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Biochemical, behavioral, physiologic, and neurodevelopmental changes in mice deficient in monoamine oxidase A or B

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Abstract

The availability of mutant mice that lack either MAO A or MAO B has created unique profiles in the central and peripheral availability of serotonin, norepinephrine, dopamine, and phenylethylamine. This paper summarizes some of the current known phenotypic findings in MAO A knock-out mice and contrast these with those of MAO B knock-out mice. Differences are discussed in relation to the biochemical, behavioral, and physiologic changes investigated to date, as well as the role played by redundancy mechanisms, adaptational responses, and alterations in neurodevelopment.

Keywords

Monoamine oxidase; Serotonin; Norepinephrine; Phenylethylamine; Dopamine

INTRODUCTION

Monoamine oxidase A (MAO A) and monoamine oxidase B (MAO B) are isoenzymes whose primary roles are in the metabolism of amines, and in the regulation of neurotransmitter levels and intracellular amine stores. The isoenzymes are located in the outer membrane of the mitochondria and demonstrate overlapping substrate specificities and tissue distributions. MAO A preferentially metabolizes serotonin (5-HT) and norepinephrine (NE), whereas MAO B has a higher affinity for phenylethylamine (PEA). Dopamine (DA)

in the mouse under normal physiological conditions is largely metabolized by MAO A, though at higher concentration a contribution by MAO B also becomes significant [22]. The availability of mutant mice that lack either MAO A [12] or MAO B [27] has created unique profiles in the central and peripheral availability of 5-HT, NE, DA, and PEA. Such targeted disruptions of single genes are unique in that they promise to elucidate the precise role of these bioamines during ontogeny and adult life in a manner which is relatively precise and free of the side effects encountered with many pharmacological probes. Though the deletion of a gene may be a relatively “clean” process, interpretation of the resulting phenotype is not necessarily straightforward. In certain instances, the knocking out of a single gene results in no obvious phenotypic change. This is the case, in particular, if there exist gene redundancy mechanisms whose presence may prevent abnormal phenotypes from becoming unmasked. Adaptational responses can also be activated in these animals, which may mask the expression of the resulting phenotypes. Furthermore, because the enzymatic loss in these animals exists from the earliest stages of embryogenesis throughout development, phenotypes examined postnatally are shaped by significant alterations in the normal pattern of ontogeny.

This paper summarizes some of the current known phenotypic findings in MAO A-deficient mice and contrasts these with those of MAO B-deficient mice (Tables 1 and 2). Differences are discussed in relation to the biochemical, behavioral, and physiologic changes investigated to date, as well as the role played by redundancy mechanisms, adaptational responses, and alterations in neurodevelopment.

GENERATION OF MAO A AND MAO B DEFICIENT MICE

MAO A and MAO B share 70% similarity in amino acid sequence [2], and are encoded by separate genes that are closely linked on the X-chromosome [42]. The genes have identical intron-exon organization, and likely are derived from the same ancestral gene [26]. Transgenic mice lacking MAO A were generated through the integration of an interferon β transgene into the gene encoding MAO A [12]. The interferon β gene was injected into embryos of C3H/HeJ mice and was integrated between exons 1 and 4 of the MAO A gene, resulting in a resultant truncated polypeptide devoid of MAO A catalytic activity. The transgene was transmitted by X-linked inheritance. To rule out the possibility that interferon β affected the phenotypic features of the transgenic animals, F2 progeny of transgenic females were mated to knockout males lacking the interferon-beta receptor [55]. F2 and F3 pups having the interferon β transgene displayed the same abnormal behaviors described below, independent of the presence or absence of the interferon β receptor [12].

Transgenic mice lacking MAO B were generated by insertion of a transcriptionally active neomycin resistance gene into exon 6 of the MAO B gene [27]. Once the targeting construct had recombined with the MAO B allele, it generated a stop codon that resulted in a truncated, inactive MAO B enzyme. The mutated cDNA was microinjected into C57 BL/6 mouse blastocytes, which gave rise to male chimeric mice. Mating of chimeric males with 129/Sv females demonstrated an X-linked transmission.

PHENOTYPE OF MAO A-DEFICIENT MICE

Biochemical Changes

MAO A knock-out mice (MAO A KO) display an absence of MAO A enzymatic activity in every organ examined including the brain, liver, lung, kidney, testes, spleen, heart, and harderian gland ([12,32]; unpublished observation). There is no indication of a significant compensatory up-regulation of MAO B in these tissues. Mutant mice compared to their wild-type counterparts demonstrate an increase in the amount of 5-HT, NE, and DA in brain. Serotonin levels are considerably increased in MAO A KO pups (ninefold at day 1, sixfold at day 12), whereas levels of the 5-HT metabolite 5-hydroxy-indoleacetic acid (5-HIAA) are markedly decreased in brain (–85% below control levels at day 1, –75% at day 12). Interestingly, 5-HT increases become less marked over time, decreasing to 92% to 54% above control levels for mice ages 1–3 months, and to ~10% above control levels for mice ages 7 months. Likewise, decreases in 5-HIAA levels noted in MAO A KO neonates diminish over time and appear at ~35% and ~20% below normal levels for mice ages 3 and 7 months, respectively. Norepinephrine levels in brain are maximal for adult MAO A KO mice ages 1–3 months (100% increase over wild-type), with smaller increases noted in neonates and 7-month-old animals. The elevation in DA is slight, although the DA metabolite dihydroxy-phenylacetic acid (DOPAC) is markedly decreased (3.5 times less in 3-month-old adults).

These findings highlight the following: (1) Amine increases in MAO A KO mice are highest for those substrates with greatest specificity for MAO A (5-HT, NE), and less for substrates which demonstrate a substantial shared substrate specificity for MAO A and B (DA). Increases are higher for 5-HT than NE or DA, possibly the result of the fact that 5-HT, unlike NE and DA, cannot be metabolized by alternate metabolic routes such as catechol-o-methyl transferase (COMT). (2) There is no compensatory increase in MAO B, suggesting that the presence of alternate metabolic pathways such as MAO B, COMT, and other amine oxidases is sufficient to maintain DA at near physiologic levels. (3) The increases of 5-HT and NE in MAO A KO mice compared to normal mice are age-dependent in the postnatal period. Differences are likely influenced by the natural developmental delay in the maturation of MAO B enzymatic activity seen in the neonate and young adult [38,78], as well as by the increasing importance of adaptational mechanisms which compensate for the underlying absence of MAO A activity. Such mechanisms could include decreases in substrate synthesis and/or enhancement of alternate metabolic routes. (4) Presence of 5-HIAA in MAO A KO mice suggest that MAO B can play a role in 5-HT oxidation *in vivo*, unlike *in vitro* results, which suggest little to no role. This could be due to the relative low sensitivity of the *in vitro* assays.

In association with the increase in 5-HT levels in brain, MAO A deficient mice also show a modest downregulation of post-receptors, and the vesicular synaptic 5-HT_{1A}, 5-HT_{2A}, 5-HT_{2C} monoamine transporter (VMAT2) [6,74]. Quantitative autoradiography shows decreased binding of the 5-HT_{1A} receptors in the hippocampus (–16%) and dentate gyrus (–16%) of adult animals, and in the medulla and cervical cord (>–52%) of pups. Low expression of 5-HT_{2A} receptors in adult animals are seen in layer 4 of the frontal cortex

(−31%), the 5-HT_{2C} receptors in the choroid plexus (−37%), and the VMAT2 in the striatum (−10%), dorsal raphe nucleus (−12%), and locus coeruleus (−7%). These findings highlight the occurrence of adaptational changes that may occur downstream from the functions of the disrupted gene.

Neurodevelopmental Changes

Since genetic mutations in conventional transgenic and knockout mice exist from the earliest stages of embryogenesis throughout development, phenotypes examined in adulthood reflect their altered ontogenetic histories. In MAO A KO mice excess 5-HT disrupts the segregation of somatosensory thalamocortical afferents [13,46,82]. This results in a complete absence in the cortex of the barrelfield formation, with a resultant alteration of the spatio-temporal processing of whisker information [87]. Similar alterations are caused by pharmacological inhibition of MAO A in developing wild-type animals [82]. In addition, during development mutant mice demonstrate transient and atypical 5-HT immunolabeling in a number of catecholaminergic structures in the substantia nigra, ventral tegmental area, hypothalamus, and locus ceruleus [11]. This phenomenon is reproduced in wild-type mice when degradation of 5-HT is prevented by MAO A inhibitors [46,82]. Serotonin immunolabeling in nonaminergic neurons in the telencephalon (CA1 and CA3 fields of the hippocampus, the central amygdala, the indusium griseum, the deep layers of the anterior cingulate and retrosplenial cortices), the diencephalon (primary sensory nuclei, as well as the mediodorsal, oval paracentral, submedial thalamic nuclei) are attributable to the existence of functional transport of 5-HT by the serotonin plasma membrane transporter (SERT), which is transiently expressed in nonmono-aminergic neurons [11]. Indeed, specific inhibitors of SERT abolish the 5-HT immunolabelling in these nonaminergic pathways. In catecholaminergic neurons, however, increased 5-HT immunolabelling occurs by alternate mechanisms. Using pharmacological experiments and genetic crosses with mice lacking the DA plasma membrane transporter, abnormalities in 5-HT uptake in catecholaminergic neurons can be entirely accounted for by the plasma membrane transporters for DA and NE [11].

In addition, MAO A deficiency alters the segregation of retinal afferents in the dorsal lateral geniculate nucleus and superior colliculus [80]. A primary role of 5-HT in this effect is suggested by the fact that such developmental abnormalities can be reversed by inhibiting 5-HT synthesis. Normalization of the faulty retinal projections, as well as the thalamocortical projections, is seen in mice doubly deficient for MAO A and the 5-HT_{1B} receptor gene [69]. This suggests that the 5-HT_{1B} receptor is a key factor in the abnormal segregation of sensory projections in MAOA KO mice. Abnormalities of neuronal retinal projections, however, have been made also in developing rats in response to subcutaneous injections of NE during the first 4 days of postnatal life [43]. Hence, in MAO A mutant mice a contributory role of the elevated brain levels of NE cannot be fully ruled out.

An excess of 5-HT in neonatal MAO A deficient mice appears to also give rise to abnormalities in morphology and activity of the phrenic motoneurons [6]. Experiments conducted *in vitro* on the brainstem-spinal cord preparation of neonatal mice reveals that at birth, the respiratory network of mutant animals is unable to generate a respiratory pattern as

stable as that produced by the wild-type network. The modulation by 5-HT of the network activity present in wild-type pups is lacking in the MAO A deficient pups. In addition, the morphology of the phrenic motoneuron is altered in mutant neonates, with the motoneuron dendritic tree losing its bipolar aspect and exhibiting an increased number of spines and varicosities. It seems likely that the maturational abnormalities observed here originate mainly from the 5-HT excess, because prenatal treatments inhibiting 5-HT synthesis or treatment with a 5-HT_{2A} receptor antagonist prevents these abnormalities [6].

That 5-HT may play a role in the maturation of locomotor networks, has been suggested by the transient difficulties in swimming abilities observed at birth in rats prenatally treated with the serotonin synthesis inhibitor para-chlorophenylalanine [58]. Consistent with these earlier results, recent work in MAO A KO mice suggests that these animals compared to their wild-type counterparts demonstrate a transient delay in the maturation of locomotor networks governing the role of the hindlimbs in swimming behavior [14]. A delay in the development of normal motor tone and motor function is further suggested by the fact that neonates between days 5 and 10 demonstrate these abnormalities: (i) prolonged righting, (ii) trembling upon locomotion and suspension by the tail, and (iii) moving backward instead of pivoting when placed on a new surface. These behaviors can be reproduced in wild-type pups, but with less intensity, by daily injection of the MAO A inhibitor clorgyline [12]. The exact mechanism by which perinatal excesses of 5-HT, and possibly NE, elicit locomotor deficits remains to be determined, since alterations may have been induced at several levels, including (1) the maturation of the locomotor network itself, (2) the maturation of the descending central projections, and/or (3) the expression of spinal 5-HT receptors. That aspects of hindlimb dysfunction may extend into adult life is suggested by the observation that adult MAO A KO mice display abnormal walking on a balance-beam, with adults grasping the lateral aspects of the beam with the hindlimbs during movement [12,69]. However, the possibility needs to be considered that such abnormalities may also be the result of altered somatosensory processing [13,29].

Behavioral Changes

Serotonin has been shown to influence a broad range of behavioral functions, including tonic and repetitive behavioral patterns, aggression, sexual behavior, learning, sensorimotor reactivity, circadian rhythm entrainment, sleep-wake cycles, and pain sensitivity. Many of the phenotypic aspects of MAO A KO mice are consistent with this general framework. MAO A deficient mice demonstrate altered courtship with increased grasping events in males and concurrent increased frequency and intensity of vocalization in females [12]. In addition, MAO A deficient mice demonstrate an increased susceptibility to developing the serotonin syndrome. In this syndrome, accentuation of serotonergic neuro-transmission has been implicated, possibly as a result of activation or modification of the 5-HT_{1A} receptor in the brainstem and spinal cord [21,25,34]. Aspects of this syndrome (restlessness, attentional deficits, disrupted social interaction) are elicited in MAO A deficient mice after inhibition of MAO B with either L-deprenyl or lazabemide, whereas this treatment does not overtly affect wild-type behavior [12]. Administration of fenfluramine which increases brain levels of 5-HT and NE through reuptake inhibition and presynaptic release [9,40,73,75] results in a more severe syndrome (tremor, rigidity, Straub tail, hindlimb abduction, lateral head

weaving, and reciprocal forepaw treading). Here the serotonin syndrome is elicited at estimated doses in MAO A KO mice that are half those required to elicit this syndrome in wild-type animals [29].

Most studies suggest that low brain 5-HT concentrations are associated with enhanced aggression since reduced concentrations of 5-HIAA in the cerebrospinal fluid have been correlated with aggressive behaviors in humans [17,49,53], and 5-HT receptor agonists reduce aggression [61]. Thus, it may appear paradoxical that male MAO A KO mice with their elevated levels of 5-HT demonstrate a marked increase in aggressive behavior. In the resident-intruder paradigm, a test of territorial aggression, male MAO A mutant mice demonstrate decreased latency to attack, prolonged attack duration, and decreased social interaction [12, 74]. Males housed in groups from the time of weaning show signs of offensive aggressive behavior, notably bite wounds that are most apparent from the age of 3 months [12]. These findings are consistent with studies demonstrating a correlation between elevated levels of 5-HT in the blood with aggressive behavior in humans [51,54,63,79]. Furthermore, they are consistent with reports of increased impulsive aggression reported in male human subjects suffering a point mutation in the MAO A gene [8].

Aggression of MAO A-deficient mice is markedly decreased by ketanserin and MDL 100907, both antagonists at the 5-HT_{2A} receptor, suggesting a role for this receptor in aggressive behavior [74]. By contrast, neither spiperone (a D₂/5-HT_{1A} antagonist), nor propranolol (a nonspecific β receptor antagonist) affect the behavior of mutant animals, suggesting that D₂, 5-HT_{1A}, and β -adrenergic receptors may not mediate the aggressive behavior of mutant mice. Despite its higher affinity for the 5-HT_{2A} receptor, MDL 100907 demonstrates a lower potency in decreasing aggression than ketanserin. The possibility needs to be considered that such differences might be due to ketanserin's additional antagonism of the vesicular monoamine transporter (VMAT2). This explanation is supported by the fact that tetrabenazine, also a VMAT2 antagonist, abolishes aggression in mutant and wild-type animals [74].

MAO A-deficient mice demonstrate a selective enhancement of emotional learning, as evidenced by an enhancement of classical fear conditioning (freezing to both tone and contextual stimuli) and an enhancement of step-down inhibitory avoidance learning [36]. In fear conditioning animals learn to associate a stimulus such as a generic tone with an aversive unconditioned stimulus such as a footshock. In step-down inhibitory avoidance, animals must remember that stepping off of a "safe" platform results in a foot-shock. The enhancement of fear learning in MAO A KO mice may be due to elevated catecholamine levels, because injections of catecholamines into the brains of mice enhances fear memory formation, whereas drugs that lower the levels of catecholamines impair it. Since the amygdala is critically involved in both fear conditioning and inhibitory avoidance learning, it is possible that the alteration of catecholamine levels in the amygdala is responsible for the enhancement of fear learning observed in the MAO A mutant mice [48,52]. In contrast, eyeblink conditioning is normal in the MAO A mutants, suggesting that motor learning remains unaffected [36]. The lack of effect on eyeblink conditioning is surprising given that monoaminergic afferents constitute one of the three major afferent systems in the cerebellum [33]. The monoaminergic inputs include well-defined noradrenergic and

serotonergic afferents from the locus coeruleus and raphe nucleus, respectively. It is possible that such a paradoxical finding may be the result of some form of developmental compensation.

MAO A KO mice show increased mobility in the Porsolt Forced Swim Test. The forced swimming test (FST), or “behavioral despair” test, is a behavioral test used to measure potential antidepressant activity [64,65]. The procedure consists of placing an animal in a cylinder of water from which it cannot escape. Failure to persist in escape-directed behavior in this paradigm is thought to represent the development of behavioral passivity in response to prolonged stress. Treatment with antidepressant drugs decreases the duration of immobility in the FST. Although the FST is sensitive to most antidepressants that increase NE transmission, the effects of serotonin-mimetic drugs on the classical FST have been mixed [4,5]. Thus, increased mobility of MAO A KO mice on the FST may be a result of either the increase of NE and/or 5-HT. The increased mobility of the MAO A mutant mice, however, was not due to increased basal levels of locomotor activity, as there was no evidence of hyperactivity in mice placed in a novel Open Field environment [12].

Physiological Changes

Serotonin has been shown to influence a broad range of physiologic functions. Stimulants of 5-HT reduce food intake and weight gain and increase energy expenditure, both in animals and in humans. MAO A mutant mice compared to wild-type mice demonstrate lower body weights (~10% lower), though it remains to be clarified whether this is related to alterations in serotonergic tone or if this simply reflects a slower weight gain due to behavioral difficulties and poor feeding in infancy. Consistent with the role of 5-HT in thermoregulation [45,50,86], fenfluramine elicits an exaggerated increase in core temperature in knock-out mice compared to wild-type mice [29]. Central serotonergic, as well as noradrenergic mechanisms have been implicated in the control of blood pressure and heart rate with most studies suggesting pressor and chronotropic effects of these biogenic amines. Hence, it is surprising that in the resting, restrained state MAO A KO mice compared to wild-type mice demonstrate a low to normal basal blood pressure and heart rate. These findings are consistent with anecdotal reports of altered peripheral autonomic function and resting hypotension in patients with Norrie disease, an X-linked recessive disorder which may encompass deletions in the gene for MAO-A [57,77]. It is possible in the face of the markedly enhanced levels of pressor amines that adaptational responses may be elicited in MAO A KO mice, which act to maintain a lowered blood pressure and heart rate. Though the nature of such compensatory changes remains to be clarified, there is suggestive evidence that such decreases may be the result of an increase in gain of the baroreflex, which under physiologic conditions acts to regulate heart rate and blood pressure [68]. Such a physiologic overcompensation, which resets the baseline autonomic tone of MAO A KO mice to a lower set point, may function to attenuate the physiologic consequences of acute, exaggerated metabolic shifts in the concentration of pressor amines.

Knock-out animals, compared to their wild-type counterparts, demonstrate a significantly higher regional cerebral cortical blood flow (CBF) in primary somatosensory and barrel field neocortex. Regional CBF is significantly lower in the entorhinal and midline secondary

motor cortex, with smaller decreases noted in the olfactory, piriform, visual, and retrosplenial cortices and the amygdala. Several possible explanations present themselves to account for such genotypic differences. Differences in regional cortical perfusion between wild-type and knock-out mice may reflect underlying differences in the regional concentrations of 5-HT and/or NE. Interestingly, the topography of serotonergic, cortical projection fibers from the dorsal raphe nucleus parallels some of the genotypic differences in regional cortical perfusion noted in these animals. Dorsal raphe neurons project densely to lateral cortical structures (piriform/entorhinal cortex), the amygdala and regions in the medial frontal cortex, with more sparing innervation of sensorimotor areas [81]. On the other hand, locus coeruleus neurons, the primary source of cortically projecting noradrenergic fibers, project broadly across the cortex, including the sensorimotor area [83,84]. Differences in perfusion patterns may also reflect neurodevelopmental abnormalities in the knock-out animals in whom excess 5-HT disrupts the segregation of somatosensory thalamocortical afferents (see above).

Administration of fenfluramine results in changes in regional cortical perfusion in most brain regions of both knock-out and wild-type mice that are opposite to the genotypic differences seen in perfusion in response to saline. Fenfluramine significantly increases regional CBF in the allocortex (olfactory, piriform, entorhinal) and the amygdala, and significantly decreases regional CBF in the somatosensory, barrel field, midline motor, auditory, visual, and retrosplenial cortices. Changes in regional perfusion in response to fenfluramine are topographically equivalent in knock-out and wild-type mice, although in knock-out mice such changes are of greater magnitude. These findings suggest that the effects on regional CBF of a life-long absence of MAO A, and the consequent chronic increase in 5-HT and NE, differ from those attributable to acute increases in these neurotransmitters following fenfluramine administration. Such a differential response may reflect neurodevelopmental abnormalities and/or effects of a chronic physiological adaptation on the regulation of cortical activation.

PHENOTYPE OF MAO B DEFICIENT MICE

Biochemical Changes

MAO B knock-out mice (MAO B KO) display an absence of MAO B enzymatic activity in every organ examined including the brain, liver, lung, kidney testes, spleen, heart, and hardierian gland ([27]; unpublished observation). Furthermore, there is no indication of a compensatory up-regulation of MAO A in these tissues. Brain levels of PEA, as well as urinary excretion of PEA, are increased approximately eightfold in MAO B-deficient animals [27]. Lack of MAO B does not alter extracellular levels of DA in striatum [22]. Similarly, the synthesis, storage, uptake, and release of DA are also unaltered [16]. There are no statistically significant differences in the concentrations of 5-HT, 5-HIAA, NE, DA, DOPAC, and homovanillic acid (HVA) in cerebral cortex, hippocampus, raphe nucleus, substantia nigra, thalamus, or striatum [16,27]. Furthermore, no differences in levels of 3-methoxytyramine (3-MT), 3-methoxy-4-hydroxyphenylethyleneglycol (MHPG) are found between mutant and control mice in various brain regions. The unchanged levels of these neurotransmitters and their metabolites, especially those of DA, in the MAO B KO mice are

surprising, because previous studies with specific inhibitors found that inhibition of MAO B *in vivo* results in increases in DA levels. Discrepancies here are likely due to the fact that *in vitro* near complete (> 90%) blockade of MAO B by selective inhibitors is accompanied by a significant inhibition (10% to 40%) also of MAO A [41]. Moreover, the prototype MAO B inhibitor L-deprenyl is transformed to amphetamine, which can also alter DA neurotransmission [35]. 3-methoxytyramine is produced by the metabolism of COMT. The normal levels of 3-MT in MAO B mutant mice suggest that COMT does not compensate for the loss of MAO B, and that MAO A alone is sufficient to metabolize DA.

The effect of loss of MAO B activity extends not only to alterations of its preferred substrate PEA, but also to changes in the expression of dopaminergic receptors. Levels of D₂-like receptors (D₂, D₃, and D₄), measured by [³H]YM-09151-2 binding are significantly increased by ~30% in the striatum, and nonsignificantly increased (17%) in the nucleus accumbens shell. In contrast to striatum, receptor autoradiography showed no difference in D₂-like autoreceptor density between knock-out and wild-type mice in the substantia nigra compacta, substantia nigra pars reticulata, and ventral tegmental area [16]. No difference was noted in the density of D₁-like receptors, as measured by [³H]SCH 23390 binding, or in the dopamine transporter by evaluation of [¹²⁵I]RTI-121 binding in striatum, nucleus accumbens, and olfactory tubercle. Mutant mice, however, exhibited a functional supersensitivity of D₁-dopamine receptors in the nucleus accumbens, with the agonist SKF 38,393 inducing c-Fos immunoreactivity to a greater extent in knock-out as compared to wild-type mice.

These findings highlight the following: (1) Amine increases in MAO B KO mice are highest for those with substrate specificity for MAO B (PEA), least for substrates with specificity for MAO A (5-HT, NE) or shared substrate specificity for MAO A and B (DA). (2) There is no evidence for a compensatory up-regulation of MAO A or likely, COMT. (3) Although MAO B KO mice do not show overt changes in dopamine, increases in PEA may still influence the dopaminergic system, possibly through its own indirect dopaminergic actions [3,66].

Though the age dependency of PEA increases has not been extensively studied, results show that PEA excretion in urine of MAO B KO mice at ages 4 to 6 months remains nearly tenfold above that seen in wild-type mice. Likewise PEA levels in MAO B KO brains remain eightfold above those of wild-type mice for animals 9 to 10 months of age [27].

Behavioral Changes

Phenylethylamine when administered pharmacologically has been found to exert strong amphetamine-like effects, with increased locomotor activity, anxiety, and at high doses, stereotypic behaviors [1,39,44]. Paradoxically MAO B mutant mice, despite their markedly elevated PEA levels, do not display evidence of hyperactivity, either in their home cages or when tested in a novel Open Field environment [27]. Nor is there evidence of increased anxiety as tested in the Elevated Plus-Maze [27] or evidence of stereotypic behaviors. In addition, unlike MAO A KO mice, MAO B KO mice do not demonstrate an increase in aggressive behavior in the resident-intruder paradigm [27]. These latter results are consistent with observations in human subjects with Norrie disease. Here those subjects with gene

deletions encompassing the MAO B gene do not demonstrate increased aggressive behavior [47].

Because of reports that the MAO B inhibitor L-deprenyl may facilitate maze performance in rodents [7,24,88], as well as aspects of cognition in humans [10,20,71], we examined memory and learning in the MAO B mutants. Adult animals show normal working memory as tested in a Y-maze [27], and both adult and aged mice demonstrate visual spatial learning in the Morris Water Maze equivalent to that seen in wild-type mice [28]. This work suggests that mechanisms by which L-deprenyl may improve maze performance are likely to be independent of the inhibition of MAO B, possibly acting through deprenyl's amphetamine metabolite [35].

Mutant mice compared to wild-type mice show increased mobility in the Porsolt Forced Swim Test, and show no adaptation at reexposure after 4 weeks [27]. This data is consistent with evidence that selective blockade of MAO B with deprenyl decreases immobility in this paradigm [64].

Physiological Changes

MAO B mutant mice in the resting, restrained state show normal or reduced blood pressures and heart rates compared to wild-type mice, in spite of showing up to eightfold elevations of PEA in tissues [72]. Such results appear counter-intuitive based on the known sympathetic central, as well as peripheral effects of PEA. When tested on resistance vessels *in vitro*, PEA increases smooth muscle tone and enhances stimulation-induced contractions by a NE displacing effect [37]. In the central nervous system, it has been suggested that PEA has a relatively potent effect inhibiting the uptake of DA, NE, and possibly 5-HT [31,66]. In addition, PEA stimulates the release of catecholamines and to a lesser extent 5-HT, and thus, may exert vasoactive effects indirectly through alternate neurotransmitter systems [3,66]. These findings emphasize that a lifelong absence of the MAO B gene likely activates adaptational mechanisms that serve to maintain the autonomic function of the mutant mice within the normal range.

In light of the vasoactive properties of PEA, we examined CBF measured with the autoradiographic [¹⁴C]-iodo-antipyrine method in MAO B knock-out mice [72]. MAO B KO mice exhibit a trend toward lower global CBF than their wild-type counterparts. This trend is accentuated in response to acute administration of PEA, with mutant animals showing a decrease in global CBF of ~38% compared to ~20% for controls. The most significant difference between mutant and wild-type mice, however, is a redistribution of regional CBF. Compared to wild-type mice, MAO B-deficient mice examined in the resting, restrained state show a higher regional CBF in midline motor cortex and medial portions of somatosensory and visual cortex; lower regional CBF is seen in the piriform cortex and anterolateral frontal cortex (including primary motor, primary somatosensory and anterior portions of the insular cortex). Administration of PEA enhances CBF in lateral frontal and piriform cortex in both MAO B deficient and wild-type mice. These changes may reflect a differential activation due to chronic and acute PEA elevations on motor and olfactory function.

MAO B KO mutants, unlike normal mice, are resistant to the neurodegenerative effects of MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine), a neurotoxin that induces a condition in animals that is reminiscent of Parkinson's disease [27]. Striatal brain tissue in MAO B mutants treated with MPTP showed none of the depletion of DA and DOPAC characteristic of wild-type mice. This confirms previous findings using pharmacological inhibitors of MAO B, that neurotoxicity of MPTP is dependent on its deamination to MPP⁺ (1-methyl-4-phenylpyridinium) by MAO B, which upon entering the dopaminergic neuron via the nerve terminal uptake carrier, damages the cell by inhibiting cellular respiration [67].

Whether MAO B in the brain represents a risk factor or a protective factor for neuronal damage following ischemia remains unclear. Two opposing mechanisms present themselves for clarification. The first mechanism proposes a protective role for MAO B based on the ability of PEA to regulate CBF distribution, and in particular to decrease CBF after acute administration. This hypothesis suggests that mice lacking MAO B might be at increased risk following cerebral ischemia because following a cerebral infarction a PEA-dependent hypoperfusion in the brain might result in a decreased perfusion of the ischemic penumbra, and a consequent increase in infarct size. The second mechanism, contrary to the first, proposes that MAO B activity might be a risk factor in determining outcome following ischemia. The catabolism of bio-amines by monoamine oxidase is reported to increase during ischemia [18] with a resultant increased production in hydrogen peroxide (H₂O₂) [76]. Inhibition of MAO B by L-deprenyl decreases the production of H₂O₂ [59], and it has been hypothesized that this may attenuate neuronal damage by H₂O₂ or the highly toxic hydroxyl radical which can be formed from H₂O₂ by iron-mediated Fenton reactions. To examine the role of MAO B in cerebral ischemia, we subjected MAO B KO and wild-type mice to unilateral middle cerebral artery occlusion [30]. No significant genotypic differences were detected in infarct volume or in the extent of brain edema. Similarly, no difference in these variables was detected for wild-type mice pretreated with L-deprenyl. Thus, neither absence of the MAO B gene nor pharmacological inhibition of the enzyme proved to be protective or detrimental in this animal model of acute cortical infarction. This suggests the existence of alternate mechanisms during ischemia that can accommodate the vasoactive effect of increases in PEA or the metabolic stress elicited by H₂O₂ following the increased catabolism of catecholamines by MAO A.

DISCUSSION

Study of transgenic and knock-out mice is increasingly demonstrating that disruption of a single gene has substantial effects, both direct and indirect throughout the organism at the biochemical, physiological, and behavioral levels. Though some of these represent an immediate consequence of the gene disruption, others represent changes downstream from the primary defect. Thus, in association with the increase in 5-HT levels in brain, MAO A KO mice show a down-regulation of post-synaptic 5-HT_{1A}, 5-HT_{2A}, and 5-HT_{2C} receptors; whereas, high levels of PEA in MAO B KO mice are associated with increases in D₂-like receptors. The latter example demonstrates that adaptational changes may also occur indirectly across different biochemical systems. The changes in D₂-like receptors binding in MAO B KO mice presumably are related to the fact that PEA exerts a strong action on the release and reuptake of DA. The fact that regional differences were noted in D₂-like

receptors binding in the brains of MAO B mutants, suggests that this adaptive regulation does not occur throughout the central nervous system. This further illustrates the complexity of downstream effects that may lead to marked regional differences in functional response. Such findings suggest that mice with single gene defects may be powerful tools in studying plasticity in development.

Absence of the MAO A gene results in widespread neurodevelopmental changes which in specific instances can prove a hindrance to the accurate interpretation of the role MAO plays in adult life in shaping behavior and physiologic function. Though future inducible knock-outs of MAO may avoid some of the ontogenetic problems inherent with conventional knock-outs, caution is required here as well in interpreting the resulting phenotypes. The potential for redundancy mechanisms masking changes in behavior or physiologic function remains, even if a gene is acutely turned “on” in late life. Furthermore, the complex orchestration of gene activation and inactivation during development is essential in determining neuronal migration and brain structure in a manner which may either facilitate or constrain the postnatal repertoire of functional changes. Eliciting functional changes in adult life by turning a gene “on” or “off” may, in specific instances, require gene activation/inactivation at specified antenatal stages in such a manner as to reproduce “normal” development.

We need to be careful about the anthropomorphic interpretations that we impose on knock-out and transgenic animals, and refrain from overgeneralizations about our findings. Low levels of MAO A in human subjects do not necessarily lead to violent or impulsive behavior. Acute inhibition of MAO A and B with tranylcypromine or phenelzine in human subjects does not result in significant aggressive personality changes, though occasional hypomania has been reported in patients with an underlying manic-depressive diathesis [62]. In rodents pharmacologic inhibition of MAO A can both induce or reduce aggressive behavior [19,23,89], likely due to the fact that aggression in animals can be subdivided into several subtypes [61]. Deficiencies of MAO A throughout life [8] likely have different consequences on aggression than acute pharmacologic inhibition of the enzyme. However, marked lifelong reductions in MAO activity are highly uncommon in human subjects [56] and are unlikely to be implicated as a cause in everyday aggressive behavior. The question to what extent moderate reductions in MAO activity might contribute to dysfunctional behavior remains to be defined, particularly if such reductions occur at specific time points during early development [12,85]. Apart from the MAO A-deficient mice, mice with targeted disruptions of genes for α -calcium-calmodulin kinase II [15], the 5-HT_{1B} receptor [70], and the neuronal isoform of nitric oxide synthetase [60], all display increased aggression. These observations highlight the complexity of the multiple behavioral and physiologic parameters that underlie the characteristic phenotypes associated with low MAO activity.

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References

1. Aliyu SU, Sewell RD. Effects of beta-phenylethylamine on locomotor activity, body temperature and ethanol blood concentrations during acute ethanol intoxication. *Psychopharmacology (Berl)*. 1987; 93:69–71. [PubMed: 3114816]
2. Bach AW, Lan NC, Johnson DL, Abell CW, Bembenek ME, Kwan SW, Seeburg PH, Shih JC. cDNA cloning of human liver monoamine oxidase A and B: Molecular basis of differences in enzymatic properties. *Proc Natl Acad Sci USA*. 1988; 85:4934–4938. [PubMed: 3387449]
3. Bailey BA, Phillips SR, Boulton AA. In vivo release of endogenous dopamine, 5-hydroxytryptamine and some of their metabolites from rat caudate nucleus by phenylethylamine. *Neurochem Res*. 1987; 12:173–178. [PubMed: 2437473]
4. Borsini F. Role of the serotonergic system in the forced swimming test. *Neurosci Biobehav Rev*. 1995; 19:377–395. [PubMed: 7566740]
5. Borsini F, Meli A. Is the forced swimming test a suitable model for revealing antidepressant activity? *Psychopharmacology (Berl)*. 1988; 94:147–160. [PubMed: 3127840]
6. Bou-Flores C, Lajard AM, Monteau R, De Maeyer E, Seif I, Lanoir J, Hilaire G. Abnormal phrenic motoneuron activity and morphology in neonatal monoamine oxidase A-deficient transgenic mice: Possible role of a serotonin excess. *J Neurosci*. 2000; 20:4646–4656. [PubMed: 10844034]
7. Brandeis R, Sapir M, Kapon Y, Borelli G, Cadel S, Valsecchi B. Improvement of cognitive function by MAO-B inhibitor L-deprenyl in aged rats. *Pharmacol Biochem Behav*. 1991; 39:297–304. [PubMed: 1946572]
8. Brunner HG, Nelen M, Breakefield XO, Ropers HH, van Oost BA. Abnormal behavior associated with a point mutation in the structural gene for monoamine oxidase A. *Science*. 1993; 262:578–580. [PubMed: 8211186]
9. Caccia S, Ballabio M, Guiso G, Rocchetti M, Garattini S. Species differences in the kinetics and metabolism of fenfluramine isomers. *Arch Int Pharmacodyn Ther*. 1982; 258:15–28. [PubMed: 7138139]
10. Campi N, Todeschini GP, Scarzella L. Selegiline versus L-acetylcarnitine in the treatment of Alzheimer-type dementia. *Clin Ther*. 1990; 12:306–314. [PubMed: 2121359]
11. Cases O, Lebrand C, Giros B, Vitalis T, De Maeyer E, Caron MG, Price DJ, Gaspar P, Seif I. Plasma membrane transporters of serotonin, dopamine, and norepinephrine mediate serotonin accumulation in atypical locations in the developing brain of monoamine oxidase A knock-outs. *J Neurosci*. 1998; 18:6914–6927. [PubMed: 9712661]
12. Cases O, Seif I, Grimsby J, Gaspar P, Chen K, Pournin S, Muller U, Aguet M, Babinet C, Shih JC, De Maeyer E. Aggressive behavior and altered amounts of brain serotonin and nor-epinephrine in mice lacking MAOA. *Science*. 1995; 268:1763–1766. [PubMed: 7792602]
13. Cases O, Vitalis T, Seif I, De Maeyer E, Sotelo C, Gaspar P. Lack of barrels in the somatosensory cortex of monoamine oxidase A-deficient mice: Role of a serotonin excess during the critical period. *Neuron*. 1996; 16:297–307. [PubMed: 8789945]
14. Cazalets JR, Gardette M, Hilaire G. Locomotor network maturation is transiently delayed in the MAOA-deficient mouse. *J Neurophysiol*. 2000; 83:2468–2470. [PubMed: 10758149]
15. Chen C, Rainnie DG, Greene RW, Tonegawa S. Abnormal fear response and aggressive behavior in mutant mice deficient for alpha-calcium-calmodulin kinase II. *Science*. 1994; 266:291–294. [PubMed: 7939668]
16. Chen L, He M, Sibille E, Thompson A, Sarnyai Z, Baker H, Shippenberg T, Toth M. Adaptive changes in postsynaptic dopamine receptors despite unaltered dopamine dynamics in mice lacking mono-amine oxidase B. *J Neurochem*. 1999; 73:647–655. [PubMed: 10428061]
17. Constantino JN, Morris JA, Murphy DL. CSF 5-HIAA and family history of antisocial personality disorder in newborns. *Am J Psychiatry*. 1997; 154:1771–1773. [PubMed: 9396964]
18. Damsma G, Boisvert DP, Mudrick LA, Wenkstern D, Fibiger HC. Effects of transient forebrain ischemia and pargyline on extra-cellular concentrations of dopamine, serotonin, and their metabolites in the rat striatum as determined by in vivo microdialysis. *J Neurochem*. 1990; 54:801–808. [PubMed: 2303812]

19. Datla KP, Bhattacharya SK. Effect of selective monoamine oxidase A and B inhibitors on footshock induced aggression in paired rats. *Indian J Exp Biol.* 1990; 28:742–745. [PubMed: 2123820]
20. Falsaperla A, Monici Preti PA, Oliani C. Selegiline versus oxiracetam in patients with Alzheimer-type dementia. *Clin Ther.* 1990; 12:376–384. [PubMed: 2125242]
21. Fletcher A, Forster EA, Bill DJ, Brown G, Cliffe IA, Hartley JE, Jones DE, McLenachan A, Stanhope KJ, Critchley DJ, Childs KJ, Middlefell VC, Lanfumey L, Corradetti R, Laporte AM, Gozlan H, Hamon M, Dourish CT. Electro-physiological, biochemical, neurohormonal and behavioural studies with WAY-100635, a potent, selective and silent 5-HT_{1A} receptor antagonist. *Behav Brain Res.* 1996; 73:337–353. [PubMed: 8788530]
22. Fornai F, Chen K, Giorgi FS, Gesi M, Alessandri MG, Shih JC. Striatal dopamine metabolism in monoamine oxidase B-deficient mice: A brain dialysis study. *J Neurochem.* 1999; 73:2434–2440. [PubMed: 10582603]
23. Gao B, Cutler MG. Effects of acute and chronic administration of the antidepressants, imipramine, phenelzine and mianserin, on the social behaviour of mice. *Neuropharmacol.* 1994; 33:813–824.
24. Gelowitz DL, Richardson JS, Wishart TB, Yu PH, Lai CT. Chronic L-deprenyl or L-amphetamine: Equal cognitive enhancement, unequal MAO inhibition. *Pharmacol Biochem Behav.* 1994; 47:41–45. [PubMed: 8115427]
25. Goodwin GM, De Souza RJ, Green AR. Attenuation by electroconvulsive shock and antidepressant drugs of the 5-HT_{1A} receptor-mediated hypothermia and serotonin syndrome produced by 8-OH-DPAT in the rat. *Psychopharmacology.* 1987; 91:500–505. [PubMed: 2954178]
26. Grimsby J, Chen K, Wang LJ, Lan NC, Shih JC. Human monoamine oxidase A and B genes exhibit identical exon-intron organization. *Proc Natl Acad Sci USA.* 1991; 88:3637–3641. [PubMed: 2023912]
27. Grimsby J, Toth M, Chen K, Kumazawa T, Klaidman L, Adams JD, Karoum F, Gal J, Shih JC. Increased stress response and beta-phenylethylamine in MAOB-deficient mice. *Nat Genet.* 1997; 17:206–210. [PubMed: 9326944]
28. Holschneider DP, Scremin OU, Chen K, Shih JC. Lack of protection of monoamine oxidase B-deficient mice from age-related spatial learning deficits in the Morris water maze. *Life Sci.* 1999; 65:1757–1763. [PubMed: 10576555]
29. Holschneider DP, Scremin OU, Huynh L, Chen K, Seif I, Shih JC. Regional cerebral cortical activation in monoamine oxidase A-deficient mice: Differential effects of chronic versus acute elevations in serotonin and norepinephrine. *Neuroscience.* 2000; 101:869–877. [PubMed: 11113335]
30. Holschneider DP, Scremin OU, Huynh L, Chen K, Shih JC. Lack of protection from ischemic injury of monoamine oxidase B-deficient mice following middle cerebral artery occlusion. *Neurosci Lett.* 1999; 259:161–164. [PubMed: 10025583]
31. Horn AS, Snyder SH. Steric requirements for catecholamine uptake by rat brain synaptosomes: Studies with rigid analogs of amphetamine. *J Pharmacol Exp Ther.* 1972; 180:523–530. [PubMed: 5012779]
32. Ikemoto K, Kitahama K, Seif I, Maeda T, De Maeyer E, Valatx JL. Monoamine oxidase B (MAOB)-containing structures in MAOA-deficient transgenic mice. *Brain Res.* 1997; 771:121–132. [PubMed: 9383015]
33. Itoh, M. *The cerebellum and neural control.* New York: Raven; 1984.
34. Jacobs BL, Klemfuss H. Brain stem and spinal cord mediation of a serotonergic behavioral syndrome. *Brain Res.* 1975; 100:450–457. [PubMed: 1192191]
35. Karoum F, Chuang LW, Eisler T, Calne DB, Liebowitz MR, Quitkin FM, Klein DF, Wyatt RJ. Metabolism of (–) deprenyl to amphetamine and methamphetamine may be responsible for deprenyl's therapeutic benefit: A biochemical assessment. *Neurology.* 1982; 32:503–509. [PubMed: 6803190]
36. Kim JJ, Shih JC, Chen K, Chen L, Bao S, Maren S, Anagnostaras SG, Fanselow MS, De Maeyer E, Seif I, Thompson RF. Selective enhancement of emotional, but not motor, learning in

- monoamine oxidase A-deficient mice. *Proc Natl Acad Sci USA*. 1997; 94:5929–5933. [PubMed: 9159177]
37. Knoll J, Miklya I, Knoll B, Marko R, Racz D. Phenylethylamine and tyramine are mixed-acting sympathomimetic amines in the brain. *Life Sci*. 1996; 58:2101–2114. [PubMed: 8649195]
 38. Koide Y, Kobayashi K. Developmental changes in the activity and substrate specificities of mouse brain monoamine oxidase. *Neurochem Res*. 1984; 9:595–606. [PubMed: 6472564]
 39. Kuroki T, Tsutsumi T, Hirano M, Matsumoto T, Tatebayashi Y, Nishiyama K, Uchimura H, Shiraishi A, Nakahara T, Nakamura K. Behavioral sensitization to beta-phenylethylamine (PEA): Enduring modifications of specific dopaminergic neuron systems in the rat. *Psychopharmacology (Berl)*. 1990; 102:5–10. [PubMed: 2392508]
 40. Laferriere B, Wurtman RJ. Effect of D-fenfluramine on serotonin release in brains of anaesthetized rats. *Brain Res*. 1989; 504:258–263. [PubMed: 2480833]
 41. Lamensdorf I, Youdim MB, Finberg JP. Effect of long-term treatment with selective monoamine oxidase A and B inhibitors on dopamine release from rat striatum in vivo. *J Neurochem*. 1996; 67:1532–1539. [PubMed: 8858937]
 42. Lan NC, Heinzmann C, Gal A, Klisak I, Orth U, Lai E, Grimsby J, Sparkes RS, Mohandas T, Shih JC. Human monoamine oxidase A and B genes map to Xp 11.23 and are deleted in a patient with Norrie disease. *Genomics*. 1989; 4:552–559. [PubMed: 2744764]
 43. Land PW, Rose LL. Exogenous monoamines affect the segregation of retinogeniculate fibers in developing rats. *Brain Res*. 1985; 354:135–140. [PubMed: 3930005]
 44. Lapin IP. Anxiogenic effect of phenylethylamine and amphetamine in the elevated plus-maze in mice and its attenuation by ethanol. *Pharmacol Biochem Behav*. 1993; 44:241–243. [PubMed: 8094251]
 45. Le Douarec JC, Schmitt H, Laubie M. Pharmacologic study of fenfluramine and its optical isomers. *Arch Int Pharmacodyn Ther*. 1966; 161:206–232. [PubMed: 5941878]
 46. Lebrand C, Cases O, Adelbrecht C, Doye A, Alvarez C, El Mestikawy S, Seif I, Gaspar P. Transient uptake and storage of serotonin in developing thalamic neurons. *Neuron*. 1996; 17:823–835. [PubMed: 8938116]
 47. Lenders JW, Eisenhofer G, Abeling NG, Berger W, Murphy DL, Konings CH, Wagemakers LM, Kopin IJ, Karoum F, van Gennip AH, Brunner HG. Specific genetic deficiencies of the A and B isoenzymes of monoamine oxidase are characterized by distinct neurochemical and clinical phenotypes. *J Clin Invest*. 1996; 97:1010–1019. [PubMed: 8613523]
 48. Liang KC, McGaugh JL, Yao HY. Involvement of amygdala pathways in the influence of post-training intra-amygdala norepinephrine and peripheral epinephrine on memory storage. *Brain Res*. 1990; 508:225–233. [PubMed: 2306613]
 49. Lidberg L, Tuck JR, Asberg M, Scalia-Tomba GP, Bertilsson L. Homicide, suicide and CSF 5-HIAA. *Acta Psychiatr Scand*. 1985; 71:230–236. [PubMed: 2580421]
 50. Maj J, Chojnacka-Wojcik E, Lewandowska A, Tatarczynska E. Central antiserotonin action of fluperlapine. *Pol J Pharmacol Pharm*. 1985; 37:517–524. [PubMed: 2867538]
 51. Mann JJ, McBride PA, Anderson GM, Mieczkowski TA. Platelet and whole blood serotonin content in depressed inpatients: Correlations with acute and life-time psychopathology. *Biol Psychiatry*. 1992; 32:243–257. [PubMed: 1420642]
 52. McGaugh JL, Introini-Collison IB, Nagahara AH, Cahill L, Brioni JD, Castellano C. Involvement of the amygdaloid complex in neuromodulatory influences on memory storage. *Neurosci Biobehav Rev*. 1990; 14:425–431. [PubMed: 1981091]
 53. Miczek KA, Mos J, Olivier B. Brain 5-HT and inhibition of aggressive behavior in animals: 5-HIAA and receptor subtypes. *Psychopharmacol Bull*. 1989; 25:399–403. [PubMed: 2483273]
 54. Moffitt TE, Brammer GL, Caspi A, Fawcett JP, Raleigh M, Yuwiler A, Silva P. Whole blood serotonin relates to violence in an epidemiological study. *Biol Psychiatry*. 1998; 43:446–457. [PubMed: 9532350]
 55. Muller U, Steinhoff U, Reis LF, Hemmi S, Pavlovic J, Zinkernagel RM, Aguet M. Functional role of type I and type II interferons in antiviral defense. *Science*. 1994; 264:1918–1921. [PubMed: 8009221]

56. Murphy DL, Sims K, Eisenhofer G, Greenberg BD, George T, Berlin F, Zametkin A, Ernst M, Breakefield XO. Are MAO-A deficiency states in the general population and in putative high-risk populations highly uncommon? *J Neural Transm.* 1998; 52(suppl):29–38.
57. Murphy DL, Sims KB, Karoum F, de la Chapelle A, Norio R, Sankila EM, Breakefield XO. Marked amine and amine metabolite changes in Norrie disease patients with an X-chromosomal deletion affecting monoamine oxidase. *J Neurochem.* 1990; 54:242–247. [PubMed: 2293615]
58. Nakajima K, Matsuyama K, Mori S. Prenatal administration of parachlorophenylalanine results in suppression of serotonergic system and disturbance of swimming movements in newborn rats. *Neurosci Res.* 1998; 31:155–169. [PubMed: 9700721]
59. Nakano T, Kawai S, Nagatsu T. A simple assay for monoamine oxidase using glutathione peroxidase and glutathione reductase. *Jpn J Pharmacol.* 1984; 35:163–167. [PubMed: 6748378]
60. Nelson RJ, Demas GE, Huang PL, Fishman MC, Dawson VL, Dawson TM, Snyder SH. Behavioural abnormalities in male mice lacking neuronal nitric oxide synthase. *Nature.* 1995; 378:383–386. [PubMed: 7477374]
61. Olivier B, Mos J, van Oorschot R, Hen R. Serotonin receptors and animal models of aggressive behavior. *Pharmacopsychiatry.* 1995; 28(suppl. 2):80–90. [PubMed: 8614705]
62. Pickar D, Murphy DL, Cohen RM, Campbell IC, Lipper S. Selective and nonselective monoamine oxidase inhibitors: Behavioral disturbances during their administration to depressed patients. *Arch Gen Psychiatry.* 1982; 39:535–540. [PubMed: 7092487]
63. Pliszka SR, Rogeness GA, Renner P, Sherman J, Broussard T. Plasma neurochemistry in juvenile offenders. *J Am Acad Child Adolesc Psychiatry.* 1988; 27:588–594. [PubMed: 3182623]
64. Porsolt RD, Bertin A, Jalfre M. Behavioral despair in mice: A primary screening test for antidepressants. *Arch Int Pharmacodyn Ther.* 1977; 229:327–336. [PubMed: 596982]
65. Porsolt RD, Le Pichon M, Jalfre M. Depression: A new animal model sensitive to antidepressant treatments. *Nature.* 1977; 266:730–732. [PubMed: 559941]
66. Raiteri M, Del Carmine R, Bertollini A, Levi G. Effect of sympathomimetic amines on the synaptosomal transport of noradrenaline, dopamine and 5-hydroxytryptamine. *Eur J Pharmacol.* 1977; 41:133–143. [PubMed: 832672]
67. Ramsay RR, Krueger MJ, Youngster SK, Gluck MR, Casida JE, Singer TP. Interaction of 1-methyl-4-phenylpyridinium ion (MPP+) and its analogs with the rotenone/piroxicin binding site of NADH dehydrogenase. *J Neurochem.* 1991; 56:1184–1190. [PubMed: 2002336]
68. Roos K, Holschneider DP, Scremin OU, Chen L, Shih JC. Monoamine oxidase A and B deficient mice exhibit altered baroreceptor reflexes. *Circulation.* 2000; 102:11–13. [PubMed: 10880408]
69. Salichon N, Gaspar P, Upton AL, Picaud S, Hanoun N, Hamon M, De Maeyer E, Murphy DL, Mossner RLKP, Hen R, Seif I. Excessive activation of 5-HT_{1B} receptors disrupts the formation of sensory maps in monoamine oxidase A and 5-HT transporter knockout mice. *J Neuroscience.* 2001; 21:884–896.
70. Saudou F, Amara DA, Dierich A, LeMeur M, Ramboz S, Segu L, Buhot MC, Hen R. Enhanced aggressive behavior in mice lacking 5-HT_{1B} receptor. *Science.* 1994; 265:1875–1878. [PubMed: 8091214]
71. Schneider LS, Pollock VE, Zemansky MF, Gleason RP, Palmer R, Sloane RB. A pilot study of low-dose L-deprenyl in Alzheimer's disease. *J Geriatr Psychiatry Neurol.* 1991; 4:143–148. [PubMed: 1953966]
72. Scremin OU, Holschneider DP, Chen K, Li MG, Shih JC. Cerebral cortical blood flow maps are reorganized in MAOB-deficient mice. *Brain Res.* 1999; 824:36–44. [PubMed: 10095040]
73. Series HG, Cowen PJ, Sharp T. p-Chloroamphetamine (PCA), 3,4-methylenedioxy-methamphetamine (MDMA) and d-fenfluramine pretreatment attenuates d-fenfluramine-evoked release of 5-HT in vivo. *Psychopharmacology (Berl).* 1994; 116:508–514. [PubMed: 7535469]
74. Shih JC, Ridd MJ, Chen K, Meehan WP, Kung MP, Seif I, De Maeyer E. Ketanserin and tetrabenazine abolish aggression in mice lacking monoamine oxidase A. *Brain Res.* 1999; 835:104–112. [PubMed: 10415365]
75. Shimizu H, Bray GA. Hypothalamic monoamines measured by microdialysis in rats treated with 2-deoxy-glucose or d-fenfluramine. *Physiol Behav.* 1989; 46:799–807. [PubMed: 2628991]

76. Simonson SG, Zhang J, Canada AT Jr, Su YF, Benveniste H, Piantadosi CA. Hydrogen peroxide production by monoamine oxidase during ischemia-reperfusion in the rat brain. *J Cereb Blood Flow Metab.* 1993; 13:125–134. [PubMed: 8417001]
77. Sims KB, de la Chapelle A, Norio R, Sankila EM, Hsu YP, Rinehart WB, Corey TJ, Ozelius L, Powell JF, Bruns G. Monoamine oxidase deficiency in males with an X chromosome deletion. *Neuron.* 1989; 2:1069–1076. [PubMed: 2483108]
78. Tsang D, Ho KP, Wen HL. Ontogenesis of multiple forms of monoamine oxidase in rat brain regions and liver. *Dev Neurosci.* 1986; 8:243–250. [PubMed: 3829988]
79. Unis AS, Cook EH, Vincent JG, Gjerde DK, Perry BD, Mason C, Mitchell J. Platelet serotonin measures in adolescents with conduct disorder. *Biol Psychiatry.* 1997; 42:553–559. [PubMed: 9376451]
80. Upton AL, Salichon N, Lebrand C, Ravary A, Blakely R, Seif I, Gaspar P. Excess of serotonin (5-HT) alters the segregation of ipsilateral and contralateral retinal projections in monoamine oxidase A knock-out mice: Possible role of 5-HT uptake in retinal ganglion cells during development. *J Neurosci.* 1999; 19:7007–7024. [PubMed: 10436056]
81. Vertes RP. A PHA-L analysis of ascending projections of the dorsal raphe nucleus in the rat. *J Comp Neurol.* 1991; 313:643–668. [PubMed: 1783685]
82. Vitalis T, Cases O, Callebert J, Launay JM, Price DJ, Seif I, Gaspar P. Effects of monoamine oxidase A inhibition on barrel formation in the mouse somatosensory cortex: Determination of a sensitive developmental period. *J Comp Neurol.* 1998; 393:169–184. [PubMed: 9548695]
83. Waterhouse BD, Devilbiss D, Fleischer D, Sessler FM, Simpson KL. New perspectives on the functional organization and postsynaptic influences of the locus ceruleus efferent projection system. *Adv Pharmacol.* 1998; 42:749–754. [PubMed: 9328007]
84. Waterhouse BD, Lin CS, Burne RA, Woodward DJ. The distribution of neocortical projection neurons in the locus coeruleus. *J Comp Neurol.* 1983; 217:418–431. [PubMed: 6886061]
85. Whitaker-Azmitia PM, Zhang X, Clarke C. Effects of gestational exposure to monoamine oxidase inhibitors in rats: Preliminary behavioral and neurochemical studies. *Neuropsychopharmacology.* 1994; 11:125–132. [PubMed: 7840864]
86. Wiczynska B, Chojnacka-Wojcik E, Rokosz-Pelc A, Mazela H, Przegalinski E. Antiserotonin activity of 9-methyl-2-[3-(4-phenyl-1-piperazinylpropyl)]-1,2,3,4-tetrahydro-beta-carboline-1-one (B-193). *Pol J Pharmacol Pharm.* 1989; 41:169–181. [PubMed: 2556691]
87. Yang Z, Seif I, Armstrong-James M. Differences in somatosensory processing in S1 barrel cortex between normal and monoamine oxidase A knockout (Tg8) adult mice. *Cereb Cortex.* 2001; 11:26–36. [PubMed: 11113033]
88. Yavich L, Sirvio J, Haapalinna A, Puumala T, Koivisto E, Heinonen E, Riekkinen PJ. The systemic administration of tacrine or selegiline facilitate spatial learning in aged fisher 344 rats. *J Neural Transm Gen Sect.* 1996; 103:619–626.
89. Zetler G, Otten U. Aggressiveness of the rat following combined treatment with monoamine oxidase inhibitors and other psychotropic drugs, especially thymoleptics. *Naunyn Schmiedeberg Arch Pharmacol.* 1969; 264:32–54. [PubMed: 4390354]

TABLE 1

Changes of MAO A KO Mice Compared To Wild-type Mice	Ref.
Biochemical	
No enzymatic MAO A activity, protein or MAO A mRNA in brain and peripheral tissues	[12]*
Normal levels of MAO B in brain and peripheral tissues	[12]*
Increased levels of 5-HT, NE in whole brain, hippocampus, frontal cortex, and cerebellum	[12,36]
Decreased tissue levels of the 5-HT metabolite 5-HIAA	[12]
Downregulation of brain postsynaptic 5-HT _{1A} , 5-HT _{2A} , 5-HT _{2C} receptors and the vesicular monoamine transporter (VMAT2)	[6,74]
Neurodevelopmental	
Abnormal development of somatosensory thalamocortical afferent fibers	[12]
Abnormal segregation of retinal afferents to the dorsal lateral geniculate nucleus, as well as to the superior colliculus	[80]
Atypical locations of 5-HT during embryonic and postnatal development	[11]
Abnormal activity and morphology of phrenic motoneurons in neonates	[6]
Transient delay of locomotor network maturation	[14]
Behavioral	
Neonatal	
Prolonged righting, trembling upon locomotion, and hunched posture	[12]
Frantic running and falling over, jumping, or prompt digging to hide under woodshavings in response to moderate sound and movement	
Prolonged and stronger reactions to pinching	
Propensity to bite the experimenter	
Sleep accompanied by violent shaking and jumps	
Adult	
Normal nesting, nursing, and pup-retrieval behavior in females	[36]
Increased territorial aggression in the resident-intruder paradigm, increased bite wounds in male, group-housed mice	[12]
Decreased social investigative behavior in males in the resident-intruder paradigm with static, hunched, fluffed-fur posture after first olfactory stimulus.	[12]
Increased sensitivity to developing the serotonin syndrome with fenfluramine challenge	[29]
Behavioral syndrome characterized by restlessness, attentional deficits, disrupted social interaction, feeding and self-grooming after administration of the MAO B inhibitors L-deprenyl or lazabemide	[12]
Enhancement of classical fear conditioning and step-down inhibitory avoidance learning (emotional memory) but not of eyeblink conditioning (motor learning)	[36]
Increased mobility in the Porsolt Forced Swim Test	[12]
Increased time spent in the center in the Open Field test with much hesitation as to which direction to take	[12]
Abnormal walking on a balance-beam with adults grasping the lateral aspects of the beam with hindlimbs during movement	[12,69]
Altered courtship with increased grasping events in males and concurrent increased frequency and intensity of vocalization in females.	[12]
Physiologic	
Decreased blood pressure and heart rate in the resting, restrained state	[29]
Exaggerated hyperthermic and tachycardic response to fenfluramine	[29]
Regional cerebral cortical blood flow: Higher in somatosensory and barrel field neocortex; lower in entorhinal and midline motor cortex	[29]
Decreased body weight ($\approx -10\%$)	[29]

* Includes data from unpublished observations.

TABLE 2

Changes of MAO B KO Mice Compared To Wild-type Mice	Ref.
Biochemical	
No enzymatic MAO B activity, protein or MAO B mRNA in brain and peripheral tissues	[27]*
Normal levels of MAO A in brain and peripheral tissues	[27]*
Increased levels of PEA in whole brain	[27]
Increased urinary excretion of PEA	[27]
Normal levels of 5-HT, NE, DA, 5-HIAA, DOPAC, HVA in cortex, hippocampus, raphe nucleus, substantia nigra, thalamus	[27]
Normal levels of extracellular DA, DOPAC, 3-MT, HVA in striatum, but elevated DA levels after administration of high dose L-DOPA	[16,22]
Functional supersensitivity of D ₁ -like dopamine receptors in the nucleus accumbens	[16]
Normal density of D ₁ -like receptors and normal tyrosine hydroxylase immunoreactivity in striatum, nucleus accumbens, and olfactory tubercle	[16]
Up-regulation of the D ₂ -like dopamine receptors in the striatum and nucleus accumbens shell, but not in substantia nigra, and ventral tegmental area	[16]
Behavioral	
Adult	
No increase in aggressive behavior	[27]
Normal visual-spatial learning in adult and in aged animals in the Morris Water Maze	[28]
Intact working memory as tested in a Y-maze	[27]
Increased mobility in the Porsolt Forced Swim Test	[27]
Normal response in the Elevated Plus-Maze	[27]
Normal exploratory activity in the Open Field	[27]
Physiologic	
Decreased blood pressure and heart rate in the resting, restrained state	[72]
Exaggerated hypertensive response to PEA	[72]
Regional cerebral cortical blood flow: Higher in midline motor cortex and medial portions of somatosensory and visual cortex; lower in piriform and anterolateral frontal cortex	[72]
No difference from wild-type mice in infarct volume or extent of brain edema following middle cerebral artery occlusion	[30]
Lack of MPTP toxicity in brain	[27]
Normal body weight	[30,72]

* Includes data from unpublished observations.