenylethylamine) is stable for up to 8 hr at 4° and for up to 4–8 weeks at −20° and up to 6 months at −80°, in keeping with other data on the general stability of MAO during freezer storage [14, 16, 17, 21]. To evaluate the effect of storage on *l*-norepinephreine deamination, we examined tissue homogenates prior to and after 6 months of storage at −80° and found essentially identical enzyme specific activities (expressed as nmoles per mg protein per hr) for rat cortex (27.5 ± 2.4 vs 28.4 ± 2.8, N = 5) and human cortex (13.9 ± 1.2 vs 15.6 ± 0.76, N = 9).

RESULTS

In both rat and human brain, *l*-norepinephrine was deaminated less avidly by MAO than was serotonin. As indicated in Table 1, the K_m values for *l*-norepinephrine were 2- to 4-fold greater than those for serotonin. The V_{max} values were 4-fold smaller for *l*-norepinephrine (34 and 16 nmoles per mg protein per hr) than the corresponding values for serotonin (129 and 63 nmoles per mg protein per hr) in rat and human cortex respectively. In the MAO-A rich human placenta, a similar 3-fold K_m difference was found for *l*-norepinephrine compared to serotonin, while in the MAO-B-containing human

Table 1. Michaelis constants for *l*-norepinephrine and serotonin deamination by MAO from rat and human cerebral cortex, human placenta and human platelet*

Tissue	Apparent K_m ($\times 10^{-4}$ M) *l*-Norepinephrine	Serotonin
Rat cerebral cortex	4.15 ± 0.25	0.99 ± 0.14
Human cerebral cortex	2.61 ± 0.06	1.11 ± 0.09
Human placenta	4.16 ± 0.65	1.62 ± 0.82
Human platelet	3.89 ± 0.92	>10.00

* Values are means ± S.E.M.

platelet a smaller K_m was found for *l*-norepinephrine than for serotonin (Table 1). When the K_m values for norepinephrine deamination in rat cortex were examined in the presence of 10^{-6} M deprenyl or 10^{-7} M clorgyline, generally similar values for MAO-A (3.24×10^{-4} M) and MAO-B (2.96×10^{-4} M) were observed; comparable results were also found in human cortex following deprenyl (4.80×10^{-4} M) for MAO-A, and following clorgyline (3.96×10^{-4} M) for MAO-B.

Across the various rodent tissues examined, specific activities for *l*-norepinephrine were only 19–27% of those for serotonin when substrate concentrations approximating five times their respective K_m values were used (Table 2). Lower specific activities for *l*-norepinephrine as compared to serotonin (24–47%) were also observed in most human and nonhuman primate tissues (Table 2). It should be noted that even noradrenergic tissues such as rat superior cervical ganglia and mouse neuroblastoma cells possessed low specific activities for *l*-norepinephrine relative to serotonin, as did brain and other tissues. In contrast, *l*-norepinephrine deamination was greater (200%) relative to serotonin deamination in the human platelet, which contains MAO-B exclusively [14].

In the studies using the selective MAO-A inhibitor, clorgyline, a biphasic inhibition plot was observed in rat cortex for *l*-norepinephrine, while serotonin inhibition followed a simple sigmoidal curve (Fig. 1A). Following the convention established by Johnston [1] and verified by the use of selective substrate ratios, and by other studies with selective inhibitors [8], approximately 85% of the deamination of *l*-norepinephrine in rat cortex could be attributed to MAO-A, in contrast to 100% of serotonin deamination. Similar clorgyline inhibition plots suggest that *l*-norepinephrine was deaminated by MAO-A to an even lesser extent in human cortex (60%) and vervet cortex (25–30%), while serotonin was deaminated 100% by MAO-A in human cortex and 90% in vervet cortex (Fig. 1, panels B and C). In contrast to serotonin and *l*-norepinephrine, β-phenylethylamine deamination was insensitive to low concentrations of clorgyline, indicating that low concentrations of this substrate are metabolized selectively by MAO-B in the species studied. In the human platelet, neither *l*-norepinephrine nor serotonin deamination was inhibited whatsoever by 10^{-7} M clorgyline; a simple sigmoidal curve for *l*-norepinephrine was obtained, with 50% inhibition found at quite high (3×10^{-5} M) clorgyline concentrations.

Similar inhibition studies using clorgyline in brain

Table 2. *l*-Norepinephrine, serotonin and β-phenylethylamine deamination by monoamine oxidase in rodent and primate tissues*

Tissue	Specific activity (nmoles/mg protein/hr) *l*-Norepinephrine	Serotonin	β-Phenylethylamine	*l*-Norepinephrine as percent of serotonin deamination
Rodent				
Rat whole brain	42.8	158.7	30.0	27
Rat cortex	27.5	135.5	32.3	20
Rat superior cervical ganglia	120.6	645.0	28.6	19
Hamster whole brain	10.8	54.7	2.2	20
Mouse whole brain	13.5	67.2	20.8	20
Mouse neuroblastoma N1E-115	10.6	45.0	0.4	24
Primate				
Vervet cortex	21.1	89.0	42.9	24
Rhesus cortex	23.2	66.4	70.4	35
Human cortex	13.9	32.5	35.5	43
Human platelet	7.8	3.9	12.0	200
Human placenta	372.8	799.2	32.5	47

* Final concentrations of substrates used were: [^{14}C]*l*-norepinephrine, 1.7×10^{-3} M; [^{14}C]serotonin, 6.5×10^{-4} M; and [^{14}C]β-phenylethylamine, 2×10^{-5} M.

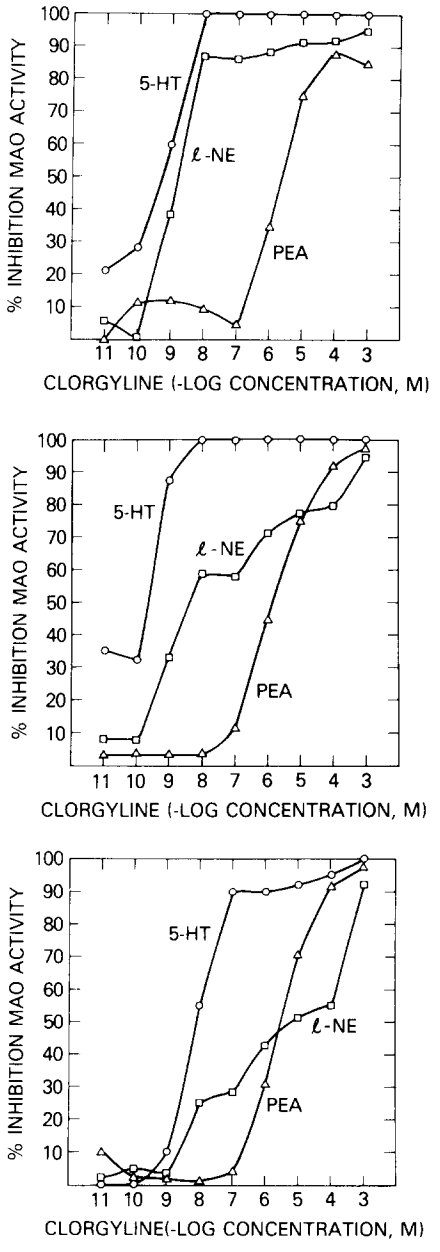

Fig. 1. Clorgyline inhibition of MAO activity, using 10^{-3} M serotonin (○), 10^{-3} M *l*-norepinephrine (□) and 2×10^{-5} M β-phenylethylamine (△) as substrates in (A) rat cortex, (B) human cortex and (C) vervet cortex. Curves represent the mean of three determinations for each substrate.

tissue from additional species, as summarized in Table 3, also indicate that serotonin was deaminated 80–100% by MAO-A in all species examined, while *l*-norepinephrine was not nearly as selective a substrate for MAO-A, being deaminated highly variably (30–85%) by MAO-A in the different tissues.

DISCUSSION

The simple sigmoidal curve obtained by Johnston [1] for the inhibition of rat brain serotonin deamination by clorgyline was the first conclusive evidence establishing the existence of the substrate-selective monoamine oxidase type A. Simple sigmoidal curves were subsequently reported for the inhibition of serotonin and norepinephrine deamination in rat brain and rat superior cervical ganglia [2, 3]. On the basis of similar data obtained in an outer mitochondrial membrane preparation from rat liver, in which simple sigmoidal inhibition curves were obtained with clorgyline, norepinephrine (and epinephrine) was classified as an exclusive substrate for MAO-A [4]. In contrast, our data from rat and hamster brain, and from three primate species (human, vervet and rhesus), all indicate that *l*-norepinephrine deamination manifests biphasic inhibition curves with clorgyline, with 30–85% of *l*-norepinephrine deamination being sensitive to 10^{-7} M clorgyline. One previous study reported that norepinephrine deamination in human brain and liver was inhibited only 50 and 65%, respectively, by 10^{-8} M to 10^{-7} M clorgyline, but complete inhibition curves were not published [13].

It should be noted that data generated from inhibition curves regarding the percent metabolism of various substrates by MAO-A compared to MAO-B in different tissues represent relative values which depend to a varying extent upon the concentration of the substrate used. Phenylethylamine deamination, for example, has been reported to be carried out in rat brain almost exclusively by MAO-B at concentrations (10 μM) near its K_m for MAO-B, while at much higher phenylethylamine concentrations (100 μM to 10 mM), nearer its K_m for MAO-A, a significant proportion of its deamination can be accomplished by the MAO-A subtype [22, 23]. When we varied norepinephrine concentrations from its approximate K_m (3×10^{-4} M) to three times and ten times its K_m and did repeated clorgyline inhibition curves for rat brain, identical plateaus, with no points varying by more than 3% S.E.M., were obtained with all three substrate concentrations (unpublished data). Thus, for substrates like norepinephrine which, in rat and human brain, have very small K_m differences between MAO-A ($3–5 \times 10^{-4}$ M) and MAO-B ($3–4 \times 10^{-4}$ M) according to our results and a previous study [13], moderate differences in substrate concentrations should not appreciably affect estimations of percent deamination by MAO-A and MAO-B from clorgyline inhibition curves. Even when K_m values are divergent for the two enzyme forms, the careful selection of substrate concentrations near the K_m for the selective enzyme form should also yield reproducible conclusions, as has been demonstrated for phenylethylamine [22, 23] and recently for serotonin [24].

These investigations in brain tissue from several species thus have demonstrated that *l*-norepinephrine is only somewhat selectively deaminated by monoamine oxidase type A. Although *l*-norepinephrine has been widely discussed as a selective MAO-A substrate, this appears not to be the case for human and other primate brain tissues where a substantial proportion of *l*-norepinephrine deamination *in vitro* is accomplished by MAO-B. The human platelet, which contains MAO-B activity only, was also the single tissue among all of those

Table 3. Percent deamination of *l*-norepinephrine and serotonin by monoamine oxidase type A in primate and rodent brain*

	% Inhibition of substrate deamination by 10^{-7} M clorgyline	
	l-Norepinephrine	Serotonin
Primate		
Human	58 ± 5	100 ± 0
Rhesus (*M. mulatta*)	37 ± 11	80 ± 6
Vervet (*C. aethiops*)	30 ± 9	90 ± 4
Rodent		
Rat	85 ± 5	100 ± 0
Hamster		
(*P. sungorus*)	75 ± 5	95 ± 2
(*M. auratus*)	80 ± 7	92 ± 3

* Percent deamination by MAO type A was determined by the inhibition of *l*-norepinephrine (10^{-3} M) and serotonin (10^{-3} M) deamination by 10^{-7} M clorgyline. The means ± S.E.M. derived from three inhibition curves obtained using 10^{-11} to 10^{-3} M clorgyline are presented.

examined which deaminated norepinephrine more avidly than serotonin; the specific activity for norepinephrine deamination in the platelet was nearly as high (7.8 nmoles per mg protein per hr) as that for the preferential MAO-B substrate phenylethylamine (12 nmoles per mg protein per hr).

Serotonin, in contrast, is either exclusively, or predominantly, deaminated by MAO-A under most assay conditions and appears to be the amine which is most selectively deaminated by MAO-A among those investigated. Only under exceptional circumstances, e.g. where high concentrations of this amine (10^{-2} M) are used [15], or in a few tissues such as bovine brain and liver, is serotonin deaminated to a significant extent by MAO-B; in these tissues serotonin is a very poor substrate for deamination in comparison to other amines [11]. It is of interest that another indoleamine closely related in structure to serotonin, tryptamine, has the lowest K_m for MAO-A of all substrates studied [13].

Serotonin was found to have a higher apparent affinity for MAO than norepinephrine, as reflected in the 2- to 4-fold lower apparent K_m values found for serotonin compared to *l*-norepinephrine in human and rat brain; a 3-fold higher affinity for serotonin was also found in the MAO-A rich human placenta. These K_m values are similar to those for serotonin and norepinephrine previously reported in separate studies of rat brain homogenates (0.6 and 3.2×10^{-4} M respectively) [25] and human cortex mitochondria (1.0 and 4.4×10^{-4} M respectively) [26]. Other evidence suggesting a higher affinity of serotonin for the MAO-A site has come from data indicating that 16-fold lower concentrations of serotonin ($K_i = 40\ \mu M$) than norepinephrine ($K_i = 640\ \mu M$) inhibited the binding of a reversible selective MAO-A inhibitor, radiolabeled harmaline [27]. Additionally, it is of note in regard to the relatively greater specificity of serotonin compared to norepinephrine for the MAO-A site that both our data and an earlier study [13] indicate that norepinephrine has nearly equivalent apparent K_m values for the MAO-A and MAO-B sites, while several other investigations of brain and also of the essentially pure MAO-A in mouse neuroblastoma cells and the MAO-B in human platelets suggest that the K_m values for serotonin for MAO-A are 10- to 40-fold smaller than those for the MAO-B enzyme [14, 17, 28].

Earlier studies indicating that norepinephrine was a selective substrate for MAO-A led to the suggestion that there existed "transmitter-specific" MAOs. This interpretation was supported by evidence from the destruction of sympathetic neurons surgically, chemically or immunologically, which led to a selective loss of clorgyline-sensitive MAO-A activity in the rat pineal gland, vas deferens, submaxillary gland, mesenteric artery and other tissues containing both MAO-A and MAO-B activities [3, 29–32]. Tissues with dense noradrenergic fibers such as the spleen and the adventitial component of blood vessels contain mostly MAO-A activity [33], as does the brain area richest in noradrenergic cell bodies, the locus coeruleus [34], and also neuroblastoma cells grown in tissue culture [17]. However, it now more clearly would seem that MAO-A is only relatively transmitter specific, as the kinetic data indicate that it deaminates *l*-norepinephrine less readily than serotonin and tryptamine. Norepinephrine is also clearly less of an exclusive substrate for MAO-A than is serotonin, particularly in primate brain where 40–70% of norepinephrine deamination *in vitro* may be accomplished by MAO-B versus only 0–20% for serotonin. Whether equivalent differences occur under *in vivo* conditions needs more direct investigation. In one study, low doses of clorgyline led to greater acute elevations of serotonin than norepinephrine in rat brain [35]. However, greater reductions in a norepinephrine metabolite, 3-methoxy-4-hydroxyphenylglycol, than in a serotonin metabolite, 5-hydroxyindoleacetic acid, were found in cerebrospinal fluid after chronic clorgyline administration in man [36]. Nonetheless, the present studies raise the possibility that when physiological or behavioral

consequences follow selective MAO-A but not MAO-B inhibitor administration (e.g. antidepressant effects [37]), the alterations may reflect serotonin rather than norepinephrine metabolic changes.

Acknowledgements—The authors thank Drs. D. E. Redmond, Jr., L. F. Major, E. Richelson, J. Hawley, W. and N. Mayer and J. Crawley for their assistance in obtaining some of the tissues from the various species, Dr. Herbert Levitan for helpful discussion, and Mrs. I. Bellesky for typing the manuscript.

REFERENCES

1. J. P. Johnston, *Biochem. Pharmac.* **17**, 1285 (1968).
2. C. Goridis and N. H. Neff, *Br. J. Pharmac.* **43**, 814 (1971).
3. C. Goridis and N. H. Neff, *Neuropharmacology* **10**, 557 (1971).
4. M. D. Houslay and K. F. Tipton, *Biochem. J.* **139**, 645 (1974).
5. C. Braestrup, H. Andersen and A. Randrup, *Eur. J. Pharmac.* **38**, 181 (1975).
6. V. Glover, M. Sandler, F. Owen and G. J. Riley, *Nature, Lond.* **265**, 80 (1977).
7. K. T. Demarest, D. J. Smith and A. J. Azzaro, *J. Pharmac. exp. Ther.* **215**, 461 (1980).
8. N. A. Garrick and D. L. Murphy, *Psychopharmacology* **72**, 27 (1980).
9. R. F. Squires, *Adv. Biochem. Psychopharmac.* **5**, 355 (1972).
10. H-Y. T. Yang and N. H. Neff, *J. Pharmac. exp. Ther.* **187**, 365 (1973).
11. C. J. Fowler and B. A. Callingham, *Biochem. Pharmac.* **27**, 97 (1978).
12. L. B. Hough and E. F. Domino, *J. Pharmac. exp. Ther.* **208**, 422 (1979).
13. H. L. White and A. T. Glassman, *J. Neurochem.* **29**, 987 (1977).
14. C. H. Donnelly and D. L. Murphy, *Biochem. Pharmac.* **26**, 853 (1977).
15. C. Mitra and S. R. Guha, *Biochem. Pharmac.* **29**, 1213 (1980).
16. D. L. Murphy, D. E. Redmond, Jr., N. Garrick and J. Baulu, *Neurochem. Res.* **4**, 53 (1979).
17. C. H. Donnelly, E. Richelson and D. L. Murphy, *Biochem. Pharmac.* **25**, 1639 (1976).
18. D. S. Robinson, W. Lovenberg, H. Keiser and J. Sjoerdsma, *Biochem. Pharmac.* **17**, 109 (1968).
19. M. D. Houslay, K. F. Tipton and M. B. H. Youdim, *Life Sci.* **19**, 467 (1976).
20. O. H. Lowry, N. J. Rosebrough, A. L. Farr and R. J. Randall, *J. biol. Chem.* **193**, 265 (1951).
21. A. V. P. Mackay, P. Davies, A. J. Dewar and C. M. Yates, *J. Neurochem.* **30**, 827 (1978).
22. H. Kinemuchi, Y. Wakui and K. Kamijo, *J. Neurochem.* **35**, 109 (1980).
23. O. Suzuki, H. Hattori, M. Oya, Y. Katsumata and T. Matsumoto, *Life Sci.* **25**, 1843 (1979).
24. C. J. Fowler and K. F. Tipton, *J. Neurochem.* **38**, 733 (1982).
25. Z. Huszti, *Molec. Pharmac.* **8**, 385 (1972).
26. H. L. White and J. C. Wu, *J. Neurochem.* **25**, 21 (1975).
27. D. L. Nelson, A. Herbert, Y. Petillot, L. Pichot, J. Glowinski and M. Hamon, *J. Neurochem.* **32**, 1817 (1979).
28. B. Ekstedt, *Biochem. Pharmac.* **25**, 1133 (1976).
29. B. Jarrott, *J. Neurochem.* **18**, 7 (1971).
30. H-Y. T. Yang, C. Goridis and N. H. Neff, *J. Neurochem.* **19**, 1241 (1972).
31. J. F. Coquil, C. Goridis, G. Mack and N. H. Neff, *Br. J. Pharmac.* **48**, 590 (1973).
32. R. E. Schmidt, C. D. Ross and D. B. McDougal, Jr., *J. Neurochem.* **30**, 537 (1978).
33. J. A. Levin and S. E. Wilson, *J. Pharmac. exp. Ther.* **203**, 598 (1977).
34. J. M. Saavedra, M. J. Brownstein and M. Palkovits, *Brain Res.* **118**, 152 (1976).
35. H-Y. T. Yang and N. H. Neff, *J. Pharmac. exp. Ther.* **189**, 733 (1974).
36. L. F. Major, D. L. Murphy, S. Lipper and E. Gordon, *J. Neurochem.* **32**, 229 (1979).
37. S. Lipper, D. L. Murphy, S. Slater and M. S. Buchsbaum, *Psychopharmacology* **62**, 123 (1979).