CNS Drug Reviews
Vol. 7, No. 3, pp. 317–345

NEW CONCEPTS

Antiaging Compounds: (–)Deprenyl (Selegiline) and (–)1-(Benzofuran-2-yl)-2-propylaminopentane, [(–)BPAP], a Selective Highly Potent Enhancer of the Impulse Propagation Mediated Release of Catecholamines and Serotonin in the Brain

J. Knoll

Department of Pharmacology, Faculty of General Medicine, Semmelweis University, Budapest, Hungary

Key Words: Selegiline—Deprenyl—Antiaging drugs—BPAP.

ABSTRACT

Hundreds of millions of people now die over the age of 80 years primarily due to twentieth century progress in hygiene, chemotherapy, and immunology. With a longer average lifespan, the need to improve quality of life during the latter decades is more compelling. "Aging — The Epidemic of the New Millenium," a recent international conference (Monte Carlo, June 17–18, 2000), showed with peculiar clarity that a safe and efficient drug strategy to slow the age-related decay of brain performance is still missing. This review summarizes the physiologic and pharmacologic arguments in favor of a peculiar lifelong prophylactic medication with reasonable chances to keep in check brain aging and decrease the precipitation of age-related neurological diseases.

Address correspondence and reprint requests to: Prof. J. Knoll, Department of Pharmacology, Faculty of General Medicine, Semmelweis University, Budapest, P.O.B., H-1445, Hungary.
Tel./Fax: +36 (1) 210-4405; E-mail: joszefknoll@hotmail.com.

INTRODUCTION

A still not fully understood brain activation mechanism ensures the survival of mammalian organisms in their hostile natural environment. Deceptive descriptions (e.g., "drive" or "motivation") camouflage our dense ignorance regarding the essence of the specific activation mechanism in the mammalian brain that ensures that living beings surmount every obstacle to reach a goal, even if life is in the balance. Well-known large individual differences in performance and lifespan in any random population of the same strain are due primarily to inborn differences in the ability to properly activate the brain when needed (for review see ref. 26,35).

The catecholaminergic machinery, the engine of the brain, plays a rate-limiting role in the basic activation of the central nervous system (CNS). We may say that an animal born with a better engine will be the better-performing, longer-living individual. Indeed, we found that lower-performing rats died significantly earlier than their higher-performing peers (31). We analyzed, therefore, in a more concrete manner the relationship between performance and longevity in the rat (46). We selected from a large random population of young male rats ($n = 1600$) the sexually inactive (low performing [LP]; $n = 94$) and sexually most active rats (high performing [HP]; $n = 99$) and treated thereafter the rats with saline and (–)deprenyl, respectively, until they died. The study proved that HP rats, selected as the most active copulators, performed significantly better on a learning test and lived significantly longer than their LP peers. For example, saline-treated LP rats lived 134.58 ± 2.29 weeks, while their HP peers lived 151.24 ± 1.36 weeks ($P < 0.001$). On the other hand, both LP and HP rats treated with (–)deprenyl, an enhancer of the impulse-evoked release of catecholamines in the brain (for review see ref. 37), performed in fact significantly better in sexual and learning tests and lived longer than the saline-treated rats. For example, the lifetime of (–)deprenyl-treated LP rats (152.54 ± 1.36 weeks) was significantly ($P < 0.001$) longer than the lifetime of their saline-treated peers (134.58 ± 2.29 weeks), and HP rats treated with (–)deprenyl lived 185.30 ± 1.96 weeks, significantly ($P < 0.001$) longer than their saline-treated peers (151.24 ± 1.36).

A better understanding of the reason for the large individual differences in brain performance and longevity, as well as the motives of the slow age-related decay of brain function in mammals, is likely to leed to a better chance of developing a safe and efficient prophylactic antiaging medication.

THE CATECHOLAMINERGIC/SEROTONINERGIC ACTIVITY ENHANCER MECHANISM

Phenylethylamine and Tryptamine: Endogenous Enhancer Substances

Until recently, the brain activation mechanism of (–)deprenyl (47) and phenylethylamine (PEA) (48), the catecholaminergic/serotonergic activity enhancer (CAE/SAE) regulation, was unknown. For the sake of brevity, we shall refer to this as the "enhancer" mechanism. We may define, for the time being, the essence of the enhancer regulation as: the existence of enhancer-sensitive neurons in the brain that are capable of working in a

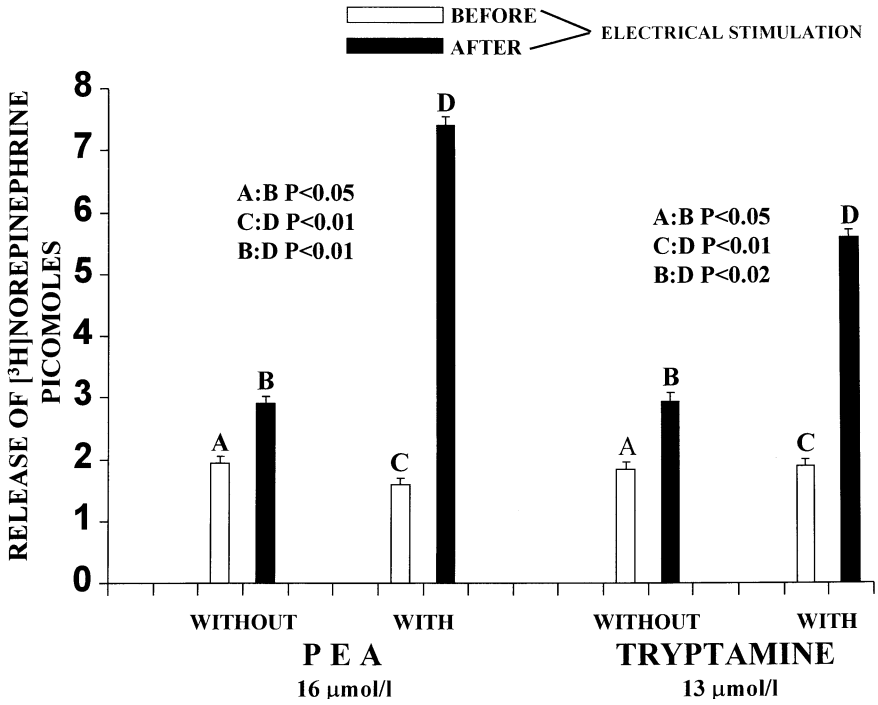


Fig. 1. The significant enhancement of the electrical-stimulation-induced release of [^{3}H]norepinephrine from isolated rat brain stem in the presence of the endogenous enhancer substances, PEA, and tryptamine, respectively. Each column represents the amount of [^{3}H]norepinephrine in picomoles released in a 3-min collection period. $N = 8$. Vertical lines show S.E.M. Paired Student's t-test. See ref. 49 for methodology.

split second on a significantly higher activity level under the influence of endogenous enhancer substances. PEA and tryptamine are presently the only experimentally analyzed examples of endogenous enhancer substances.

Although the effect of enhancer substances in the CNS is not restricted to the catecholinaminergic/serotonergic neurons, as will be described later, the most convenient method to get acquainted with this regulation is to follow an enhancer substance-induced increase in the impulse-evoked release of norepinephrine, dopamine, or serotonin. This effect is dose-dependent, immediate, and dramatic.

The existence of an enhancer regulation brings a different perspective to the brain-organized realization of goal-oriented behavior, which is the essence of plastic behavioral descriptions such as drive or motivation. Figure 1 shows, for example, the characteristic enhancer effect of PEA and tryptamine on the release of [^{3}H]norepinephrine from the isolated brain stem of the rat. A stable amount of [^{3}H]norepinephrine is released from the freshly isolated brain stem of a properly pretreated rat for a couple of hours (for methodology see ref. 48). Electrical stimulation of the brain stem significantly increases the outflow of the transmitter. The calculated average amount of [^{3}H]norepinephrine released from the stimulated brain stem is the product of a population of noradrenergic neurons with large individual variation in their response to the same stimulus. PEA and tryptamine

usually leave the resting activity of the neurons unchanged; however, the response to stimulation is highly significantly increased.

To interpret the phenomenon shown in Fig. 1, we may accept the classical view that the neuron responds to stimulation in an "all or none" manner and, when properly stimulated, it emits the same amount of transmitter. Hence, prior to the administration of PEA or tryptamine, only the most excitable neurons, the HP members of the population, responded with transmitter release to the applied electrical stimulation. In the presence of PEA or tryptamine, however, the neurons started working on a higher activity level, their excitability increased, and thus the number of neurons that responded to the applied stimulation with transmitter release increased accordingly.

Enhancer-sensitive neurons are always ready to immediately increase their activity in response to endogenous enhancer substances and represent the device in the mammalian brain that operates *de facto* as the *vis vitalis.* Any act in the endless "fight for existence" drama in nature is illustrative for the crucial importance of the enhancer regulation for survival. For example, an eagle pounces upon its chosen victim with lightning speed. To react accordingly is a matter of life and death. Both the attacker and the potential victim have only a split second to become properly activated. The chance for the eagle to obtain its food and for the victim to save its life lies in the mechanism that specific endogenous substances drive with proper speed the enhancer-sensitive neurons in the brain to reach the maximum level of performance. The partner with the more efficiently activated brain will reach its goal (26,35).

Significantly enhanced catecholaminergic/serotoninergic activity in the rat brain after weaning

In the rat, the enhancer regulation in the brain starts working at the discontinuation of breast feeding (end of the third week of age), which is the onset of the developmental ("uphill") phase of life. This period, characterized by a higher basic activity, lasts until the rat develops full sexual maturity (39). One of the tell-tale signs that makes the operation of the enhancer mechanism in the brain evident is the significantly enhanced release of catecholamines and serotonin from discrete brain regions after weaning (39). As an example, Fig. 2 illustrates that the amount of norepinephrine released from the locus coeruleus of male or female rats 1 week after weaning is significantly higher than that released 1 week before weaning.

Dampening of the Enhancer Regulation in the Rat Brain by Sexual Hormones

Further studies revealed that in both male and female rats the significantly enhanced brain release of catecholamines and serotonin, characteristic to the postweaning period, disappeared after sexual maturity was reached. In sexually mature rats the amounts of catecholamines and serotonin released from discrete brain regions did not differ significantly from the amounts released before weaning (39,50). Figure 3 illustrates that in sexually immature, 4-week-old male rats the release of norepinephrine from the locus coeruleus is significantly higher than in the sexually mature, 12-week-old rats. Similar age-related differences were measured in the release of dopamine from the substantia nigra and tuberculum olfactorium and the release of serotonin from the raphe (39).

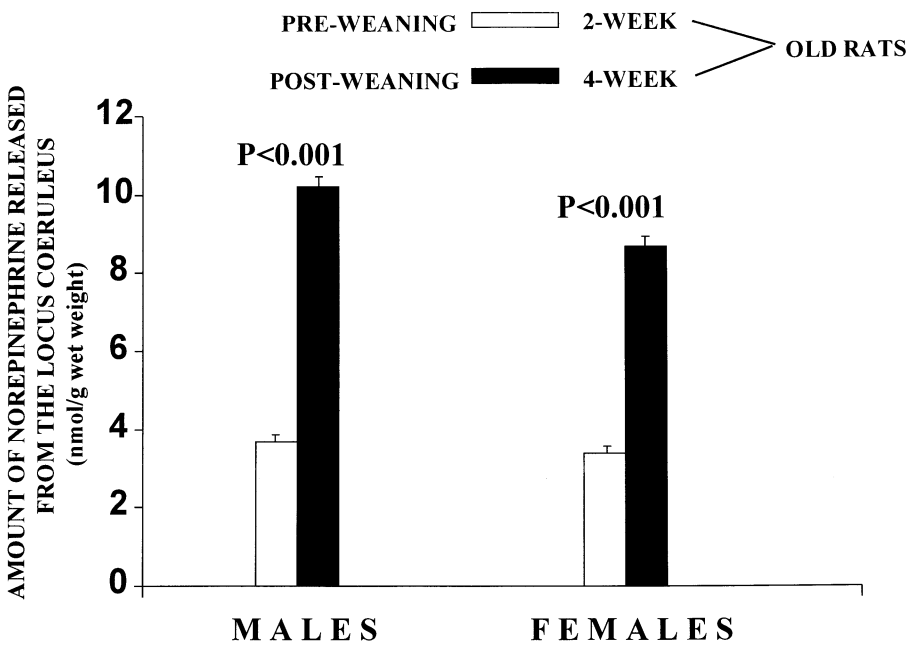


Fig. 2. The significantly enhanced release of norepinephrine from the locus coeruleus of male and female rats one week after weaning. $N = 12$. Vertical lines show S.E.M. Paired Student's t-test. See ref. 39 for methodology.

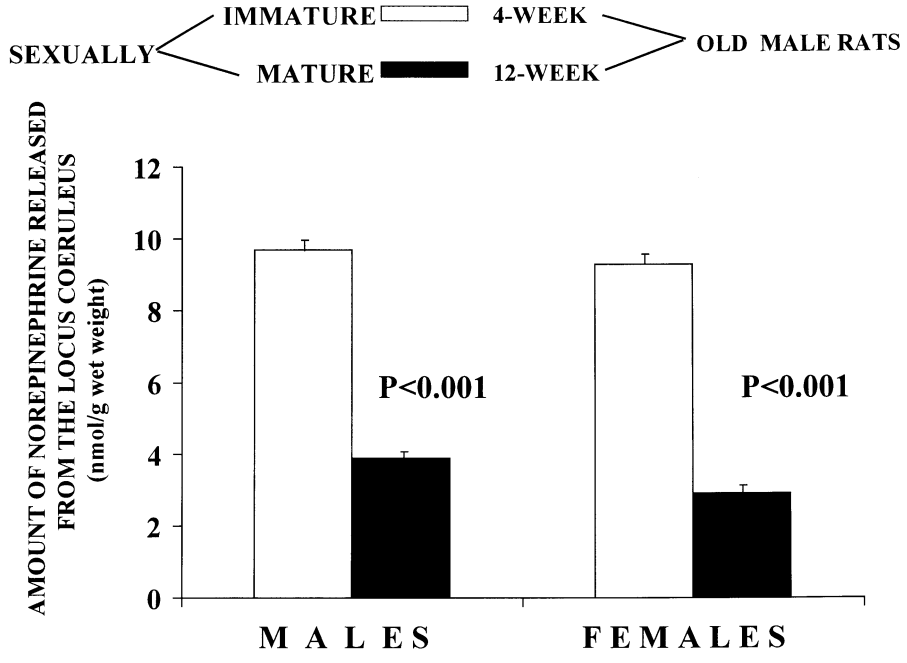


Fig. 3. The significantly dampened release of norepinephrine from the locus coeruleus of sexually mature male and female rats. $N = 12$. Vertical lines show S.E.M. Paired Student's t-test. See ref. 39 for methodology.

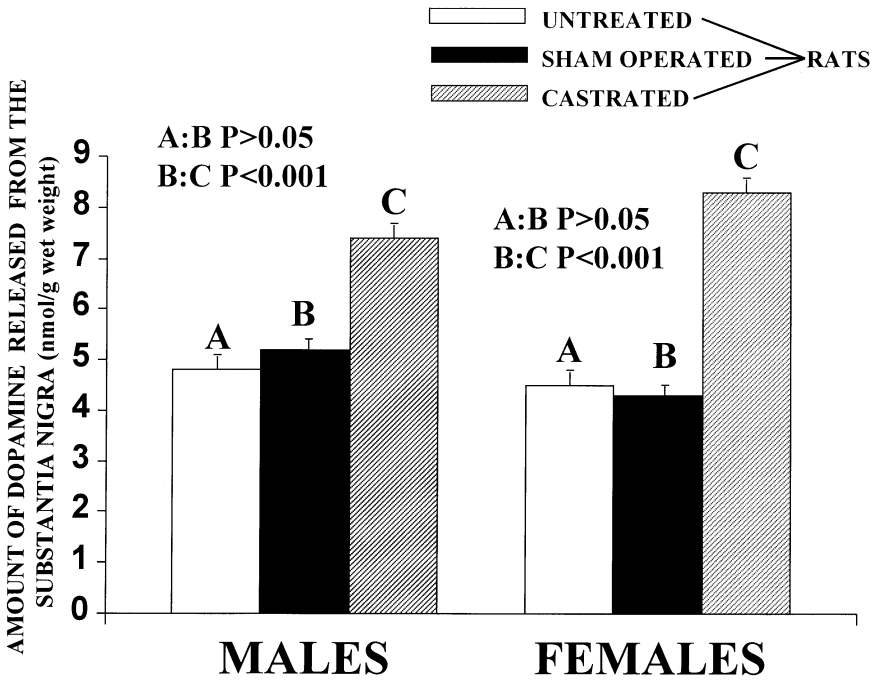


Fig. 4. The significantly enhanced release of dopamine from the substantia nigra of 3-months-old castrated male and female rats. The rats were castrated at the age of three weeks. $N = 16$. Vertical lines show S.E.M. Paired Student's *t*-test. See ref. 39 for methodology.

The detected age-related changes, shown for example in Fig. 3, clearly indicated that sexual hormones dampen the enhancer regulation. This was unequivocally proven by further analysis of this phenomenon (see ref. 50). The amount of dopamine released from the striatum, substantia nigra and tuberculum olfactorium, the amount of norepinephrine released from the locus coeruleus, and the amount of serotonin released from the raphe taken from 3-month-old castrated rats was significantly higher than the amount released from the same brain tissue samples isolated from untreated or sham-operated rats. The rats were operated at the completion of the third week of their life, which is the usual time of the discontinuation of breast feeding (50). For example, Fig. 4 shows that the amount of dopamine released from the substantia nigra of castrated 3-month-old male or female rats is significantly higher than that released from the substantia nigra of untreated or sham-operated rats.

The castration experiment already verified that sexual hormones dampen the enhancer regulation. Further experiments testing the effects of testosterone, estrone, and progesterone on the release of norepinephrine, dopamine, and serotonin from selected discrete brain regions furnished direct final evidence for this previously unknown central effect of the sexual hormones. Testosterone and estrone significantly inhibited the release of catecholamines and serotonin, but progesterone was ineffective. Unusually, estrone in male rats and testosterone in female rats was more effective in dampening the enhancer regulation in the brain (50).

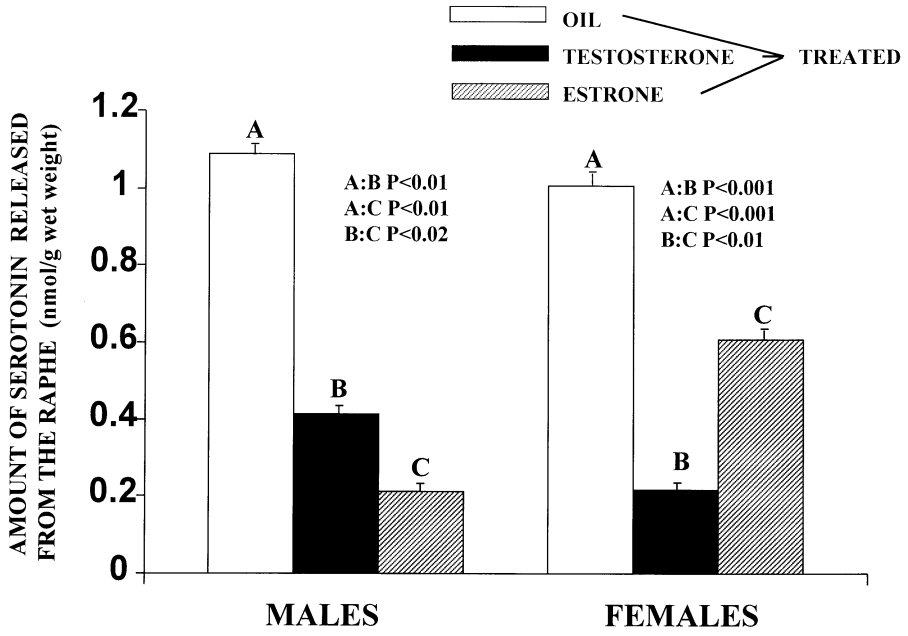
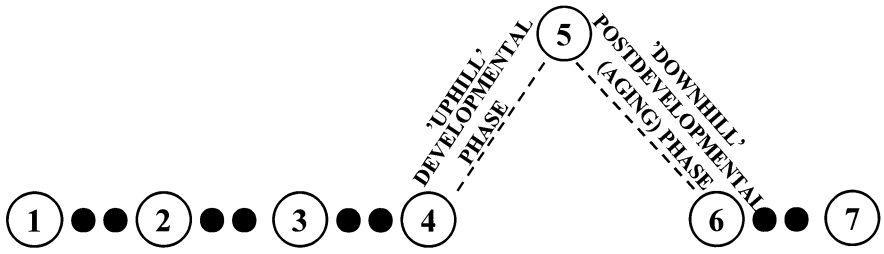


Fig. 5. The significant inhibition of the release of serotonin from the raphe of male and female rats injected s. c. once daily for 14 days with testosterone propionate (0.1 mg/rat) or estrone (0.01 mg/rat), respectively. Treatment started on 3-week-old rats. The raphe was isolated 24 h after the last injection. $N = 16$. Vertical lines show S.E.M. Paired Student's t-test. See ref. 39 for methodology.

As an example, Fig. 5 illustrates on the one hand that 2-weeks' treatment of 3-week-old male and female rats once daily with testosterone or estrone, respectively, significantly inhibited the release of serotonin from the raphe; on the other hand, estrone was significantly more effective than testosterone in the male rates ($P < 0.02$) and testosterone was significantly more effective than estrone in the female rates ($P < 0.01$).

Physiologic significance of the enhanced CAE/SAE activity during the developmental period of life

All in all, we detected two previously unnoticed mechanisms in the rat brain that seem to be a determinant for the beginning and the end of the uphill period of life. Figure 6 is a schematic illustration of the crucially important functional changes during the lifetime of mammals. The discontinuation of breast feeding is the onset of the developmental period of life and lasts until the goal of goals in nature, full scale sexual maturity, is reached. This is the most delightful phase of life, the glorious uphill journey. The individual progressively takes possession on a mature level of all abilities crucial for survival and maintenance of the species. The individual learns to avoid dangerous situations, masters the techniques to obtain its food, develops procreative powers for sexual reproduction, and copulates. This is also the time the climax of developmental longevity and the "downhill" post-developmental (aging) stage of life begins.



1) FUSION OF THE SPERMATOZOON WITH THE OVUM
2) THE INTEGRATIVE WORK OF THE CNS SETS IN. APPEARANCE OF EEG
3) BIRTH OF THE FOETUS
4) WEANING
5) SEXUAL MATURITY IS REACHED
6) THE INTEGRATIVE WORK OF THE CNS BLACKS OUT. DISAPPEARANCE OF EEG. 'NATURAL DEATH'
7) DEATH OF THE LAST CELL

Fig. 6. Conception about essential changes during the lifetime of mammals. For details see ref. 35.

Various species live together on earth in a harmonious proportion. This is obviously carefully regulated. One of the seemingly principal regulatory mechanisms that produces the balanced equilibrium among living organisms is brain aging which, in its final consequences, eliminates the individuals who already fullfilled their duty in nurturing the new generation.

Now we have to realize that the uphill period of life is hallmarked with the operation of the enhancer regulation that maintains the basic activity of the brain on a significantly higher level. The period of enhanced activity lasts until sexual hormones appear, dampen the enhancer regulation, and lower the basic activity of the brain to the preweaning level. Thus, sexual hormones care for the transition from the developmental phase of life into postdevelopmental longevity, the period of the slow age-related decay of brain performance and terminated by natural death.

This sequence of changes clearly accounts for the previously unexplained phenomenon first described by us in 1957 (25). We measured the hunger drive-induced intensive orienting-searching reflex activity of rats in a special variant of an open field. As it is well known, the intensity of the hunger drive-induced hypermotility depends on the time elapsed from the last feed. Figure 7 shows that food deprivation-induced hypermotility was substantially higher in rats at the peak of their uphill period of life than in rats in the early stage of their postdevelopmental phase of life. This phenomenon now has a logical interpretation. In the sexually immature younger group the enhancer regulation obviously worked unrestricted, while in the sexually mature elder rats sexual hormones already dampened this mechanism.

Although the slow and continuous age-related decline of the enhancer regulation (the *vis vitalis*) that is characteristic of the downhill, postdevelopmental phase of life starts

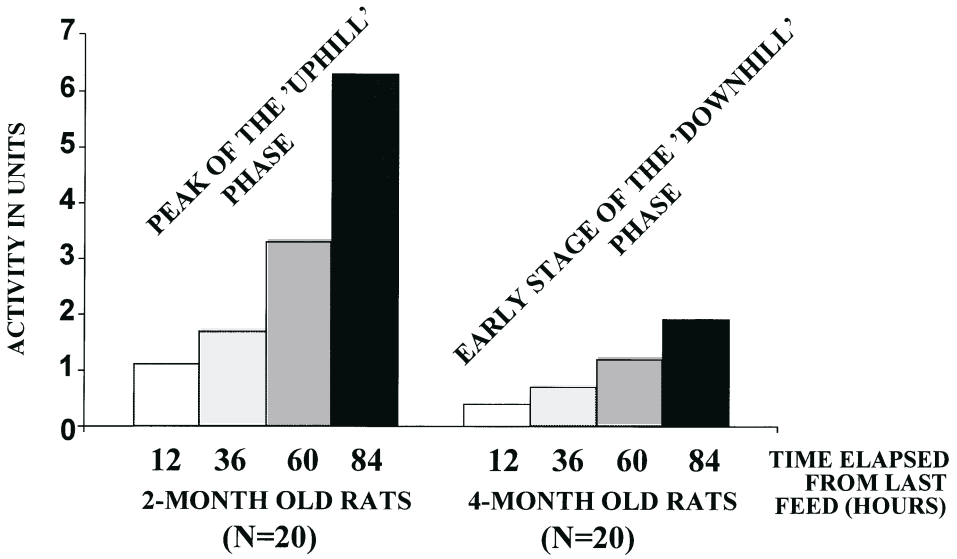


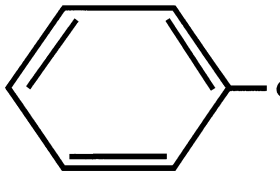
Fig. 7. Intensity of orienting-searching reflex activity of hungry rats in surroundings quite new to them as a function of time elapsed from last feed. Activity was measured and expressed in units from 0 to 10. See ref. 39 for methodology and other details.

with the full scale development of sexual hormonal regulation, it does not mean that the sexually mature individual is immediately converted to a significantly lower performer in its fight for existence. As it was shown earlier in detail, conditioning (learning) makes the performance of the experienced organism highly economic and efficient, even at a lower level of specific activation of the brain, (26). Nevertheless, the irresistible, progressive age-related decay of the enhancer regulation gradually weakens the compensatory role of experience, and even the most experienced aged organism becomes more and more vulnerable with the passing of time.

PEA-DERIVED ENHANCER SUBSTANCES

Reference Compounds: (–)Deprenyl and (–)1-Phenyl-2-propylaminopentane, (–)PPAP

(–)Deprenyl, which was the key experimental tool used to reveal the operation of the enhancer regulation in the brain (for review see ref. 37), is currently the only worldwide registered, clinically used drug with an enhancer effect. The compound was developed in the early 1960s and had, as the first selective inhibitor of MAO-B, a peculiar history before its primary important enhancer effect was discovered (for review see refs. 30 and 36).



	CH_2 - CH - N			'ENHANCER' EFFECT	RELEASING EFFECT	RELATION TO MAO
PHENYLETHYLAMINE (PEA)	H	H	H	+	+	SUBSTRATE TO MAO-B
AMPHETAMINE	CH_3	H	H	+	+	WEAK MAO INHIBITOR
METHAMPHETAMINE	CH_3	CH_3	H	+	+	WEAK MAO INHIBITOR
(-)1-PHENYL-2-METHYL-N-METHYL-PROPARGYL-AMINE, (-)DEPRENYL	CH_3	CH_3	C_3H_3	+	0	POTENT MAO-B INHIBITOR
(-)1-PHENYL-2-PROPYL-AMINOPENTANE, (-)PPAP	C_3H_7	H	C_3H_7	+	0	0

Fig. 8. The chemical structure and pharmacological spectrum of PEA and the PEA-derived, most representative enhancer substances.

(–)Deprenyl is a derivative of methamphetamine, a long-acting variant of PEA, one of the endogenous enhancer substances. PEA and the amphetamines are known to be potent releasers of the catecholaminergic transmitters from their pools, and in higher concentrations they are also releasers of serotonin. It now seems obvious that it was the releasing effect that hindered for decades the recognition of the primary important enhancer effect of PEA and its analogs (amphetamine, methamphetamine) with a long-lasting effect. (–)Deprenyl, the first amphetamine derivative devoid of the releasing property of its parent compound, allowed researchers to realize the operation of the enhancer mechanism and unmasked the enhancer effect of PEA and its derivatives (47,48).

(–)Deprenyl was originally developed as a new monoamine oxidase (MAO) inhibitor (40). It proved to be the first selective inhibitor of MAO-B (38) and became the worldwide experimental tool used to analyze this form of MAO. Recognizing that (–)deprenyl-induced activation of the nigrostriatal dopaminergic system is unrelated to the inhibition of MAO-B (32), we performed a structure-activity relationship study with the aim of developing deprenyl analogues that, on the one hand, are free of the MAO inhibitory property and, on the other hand, are, in contrast to deprenyl, not metabolized to amphetamines (44). (–)PPAP was selected as a reference substance for further studies. Although (–)PPAP was the first PEA-derived enhancer substance free of the unwanted effects of (–)deprenyl, its clinical efficiency was, in spite of all our efforts, never tested. Figure 8 shows the chemical structure and pharmacologic spectrum of the most important PEA-derived substances that have an enhancer effect.

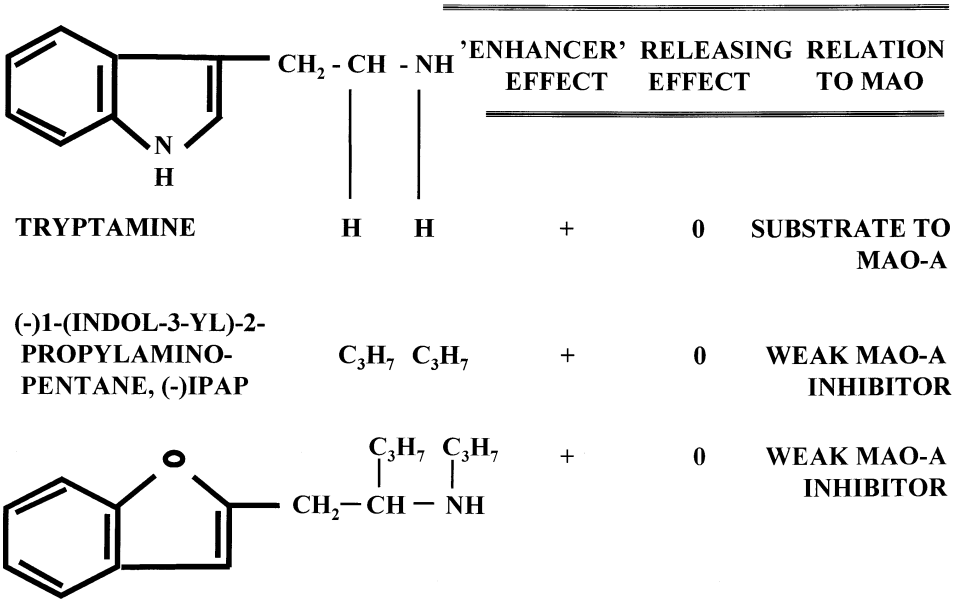


Fig. 9. The chemical structure and pharmacological spectrum of tryptamine and the tryptamine-derived, most representative enhancer substances.

TRYPTAMINE DERIVED ENHANCER SUBSTANCES

Reference Compound: (–)1-(Benzofuran-2-yl)-2-propylaminopentane [(–)BPAP]

The discovery that tryptamine is an endogenous enhancer substance (35) opened the way for a structure-activity relationship study aiming to develop a family of enhancer compounds unrelated to PEA and the amphetamines. Of the newly synthetized compounds, (–)1-(benzofuran-2-yl)-2-propylaminopentane [(–)BPAP] was selected for use in the future as: (i) the reference substance for the analysis of the enhancer mechanism in the mammalian brain; (ii) a therapeutic agent in age-related depression, Parkinson's disease, and Alzheimer's disease; and (iii) a prophylactic agent to slow the physiological aging of the brain in the healthy population (49). Figure 9 shows the chemical structure of the two most potent, tryptamine-derived selective enhancer substances.

Antagonism of tetrabenazine-induced depression by (–)BPAP

Because of the crucial importance of brain catecholamines in learning, the drastic reduction of the catecholamine stores in the brain of rats treated with tetrabenazine almost completely inhibits the acquisition of conditioned avoidance responses (CARs) in the shuttle box. Even though the avoidance response to the unconditioned stimulus is im-

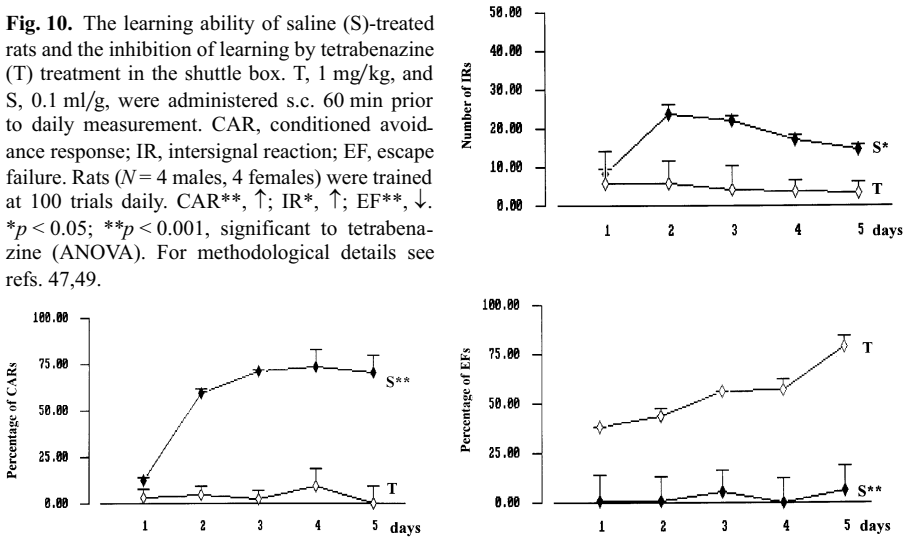


Fig. 10. The learning ability of saline (S)-treated rats and the inhibition of learning by tetrabenazine (T) treatment in the shuttle box. T, 1 mg/kg, and S, 0.1 ml/g, were administered s.c. 60 min prior to daily measurement. CAR, conditioned avoidance response; IR, intersignal reaction; EF, escape failure. Rats ($N = 4$ males, 4 females) were trained at 100 trials daily. CAR**, ↑; IR*, ↑; EF**, ↓. *$p < 0.05$; **$p < 0.001$, significant to tetrabenazine (ANOVA). For methodological details see refs. 47,49.

paired, the number of escape failures (EFs) is significantly increased. The peculiar depression of learning by tetrabenazine is illustrated in Fig. 10.

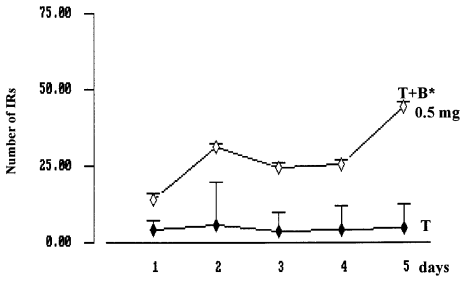
Substances that enhance the impulse-propagation-mediated release of catecholamines in the brain fully antagonize tetrabenazine-induced depression in rats. Figure 11 shows, as an example, the prevention of tetrabenazine-induced learning depression in the shuttle box by the simultaneous administration of (–)BPAP. In antagonizing the effect of tetrabenazine in the shuttle box, (–)BPAP was found to be 130 times more potent than (–)deprenyl (49).

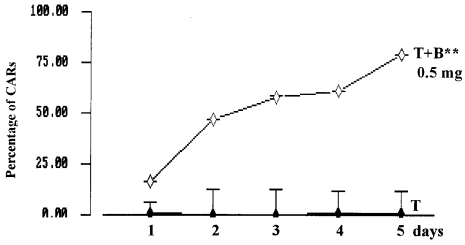
Enhancement of the impulse-propagation-mediated release of catecholamines and serotonin in the brain by (–)BPAP

The isolated rat brain stem is a sensitive test for the rapid ex vivo detection of the enhancer property of a substance. Figure 12 demonstrates that (–)BPAP is a highly potent enhancer of the electrical stimulation induced release of [^{3}H]norepinephrine, [^{3}H]dopamine, and [^{3}H]serotonin from the isolated brain stem of rats.

The measurement of the *in vivo* enhancer effect of a compound is based on the finding that, in rats treated with an enhancer substance, the catecholaminergic/serotoninergic neurons in the brain work on a higher activity level and this enhancer effect can be detected in the most simple manner. We measured the amount of norepinephrine, dopamine, and serotonin, respectively, released within a 20-min period from isolated discrete brain regions. Pretreatment of rats with 0.0001 mg/kg of (–)BPAP was already sufficient to significantly enhance the release of dopamine from the substantia nigra and the tuberculum olfactorium, norepinephrine from the locus coeruleus, and serotonin from the raphe (see Table 2 in ref. 49).

The enhancer effect of a substance can be detected in a dose-dependent manner also by adding the compound to the isolated discrete brain region containing the neurocytes of the enhancer-sensitive neuron. Figure 13 illustrates, as an example, the effect of a series of concentrations of (–)BPAP on the release of norepinephrine when given to the isolated





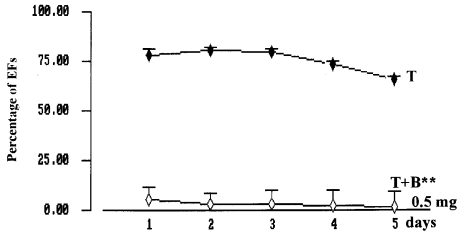


Fig. 11. Antagonistic effect of (–)BPAP (B) in rats against tetrabenazine (T)-induced depression of learning in the shuttle box. T, 1 mg/kg, or B, 0.5 mg/kg, + T were administered s.c. 60 min prior to daily measurement. For details see Fig. 10. CAR, conditioned avoidance response; IR, intersignal reaction; EF, escape failure. Rats ($N = 4$ males, 4 females) were trained at 100 trials daily. CAR**, ↑; IR*, ↑; EF**, ↓. *$p < 0.05$; **$p < 0.001$, significant to tetrabenazine (ANOVA). For methodological details see refs. 47,49.

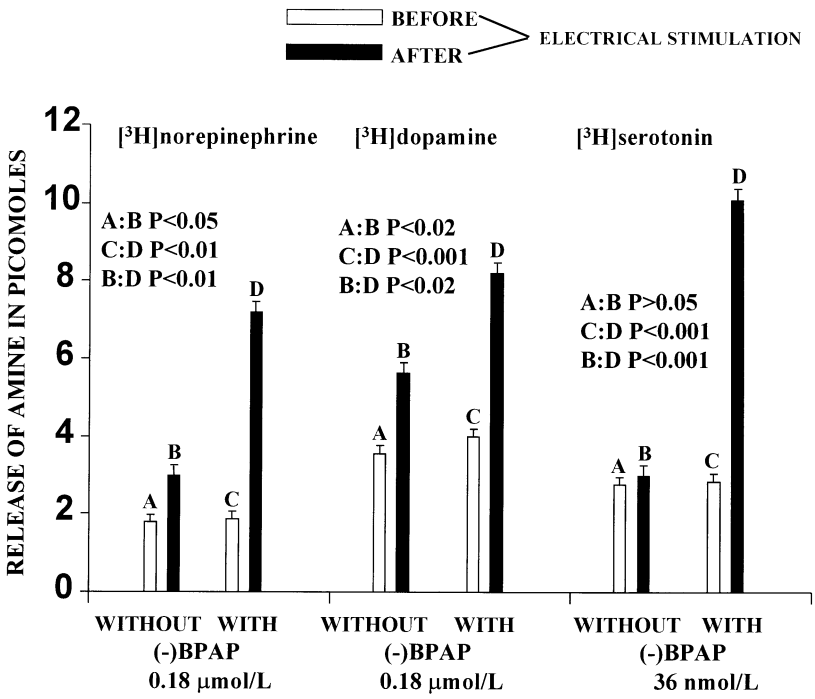


Fig. 12. The significant enhancement of the electrical stimulation induced release of [^{3}H]norepinephrine, [^{3}H]dopamine, [^{3}H]serotonin, respectively, from isolated rat brain stem in the presence of (–)BPAP. Each column represents the amount of labelled amine in picomoles released in a 3-min collection period. $N = 8$. Vertical lines show S.E.M. Paired Student's t-test. See refs. 48 and 49 for methodology.

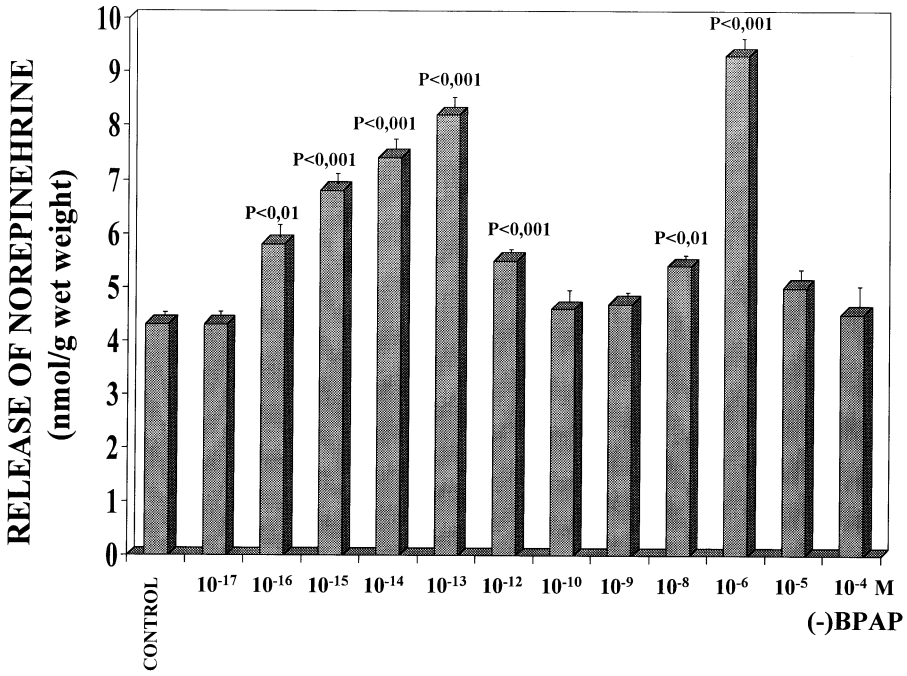


Fig. 13. The peculiar concentration dependency of the enhancer effect of (–)BPAP when added to the quickly excised locus coeruleus of male rats. Vertical lines show S.E.M. Paired Student's *t*-test. See refs. 39 and 49 for methodology.

locus coeruleus of rats. The figure demonstrates the peculiar concentration dependency of the enhancer effect of (–)BPAP. There is a highly significant increase in the amount of norepinephrine released from the tissue in the low concentration range (with a peak of 10^{-14} M) and a second one in the high concentration range (with a peak of 10^{-6} M). This behavior of the enhancer substance clearly indicates, on the one hand, the presence of highly specific enhancer receptors for binding of (–)BPAP and, on the other hand, the existence of an unusually complicated cell device for the accomodation of an enhancer substance.

The neuroprotective effect of (–)BPAP and the effect of enhancer substances on other than catecholaminergic/serotoninergic neurons in the brain

For practical reasons, we measured the effect of the enhancer substances mainly on the catecholaminergic/serotoninergic neurons, but many other groups of enhancer-sensitive neurons exist in the brain. We know this from series of experiments performed with (–)deprenyl on cultured neurons, demonstrating the neuroprotective, antiapoptotic, trophic, etc., effects of the drug. It was concluded in 1998 that all these effects of (–)deprenyl, detected on different types of cultured neurons, relate to the enhancer property of the drug (37). Final support for this conclusion was given recently by demonstrating the neuroprotective effect of racemic BPAP on primary embryonic hippocampal cultures (49).

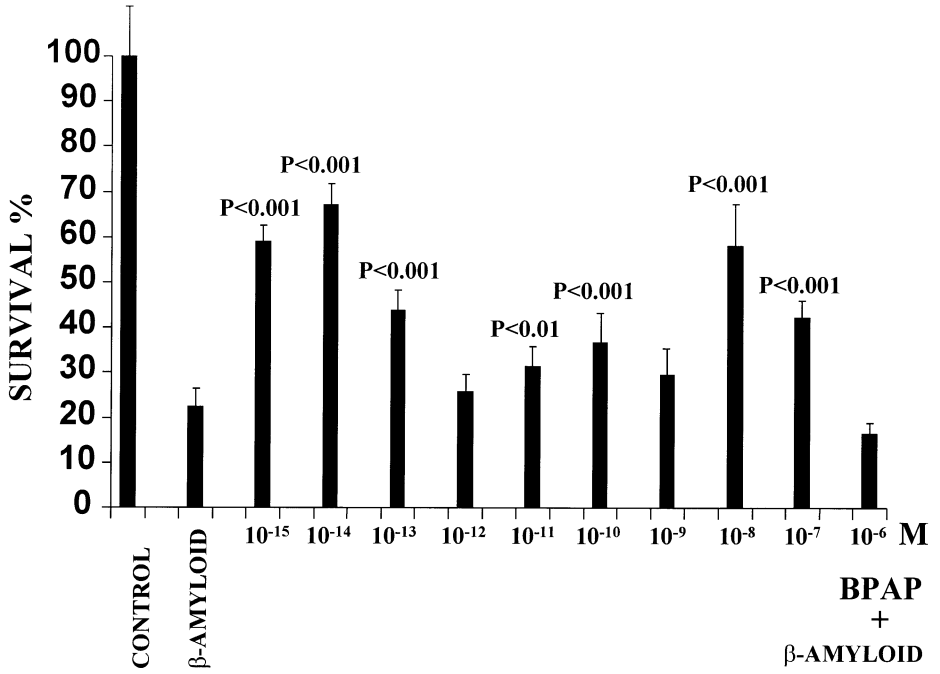


Fig. 14. The protective effect of racemic BPAP against β-amyloid induced neurotoxicity on cultured rat hippocampal neurons. Vertical lines show S.D. Statistics: Dunnet's *t*-test. See ref. 49 for methodology. Note that the neuroprotective effect of BPAP on cultured neurons and the enhancer effect of (–)BPAP on the locus coeruleus (see Fig. 13) showed essentially similar concentration dependency.

At day 10 the cultured cells were injured by exposing them for 3 days to a 20 μM concentration of β-amyloid 25–35 fragment. This concentration of β-amyloid decreased the survival of the neurons (control = 100%) to 22.4 ± 7.20% (mean ± S.D.). Figure 14 shows that BPAP significantly inhibited the β-amyloid-induced neurotoxicity in the cultured hippocampal neurons in two distinct ranges of concentration, one with a peak of 10^{-14} M and one with a peak of 10^{-8} M. The peculiar concentration dependency of the effect of BPAP on the hippocampal neurons is surprisingly identical with that on noradrenergic neurons (see Fig. 13), showing that the cell device in service of the enhancer regulation might be very similar to each other in the different enhancer-sensitive neurons.

As a matter of fact, there is a conspicuous essential similarity between the BPAP-induced effect on the cultured rat hippocampal neurons and the one induced by (–)deprenyl in rats treated with the drug for years during their postdevelopmental phase of life, which is described in more detail in the introduction of this review. In the (–)deprenyl experiment, we picked out of a population of 1600 rats, the animals with the lowest and highest sexual performance, and demonstrated that the HP rats were also better performers in the shuttle box and lived significantly longer than their LP peers and that (–)deprenyl treatment transformed the LP rats into significantly higher performing ones that lived as long as their saline-treated HP peers (46). We assumed that HP, longer-living rats possess a more efficient catecholaminergic brain engine than the LP, shorter-living rats and treatment with (–)deprenyl, an enhancer substance, acts accordingly. Whatever perfor-

mance is measured in a random population, the individual variation in performance is always considerably high. About 20% of cultured hippocampal neurons that survived the β-amyloid attack, as the most active HP members of the population possessing the most efficient enhancer regulation, successfully mobilized their resources against the attacker. In the presence of 10^{-14} M BPAP each neuron started working on a higher activity level; hence, the surviving rate increased from 20% to 70%.

The peculiar pharmacologic profile of (–)BPAP and the fact that the compound exerts its enhancer effect in a concentration as low as 10^{-14}–10^{-15} M speaks strongly in favor for the assumption that much more potent endogenous enhancer substance(s), in addition to PEA or tryptamine, may exist in the mammalian brain.

Because of the variety of enhancer-sensitive neurons, (–)deprenyl and (–)BPAP necessarily change various brain functions. The enhanced activity of the catecholaminergic system in the brain has by itself far reaching functional consequences. The finding in 1988 that the scavenger function of the nigrostriatal dopaminergic neurons is significantly enhanced in (–)deprenyl-treated rats (31), this effect is unrelated to MAO inhibition (33) and is also unrelated to a direct effect on the scavenger enzymes in general (32), was the first proof that a previously unknown, peculiar "new" effect of (–)deprenyl is just an expression of the drug-induced increased activity of a special group of enhancer-sensitive neurons in the brain (for review see ref. 37).

Recently there has been a rapid increase in the number of papers analyzing the so-called neuroprotective effect of (–)deprenyl under different experimental conditions. To mention just one of the more recent examples of the dozens of papers describing essentially the same phenomenon, Suuronen et al. (74) described that (–)deprenyl offered significant protection against the apoptotic response induced by okadaic acid in cultured hippocampal neurons, cerebral granule neurons, and Neuro-2a neuroblastoma cells and provided protection against apoptosis after cytosine-β-D-arabinoside treatment of hippocampal neurons and Neuro-2a neuroblastoma cells, as well as after etoposide treatment in Neuro-2a cells (74).

The number of papers describing an unexpected new effect of (–)deprenyl is increasing. The antitumor effect of this drug due to the enhanced activity of the tuberoinfundibular dopaminergic (TIDA) neurons in the medial basal hypothalamus is a remarkable example. It was found by ThyagaRajan et al. (79) that prolonged treatment of old acyclic female rats with (–)deprenyl decreased the incidence of mammary and pituitary tumors by augmenting hypothalamic dopaminergic activity. They also found that treatment of rats with (–)deprenyl following the development of DMBA-induced mammary tumors prevented tumor growth and decreased the tumor number (80). There is no reason to review each of the hundreds of papers describing the various outward appearance forms of the effects of (–)deprenyl on cultured neurons, denoting these as neuroprotective, antiapoptotic, antitumor, etc., because all of them are due to the drug-induced enhanced activity of enhancer-sensitive neurons.

A recently published paper by Groc et al. (21) presented evidence of (–)deprenyl-insensitive apoptosis of nigral neurons during development. The authors demonstrated that apoptotic death of dopamine neurons during development is insensitive to daily treatment of the pregnant mothers and then newborns with (–)deprenyl 0.1, 1, or 10 mg/kg. This finding deserves attention in the light that the enhancer mechanism starts working only after discontinuation of breast feeding (see Fig. 2).

The neurons of the catecholaminergic/serotoninergic system in the brain are probably the physiologically, biochemically, and pharmacologically best studied constituents of the CNS, and we have highly sensitive methods for the rapid and exact measurement of the electrical stimulation-induced activation of these neurons. Hence, an enhancer substance-induced activation of these neurons is easily detectable by the enhanced impulse-evoked release of norepinephrine, dopamine, and serotonin, respectively (see for example Figs. 1 and 12). It is at the present time more complicated to detect the activation of other enhancer sensitive groups of neurons in the brain. To date the enhancer effect was measured on cultured cells. In a random population of properly, freshly prepared cultured neurons we find the usual distribution between high-, medium-, and low-performing neurons and apoptotic cells. As time passes the performance of the neurons progressively decreases until the cells die out. Addition of an enhancer substance to an enhancer-sensitive population of cultured neurons stimulates performance, slows the degradation process, and prolongs the life of the cell ("antiapoptotic" effect). In the more active, better-performing neurons, the basic mechanisms of cell life obviously operate on a higher activity level. For example, Tatton (76) found that the rate of new protein synthesis was enhanced and the mitochondrial membrane potential was better maintained in neurons treated with (-)deprenyl.

The main aims of current research to clarify the enhancer regulation in the brain are as follows: (i) to show that (–)BPAP binds to specific receptors by selecting BPAP analogues with high affinity to the receptors but with significantly lower specific activity than (–)BPAP, which will then work as competitive antagonists; (ii) to identify the specific macromolecular target(s) that accommodate the endogenous enhancer substances and their synthetic derivatives; (iii) to demonstrate the existence of the predicted endogenous enhancer substance(s) that are much more potent than PEA or tryptamine; and (iv) to map the enhancer-sensitive neurons in the brain.

ENHANCER SUBSTANCES AS PROPHYLACTIC ANTIAGING DRUGS AND THERAPEUTIC AGENTS IN AGE-RELATED NEUROLOGICAL DISEASES

Rationale

History of the development of (–)deprenyl

As Figure 8 shows, (–)deprenyl differs from its parent compound methamphetamine in that it contains a relatively bulky substituent (a propargyl group) attached to the nitrogen. This chemical change eliminated the compound's releasing property, while its enhancer effect survived. This change in the pharmacologic spectrum made the discovery of its enhancer regulation possible. The releasing property masked for decades the enhancer effect in PEA and the amphetamines, the parent compounds of (–)deprenyl. The original idea of the structure-activity relationship study, leading in 1965 to the selection of the compound later named deprenyl, was to elaborate a new type of psychostimulant that is also a potent MAO inhibitor (see ref. 40). We knew from the history of pargyline that the attachment of a propargyl group to the nitrogen in benzylamine yielded a highly potent

MAO inhibitor and, as expected, we had the same result using methamphetamine as the starting material. The propargyl group binds covalently with the flavin in MAO, leading to the irreversible inhibition of the enzyme.

It was a surprisingly lucky, unexpected finding that (–)deprenyl inhibited MAO-B with high selectivity (38). (–)Deprenyl was used thereafter as a specific experimental tool to analyze MAO-B. Our first paper that describes this novel property has become a citation classic. For several years the selective MAO-B inhibitory effect was at the center of our interest and delayed the discovery of the drug's enhancer effect. It was the MAO inhibitory effect of the compound that led to the first clinical application of (–)deprenyl.

In light of the serious side effects of levodopa in Parkinson's disease, Birkmayer and Hornykiewicz (4) tried in 1962 to achieve a levodopa-sparing effect by concurrent administration with an MAO inhibitor. The blockade of the enzyme however potentiates the catecholamine releasing property of various indirectly acting amines ("cheese" effect). The precipitation of hypertensive attacks also followed the concurrent administration of levodopa with an MAO inhibitor, terminating this line of clinical research. As we have already shown in 1968, (–)deprenyl is a unique MAO inhibitor that does not potentiate the catecholamine releasing effect of tyramine, i.e., the substance is free of the cheese effect (41). Birkmayer et al. (6) combined (–)deprenyl with levodopa in Parkinson's disease and published in 1977 that the trial was successful; the levodopa-sparing effect was achieved without signs of significant hypertensive reactions. The study initiated worldwide use of (–)deprenyl in Parkinson's disease. Today the most evaluated effect of the drug is its ability to slow the progress of the disease in de novo parkinsonians. This beneficial effect of (–)deprenyl is due to its enhancer property and is unrelated to the inhibition of MAO-B (for review see ref. 37).

We showed that (–)deprenyl is free of the cheese effect in rat, which prompted and justified the Birkmayer trial, and was corroborated in man by Sandler et al. (14,66) in two studies in 1978. As a matter of fact, Varga (41), inspired by our finding that E-250 (the original code name for the racemic compound named later deprenyl) was free of the tyramine potentiating effect of the MAO inhibitors (41), was the first to demonstrate in 1968 that provocative cheese consumption failed to provoke headache or hypertensive reactions in volunteers treated with E-250. However, Varga's results (quoted in the discussion of ref. 41) remained unpublished.

The place of an enhancer drug in depression

(–)Deprenyl was found to be a potent antidepressant. This effect was originally demonstrated by Varga et al. (81,82) with the racemic compound in 1965–1967 and in 1971 with the (–)enantiomer (78), and was first corroborated by Mann and Gershon (55) in 1980. The realization of the peculiar effect of (–)deprenyl, first in Parkinson's disease and later in Alzheimer's disease, distracted attention from its antidepressant property, which remained unutilized. Unfortunately, (–)deprenyl was not registered anywhere for depression. Even an especially interesting aspect of this problem fell into oblivion. In a study on 102 outpatients and 53 inpatients (–)deprenyl was given together with (–)phenylalanine by Birkmayer et al. (7) in 1984 and it was shown that nearly 70% of the patients achieved full remission. This outstanding clinical efficiency equaled only that of electroconvulsive treatments (ECT), but without the memory-loss side effect of ECT. Making use of the promising antidepressant effect of this highly potent and selective enhancer substance will

be a challenge since (–)BPAP is about 130 times more potent than (–)deprenyl in rats for antagonizing tertrabenazine-induced depression in the shuttle box.

The Place of an Enhancer Drug in Parkinson's Disease

The neostriatum is the main input structure of the basal ganglia. We may say that a more active nigrostriatal dopaminergic system means a more active cerebral cortex and vice versa. The physiological age-related morphological and functional deterioration of the nigrostriatal dopaminergic neurons leads to an equivalent decay of cortical activity with the passing of time (for details see ref. 37).

It is reasonable to conclude that the age-related decline of the nigrostriatal dopaminergic brain mechanism plays a significant role in the progressive decline of performance in the aged. Aging of the dopaminergic system in the brain plays an undisputable leading role in the highly significant, substantial decline in male sexual activity and also in the more modest but still significant age-related decline in learning performance. In a human male study the median coital activity was the highest, 2.1/week, between ages 30–34 and decreased progressively with increasing age, sinking to 0.2/week ($P < 0.001$) in the 65–69-year-old age group (56). We found essentially the same trend of changes in male rats in different series of experiments (31,42,43,46).

We measured the age-related decay of sexual performance in male rats by selecting the best performing individuals from a large population and copulatory activity was tested once weekly during three consecutive 36-week periods. In a group of 49 rats, an average of 14.04 ± 0.56 ejaculations were displayed during the first 36-week period, dropping to 2.47 ± 0.23 ($P < 0.001$) during the third 36-week period. Also the learning performance of the same rats, tested in the shuttle box, declined significantly with age. The total number of conditioned avoidance responses displayed during the first 36-week period was 78.45 ± 3.11 and decreased to 50.67 ± 2.99 ($P < 0.001$) during the third 36-week period (46).

There is a quantitative difference only between the physiological age-related decline of the dopaminergic input and that observed in Parkinson's disease. In the healthy population the calculated loss of striatal dopamine is about 40% at the age of 75, which is about the average lifetime; the loss of dopamine in Parkinson's disease is 70% or thereabout at diagnosis and over 90% at death. The drastic reduction of the dopaminergic output in Parkinson's disease leads evidently to an accordingly drastic reduction of cortical activity (37) and this makes it understandable why an enhancer substance, like (–)deprenyl, improves cognition, attention, memory, and reaction times and brings about subjective feelings of increased vitality, euphoria, and increased energy in people with Parkinson's disease (52).

When diagnosing Parkinson's disease neurologists select subjects with the most rapidly aging striatal dopaminergic system (about 0.1% of the population). As symptoms of Parkinson's disease become visible only after the unnoticed loss of a major part (about 70%) of striatal dopamine and further deterioration is persistent, the disease is, in essense, incurable. Prevention is the only chance to fight off Parkinson's disease. We need to start slowing the age-related functional decline of the striatal dopaminergic neurons in due time. For this reason it is advisable to begin the prophylactic administration of an enhancer substance, for example (–)deprenyl, 1 mg/day, as soon as sexual maturity is reached and the postdevelopmental period of life has just started.

It has been shown in a series of experiments performed with (–)deprenyl (for review see ref. 36) that an enhancer substance protects the nigrostriatal dopaminergic neurons against selectively acting neurotoxins (13,18,22,28,83,84), facilitates scavenger function in the striatum (11,12,31,33), and prevents age-related changes in the neurocytes of the substantia nigra in rats (45) and in man (68).

(–)Deprenyl was convincingly shown to be capable of slowing the rate of the functional deterioration of the nigrostriatal dopaminergic neurons in patients with early, untreated Parkinson's disease. Age-related deterioration of the striatal machinery is a continuum and any precisely determined short segment of it is sufficient to measure the rate of decline in the presence or absence of (–)deprenyl. As a matter of fact, in the multicenter DATATOP study of the Parkinson Study Group, a segment of this continuum, the time elapsing from diagnosis of Parkinson's disease until levodopa was needed, was properly measured in untreated patients with Parkinson's disease and the effect of (–)deprenyl versus placebo was compared (61). It was first published by Tetrud and Langston (77), participants of the DATATOP study, that (–)deprenyl delayed the need for levodopa therapy. In their study, the average time until levodopa was needed was 312.1 days for patients in the placebo group and 548.9 days for patients in the (–)deprenyl group. This was clear proof that (–)deprenyl, which enhanced the activity of the surviving dopaminergic neurons, kept these neurons on a higher activity level for a longer duration of time.

The design of the DATATOP study was unintentionally the same that we have used in rat experiments with (–)deprenyl since 1980. We selected male sexual activity as a quantitatively measurable rapidly aging dopaminergic function, compared the effect of (–)deprenyl versus saline treatment on the age-related decline of this function, and demonstrated that (–)deprenyl treatment significantly slowed the age-related decay of sexual performance (29). This effect of (–)deprenyl is unrelated to the inhibition of MAO-B, as (–)PPAP, a derivative of (–)deprenyl that is free of MAO-B inhibitory property (44), enhances dopaminergic activity in the brain like (–)deprenyl (for review see ref. 36). By now it is clear that if we select a quantitatively measurable dopaminergic function and measure its age-related decline by fixing an exact endpoint, a shift from this endpoint in time in (–)deprenyl-treated rats shows the dopaminergic activity enhancer effect of the drug. For example, male rats finally lose their ability to ejaculate due to the physiological aging of the striatal dopaminergic system. We found that saline-treated rats reached this endpoint at the age of 112 ± 9 weeks, whereas their (–)deprenyl-treated peers lost the ability to ejaculate only at the age of 150 ± 12 weeks ($P < 0.001$) (34). The design of the DATATOP study was essentially the same. The authors knew that after diagnosis of Parkinson's disease the next stage downward is the need for levodopa, and they measured the (–)deprenyl-induced delay in reaching this endpoint.

The authors expected (–)deprenyl to be efficient in their trial because of its MAO-B inhibitory effect. Their hypothesis was that the activity of MAO and the formation of free radicals predispose patients to nigral degeneration and contribute to the emergence and progression of Parkinson's disease; thus, the MAO inhibitor (–)deprenyl, the antioxidant α-tocopherol, and the combination of the two compounds will slow the clinical progression of the disease to the extent that MAO activity and the formation of oxygen radicals contribute to the pathogenesis of nigral degeneration. They selected patients with early, untreated Parkinson's disease and measured the delay of the onset of disability necessitating levodopa therapy. In the first part of the trial 401, subjects were assigned to α-tocopherol or placebo and 399 subjects were assigned to (–)deprenyl alone or in combi-

nation with α-tocopherol. Only 97 subject who received (–)deprenyl reached endpoint during an average 12 months of follow up compared with 176 subjects who did not receive (–)deprenyl. The risk of reaching the endpoint was reduced by 57% for subject who received (–)deprenyl and these patients also had a significant reduction in the risk of having to give up full-time employment (61). Following the fate of the patients, they corroborated in their next paper (62) that (–)deprenyl, but not α-tocopherol, delayed the onset of disability associated with early, otherwise untreated Parkinson's disease. But as time passed, the DATATOP study also revealed that (–)deprenyl did not reduce the occurence of subsequent levodopa-associated adverse effects in the patients (63). A comparison of the enhancer effect of α-tocopherol with that of (–)deprenyl revealed that α-tocopherol is devoid of an enhancer effect; it does not change the impulse-evoked release of norepinephrine, dopamine and serotonin in the brain (unpublished results).

Although Tetrud and Langston and other authors of the DATATOP study were not aware of the dopaminergic activity enhancer effect of (–)deprenyl, their trial was the first to give convincing evidence that (–)deprenyl keeps the nigrostriatal dopaminergic neurons on a higher activity level not only in the rat but also in humans. In addition, this effect of (–)deprenyl was detected in a selected human population with the lowest striatal dopaminergic activity. The highly significant effect of (–)deprenyl and the ineffectiveness of α-tocopherol during the first years of the DATATOP study was clear proof that the drug acted by enhancing the activity of the nigrostriatal dopaminergic neurons. The patients selected for the study with early, untreated Parkinson's disease were ideal for demonstrating this effect. The subjects still had a sufficient number of dopaminergic neurons, the activity of which could be enhanced by (–)deprenyl; thus, the need for levodopa therapy was delayed.

α-Tocopherol, devoid of a dopaminergic activity enhancer effect, remained ineffective. As Parkinson's disease is incurable, drug effects are necessarily transient in nature. It is obvious that parallel with further decay of the striatal dopaminergic system, the responsiveness of the patients toward (–)deprenyl decreased with the passing of time (63).

Birkmayer et al. (6) used a (–)deprenyl 10 mg/day oral dose, calculated by us on the basis of the MAO inhibitory potency of the racemic compound during the mid-1960s, for the first clinical trial carried out by Varga in 1965 (81). Because the daily administration of (–)deprenyl 10 mg completely inhibits MAO-B in human platelets and the brain, this was from the beginning and is still the clinical (–)deprenyl dose used worldwide. Due to inhibiton of MAO-B, (–)deprenyl treatment allows for a 20–50% decrease in the levodopa dose needed in Parkinson's disease.

The DATATOP study (1989) in the United States (61), the French Selegiline Multicenter Trial (FSMP) (1991) (2), and the Finnish study (1992) (60), all multicenter studies that used (–)deprenyl as initial treatment in *de novo* patients with Parkinson's disease, revealed the safety of the long-term administration of (–)deprenyl 10 mg/day. It is commonly assumed that (–)deprenyl by itself is an exceptionally safe compound. In patients who need levodopa, however, there is always a risk that the administration of (–)deprenyl will enhance the side effects of levodopa, which can only be avoided by properly decreasing the levodopa dose according to the individual sensitivity of the patient. An example of a multicenter clinical trial, in which the improper combination of levodopa with (–)deprenyl led to confusion and misinterpretation, is the one performed by the Parkinson's Disease Research Group of the United Kingdom and published in 1995 (53; for a critical analysis see ref. 37).

The Norwegian-Danish Study Group (1999) published their results from a 5-year randomized, placebo-controlled, double-blind study in patients with early Parkinson's disease (51). They concluded that patients treated with a combination of (–)deprenyl and levodopa developed markedly less severe parkinsonism and required lower doses of levodopa during the 5-year study period than patients treated with levodopa and placebo. There was no trend towards worsening during the washout period among patients treated with (–)deprenyl. This recent study further supports the conclusion that (–)deprenyl slows the progression of early Parkinson's disease.

There are several promising opportunities in the treatment of Parkinson's disease: to slow the progress of the disease and shift the time until levodopa is needed in *de novo* patients with Parkinson's disease with (–)BPAP, a much more potent and selective enhancer substance than (–)deprenyl; to administer a carefully adjusted dose of (–)deprenyl when levodopa is already needed; and to make safe use of the levodopa-sparing effect of (–)deprenyl.

The place of an enhancer drug in Alzheimer's disease

The first two studies to demonstrate the beneficial effect of (–)deprenyl in Alzheimer's disease were published in 1987 (57,75), and a series of clinical studies with small sample sizes confirmed thereafter the usefulness of this drug in the treatment of the disease (1,9, 10,15–17,20,54,59,65,72). In some of these studies the effect of (–)deprenyl was compared with other drugs. Campi et al. (10) found (–)deprenyl to be more effective than acetyl-L-carnitine. According to Falsaperla et al. (15), (–)deprenyl is more effective than oxiracetam (a piracetam-like nootropic drug) in improving higher cognitive functions and reducing impairment of daily living. In a study by Monteverde et al. (59) (–)deprenyl proved to be more effective than phosphatidylserine.

The rationale and design of the first multicenter study of (–)deprenyl in the treatment of Alzheimer's disease using novel clinical outcomes was published by Sano et al. (70) in 1996 and the results of this study were published 1 year later (71). The primary outcomes were the time to the occurence of any of the following: death, institutionalization, loss of the ability to perform basic activities of daily living, or severe dementia. There were significant delays in the time to the primary outcomes for the patients treated with (–)deprenyl. The authors concluded that in patients with moderately severe impairment from Alzheimer's disease treatment with (–)deprenyl slows the progression of the disease.

The place of an enhancer drug in slowing aging of the brain

In light of the peculiar changes in the enhancer regulation during the developmental phase of life, the antiaging potential of the administration of a small dose of a safe enhancer substance during the postdevelopmental (aging) phase of life deserves serious consideration. It seems reasonable to shift safely the functional constellation of the brain during post-developmental longevity towards the one characteristic of the "uphill" (young) period of life. Humans need medication with a very small amount of an enhancer, probably starting immediately after sexual maturity, to keep the engine of their brain on a higher activity level during post-developmental longevity. This will work for decades. It will improve the quality of life in the latter decades, hopefully shifting the time of natural death, probably decreasing the precipitation of age-related depression, maybe eliminating

the precipitation of Parkinson's disease, and possibly reducing or delaying the onset of Alzheimer's disease.

In longevity studies in animals the antiaging effect of an enhancer substance has already been convincingly proven. Rats treated during their post-developmental phase of life with a small dose of (–)deprenyl lost their ability to ejaculate later (31,34,43,46), showed a slower decline in learning performance with the passing of time (32,43,46), and lived significantly longer than their saline-treated peers (31,43,46). Our finding that (–)deprenyl prolongs life was corroborated in mice (3,86), in rats (24,58), in hamsters (73) and in dogs (69). Nevertheless, variation in the extent of the prolongation of life between the longevity studies performed in different laboratories was unusually high.

In one strain of mice (–)deprenyl treatment had no beneficial effect on survival (23). Stoll et al. (73) found that chronic treatment of male and female Syrian hamsters with the same low dose of (–)deprenyl significantly increases lifespan in females but not in males. Substantial strain differences were found in rats in the efficiency of (–)deprenyl in the longevity studies (see ref. 31,43,46 versus 24,58). The peculiar concentration dependency of the enhancer effect (see Fig. 13) may be one of the reasons that explains the unusually high variation in the optimal dose regarding the prolongation of life. The same dose of an enhancer drug may exert a peak effect on one strain, a much lower effect on another strain, and be ineffective on a third strain. However, it is also worth mentioning that (–)deprenyl is a relatively weak enhancer substance, and longevity studies in the future with (–)BPAP, a highly potent and selective enhancer compound, may lead to more uniform results.

One group found, in striking contrast with all other studies, an increased mortality in male Wistar rats treated with (–)deprenyl 0.5 mg/kg for up to 20 months (19). The toxicological studies performed with (–)deprenyl and decades of experiences on thousands of rats make clear that the administration of (–)deprenyl 0.5 mg/kg three times a week could not be responsible for the observed increased mortality in the rats.

Because of the peculiar pharmacologic spectrum of (–)deprenyl, it was proposed already in 1982 before the realization of its enhancer effect, that the drug is primarily destined to be used as a prophylactic agent in the healthy population for slowing the physiological age-related decline of the striatal dopaminergic neurons (29). Indeed, the experimental and clinical data on (–)deprenyl that have accumulated since the early 1980s, support this view. It seems reasonable to assume that if the patients selected for the DATATOP trial had had the opportunity to take, prior to the onset of the symptoms of Parkinson's disease, just 1 mg/day of (–)deprenyl during the whole postdevelopmental period of their life, they would have either avoided reaching the low level of striatal dopamine needed for the manifestation of the disease within their lifetime (as 99.9% of the human population avoid it), or at least they would have crossed the critical threshold substantially later. It seems reasonable to expect that (–)BPAP might better serve this aim than (–)deprenyl.

It is well known that the prevalence rate of age-related depression, Parkinson's disease, and Alzheimer's disease increases sharply in individuals over 60 years of age. It is compelling to start a randomized, controlled trial on a selected 60 year old male and female population free of any sign of an age-related disease, treating them daily until death with placebo, (–)deprenyl 1 mg or (–)BPAP 0.1 mg, respectively. We may reasonably hope to find within 5–10 years a statistically significant difference in the prevalence rate of the age-related diseases between placebo and enhancer drug–treated groups, so that the pro-

phylactic medication with an enhancer drug will be indicated during the downhill period of life.

THE ADVANTAGE OF (–)BPAP OVER (–)DEPRENYL

Collating the similarities and differences in the pharmacologic spectrum of (–)deprenyl and (–)BPAP with special regard to the enhancer effect, we may conclude that (–)BPAP, as an enhancer substance, is superior over (–)deprenyl for two reasons: selectivity and substantially higher potency.

(–)Deprenyl is a highly potent and selective inhibitor of MAO-B independently from its enhancer effect. It completely inhibits the activity of MAO-B at usual doses (0.25 mg/kg in the rat and 10 mg/day in man). MAO-A is inhibited by (–)deprenyl to a physiologically significant level only at very high doses (over 10 mg/kg in the rat and 30 mg/day in man). (–)Deprenyl was the first selective inhibitor of MAO-B (for the history of the early period see refs. 27,28,30); it is still used as a reference substance for the blockade of this enzyme, and is listed accordingly in the pharmacology handbooks. In the overwhelming majority of the papers published on (–)deprenyl, the drug is still handled solely as a selective inhibitor of MAO-B. It is reasonable to expect that the development of (–)BPAP will now turn, in general, toward enhancer regulation and it will be also helpful in clarifying the primary importance of this mechanism in most of the well known effects of (–)deprenyl in animals and man.

In contrast to (–)deprenyl, (–)BPAP, even in high concentrations, leaves MAO-B activity unchanged. Due to the close structural similarity to tryptamine, the main endogenous substrate for MAO-A, (–)BPAP is a weak, selective inhibitor of MAO-A, but this effect is insignificant from a pharmacologic point of view (49).

(–)Deprenyl is metabolized to (–)amphetamine and (–)methamphetamine in animals and in man (64,67). Nevertheless, unless the upper limit of the usually used dose range (0.25 mg/kg in the rat, 10 mg/day in man) is not exceeded, the amphetamine metabolites never reach the level of pharmacological significance.

The history of (–)deprenyl research clarified that the abuse liability of an amphetamine derivative rests upon its catecholamine-releasing property. During the nearly 30 years of clinical experience with (–)deprenyl there was no mention of a single case of human abuse of this compound although tens of thousands of patients were treated with the drug. It is the unanimous opinion of clinicians that (–)deprenyl is free of amphetamine-like abuse liability. Using a battery of tests in rats and monkeys, a careful experimental analysis confirmed that (–)deprenyl lacks amphetamine-like abuse potential (85). With regard to (–)BPAP, a structure unrelated to the amphetamines, there is no reason to raise the abuse liability problem at all.

In context to the safety of enhancer substances, it is worth mentioning that doses much lower than the presently used doses of (–)deprenyl (0.01–0.05 mg/kg in the rat and 1 mg/day in man) are sufficient for exerting the enhancer effect and with (–)BPAP less than one tenth of this dose is more than enough to substitute for (–)deprenyl (37,49). As (–)deprenyl is a safe drug even in the presently applied higher dose, (–)BPAP can be expected to exert its specific enhancer effect without any noticeable side effects.

Both PEA and tryptamine, as well as their close structural relatives, do not have substantial affinity for the uptake machinery as their binding requirements for the plasma membrane amine transporter are not too strict. In accordance with this, both, (–)deprenyl and (–)BPAP, compete, for example, with the uptake of tyramine. The pulmonary artery of the rabbit is a highly sensitive preparation to detect the catecholamine-releasing effect of an indirectly acting amine. Tyramine elicits contraction of the artery strip in a dose-dependent manner by inducing the outflow of norepinephrine from the neuronal stores. This effect of tyramine is blocked not only by the uptake inhibitors, but also by (–)deprenyl and (–)PPAP, which interfere with the binding of tyramine to the plasma membrane amine transporter (for review see ref. 36). Similar to (–)deprenyl and (–)PPAP, (–)BPAP (2 mg/L) fully inhibited the tyramine-induced release of norepinephrine from the pulmonary artery strip (49). Because of the very low concentration of (–)BPAP needed to exert the enhancer effect of the substance, this interference with the binding of endogenous amines to the uptake machinery is without pharmacologic significance.

Though specific binding to the catecholamine and serotonin receptors shows a much higher structural selectivity than binding to the plasma membrane amine transporter, drugs used in therapy as agonists or antagonists of one or another type of pre- or postsynaptic catecholamine or serotonin receptors show substantial affinity to the whole group of these receptors. We measured the specific binding of (–)deprenyl, (–)BPAP, and (+) BPAP to catecholamine and serotonin receptors using the dopamine D_2 receptor agonist bromocriptine as a reference compound because (–)deprenyl is known to act primarily as a stimulant of the dopaminergic system in the brain. (–)Deprenyl and both enantiomers of BPAP showed a specific binding capacity only to the α_2-adrenoceptors, while bromocriptine inhibited the radioligand binding to all catecholamine and serotonin receptors examined (see ref. 49; Table 4).

All in all, (–)BPAP is the most potent and most selective stimulant of the enhancer-sensitive neurons in the mammalian brain presently known.

SUMMARY AND CONCLUSIONS

The specific brain activation mechanism ("drive") that ensures that living beings surmount every obstacle to reach a goal, even if life is in the balance, roots in the existence of "enhancer-sensitive" neurons in the brain that are ready to increase their activity with lightning speed in response to endogenous "enhancer" substances, of which phenylethylamine (PEA) and tryptamine are the presently known examples. PEA and tryptamine enhance the impulse-propagation-mediated release of catecholamines and serotonin in the brain (CAE/SAE effect). This is the best model for studying the enhancer regulation in the mammalian brain, which starts working at the discontinuation of breast feeding. Weaning is the beginning of the developmental ("uphill") period of life and is characterized by significantly higher brain activity levels that last until the sexual hormones dampen this regulation, thereby terminating the uphill period. This is the prelude of the postdevelopmental ("downhill") phase of life and the beginning of the slow brain aging process from which there is no escape until natural death.

It has been proposed that enhancer compounds can delay the natural age-related deterioration of brain performance and keep the brain on a higher activity level during post-development longevity. PEA, a substrate of MAO-B, and tryptamine, a substrate of MAO-A, are rapidly metabolized, short-acting endogenous enhancer compounds. PEA and its long-acting derivatives, amphetamine and methamphetamine, which are not metabolized by MAO, are enhancer substances at low concentrations but also potent releasers of catecholamines and serotonin from their pools at higher concentrations. The catecholamine-releasing effect masked for decades the enhancer property of these compounds.

(–)Deprenyl (selegiline) is the first PEA derivative free of the catecholamine-releasing property and made possible the discovery of the enhancer regulation in the brain. This drug is presently the only clinically used enhancer compound. (–)Deprenyl is also a highly potent, selective inhibitor of MAO-B and is metabolized to amphetamines. Tryptamine is an endogenous enhancer substance free of the catecholamine/serotonin-releasing property. The newly developed tryptamine derivative (–)BPAP is the first highly selective enhancer substance. It is also much more potent than (–)deprenyl.

Enhancer substances that keep the enhancer-sensitive neurons on a higher activity level slow the age-related deterioration of the mammalian brain. Maintenance of rats on (–)deprenyl during post-developmental longevity slows the age-related decline of sexual and learning performances and prolongs life significantly. Patients with early Parkinson's disease who are maintained on (–)deprenyl need levodopa significantly later than their placebo-treated peers and they live significantly longer when on levodopa plus (–)deprenyl than patients on levodopa alone. In patients with moderately severe impairment from Alzheimer's disease, treatment with (–)deprenyl slows the progression of the disease.

(–)BPAP is an especially promising prophylactic antiaging compound that may provide the opportunity to shift the functional constellation of the brain during postdevelopmental longevity towards the one characteristic to the uphill period of life. According to the available experimental and clinical data, it is reasonable to expect that daily administration of an enhancer drug [e.g., (–)deprenyl 1 mg or (–)BPAP 0.1 mg] from sexual maturity until death will improve quality of life in the latter decades, shift the time of natural death, decrease the precipitation of age-related depression, and reduce the prevalence of Parkinson's disease and Alzheimer's disease.

REFERENCES

1. Agnoli A, Fabbrini G, Fioravanti M, Martucci N. CBF and cognitive evaluation of Alzheimer-type patients before and after MAO-B treatment: A pilot study. *Eur Neuropsychopharmacol* 1992;2:31–35.
2. Allain H, Gougnard J, Naukirek HC. Selegiline in *de novo* parkinsonian patients: The French selegiline multicenter trial (FSMP). *Acta Neurol Scand* 1991;136:73–78.
3. Archer JR, Harrison DE. L-Deprenyl treatment in aged mice slightly increases lifespans and greatly reduces fecundity by aged males. *J Gerontol Sci* 1996;13A:B448–B453.
4. Birkmayer W, Hornykiewicz O. Der L-Dioxyphenyl-alanin-Effekt beim Parkinson Syndrom des Menschen. *Arch Psychiat Nervenkrh* 1962;203:560–564.
5. Birkmayer W, Riederer P. *Parkinson's Disease. Biochemistry, Clinical Pathology and Treatment.* New York: Springer-Verlag, 1983:1–194.
6. Birkmayer W, Riederer P, Ambrozi L, Youdim MBH. Implications of combined treatment with "Madopar" and L-Deprenil in Parkinson's disease. *Lancet* 1977;1:439–443.

7. Birkmayer W, Riederer P, Linauer W, Knoll J. L-Deprenyl plus L-phenylalanine in the treatment of depression. *J Neural Transm* 1984;59:81–87.
8. Birkmayer W, Knoll J, Riederer P, Youdim MBH, Hars V, Marton J. Increased life expectancy resulting from addition of L-deprenyl to Madopar treatment in Parkinson's disease: A long-term study. *J Neural Transm* 1985;64:113–127.
9. Burke WJ, Roccaforte WH, Wengel SP, Bayer BL, Ranno AE, Willcockson NK. L-Deprenyl in the treatment of mild dementia of the Alzheimer type: Results of a 15-month trial. *J Am Geriatr Soc* 1993;41:1219–1225.
10. Campi N, Todeschini GP, Scarzella L. Selegiline versus L-acetylcarnitine in the treatment of Alzheimer-type dementia. *Clin Ther* 1990;12:306–314.
11. Carrillo MC, Kanai S, Nokubo M, Kitani K. (–)Deprenyl induces activities of both superoxide dismutase and catalase but not of glutathion peroxidase in the striatum of young male rats. *Life Sci* 1991;48:517–521.
12. Carrillo MC, Kanai S, Nokubo M, Ivy GO, Sato Y, Kitani K. (–)Deprenyl increases activities of superoxide dismutase and catalase in striatum but not in hippocampus: The sex- and age-related differences in the optimal dose in the rat. *Exp Neurol* 1992;116:286–294.
13. Cohen G, Pasik P, Cohen B, Leist A, Mitileneou C, Yahr MD. Pargyline and (–)deprenyl prevent the neurotoxicity of 1-methyl-4-phenyl-1,2,3,6-tetra-hydropyridine (MPTP) in monkeys. *Eur J Pharmacol* 1984;106:209–210.
14. Elsworth JD, Glover V, Reynolds GP, et al. Deprenyl administration in man: A selective monoamine oxidase B inhibitor without the "cheese effect." *Psychopharmacology* 1978;57:33–38.
15. Falsaperle A, Monici Preti PA, Oliani C. Selegiline versus oxiracetam in patients with Alzheimer-type dementia. *Clin Ther* 1990;12:376–384.
16. Finali G, Piccirilli M, Oliani C, Piccinin GL. Alzheimer-type dementia and verbal memory performances: Influence of selegiline therapy. *Ital J Neurol Sci* 1992;13:141–148.
17. Finali G, Piccirilli M, Oliani C, Piccinin GL. L-deprenyl therapy improves verbal memory in amnesic Alzheimer patients. *Clin Neuropharmacol* 1991;14:523–536.
18. Finnegan KT, Skratt JJ, Irvin I, DeLanney LE, Langston JW. Protection against DSP-4 induced neurotoxicity by deprenyl is not related to its inhibition of MAO-B. *Eur J Pharmacol* 1990;184:119–126.
19. Gallagher IM, Clow A, Glover V. Long term administration of (–)deprenyl increases mortality in male Wistar rats. *J Neural Transm* 1998;52(Suppl):315–320.
20. Goad DL, Davis CM, Liem P, Fuselier CC, McCormack JR, Olsen KM. The use of selegiline in Alzheimer's patients with behavior problems. *J Clin Psychiatry* 1991;52:342–345.
21. Groc L, Levine RA, Foster JA, Normile HJ, Weissmann D, Bezin L. Evidence of deprenyl-insensitive apoptosis of nigral dopamine neurons during development. *Brain Res Dev Brain Res* 2000;120:95–98.
22. Hársing RG, Magyar K, Tekes K, Vizi ES, Knoll J. Inhibition by (–)-deprenyl of dopamine uptake in rat striatum: A possible correlation between dopamine uptake and acetylcholine release inhibition. *Pol J Pharmacol Pharm* 1979;31:297–307.
23. Ingram DK, Wiener HL, Chachich ME, Longo JM, Hengemihle J, Gupta M. Chronic treatment of aged mice with L-deprenyl produced marked MAO-B inhibition but no beneficial effects on survival, motor performance, or nigral lipofuscin accumulation. *Neurobiol Aging* 1993;14:431–440.
24. Kitani K, Kanai S, Sato Y, Ohta M, Ivy GO, Carillo MC. Chronic treatment of (–)deprenyl prolongs the lifespan of male Fischer 344 rats. Further evidence. *Life Sci* 1992;52:281–288.
25. Knoll J. Experimental studies on the higher nervous activity of animals. VI. Further studies on active reflexes. *Acta Physiol Hung* 1957;12:65–92.
26. Knoll J. *The Theory of Active Reflexes. An Analysis of Some Fundamental Mechanisms of Higher Nervous Activity.* New York: Hafner Publishing Company, 1969:1–131.
27. Knoll J. Analysis of the pharmacological effects of selective monoamine oxidase inhibitors. In: *Monoamine Oxidase and Its Inhibition*. Wolstenholme GES, Knight J, eds. Amsterdam: Elsevier, 1976:131–161.
28. Knoll J. The possible mechanism of action of (–)deprenyl in Parkinson's disease. *J Neural Transm* 1978;43: 177–198.
29. Knoll J. Selective inhibition of B type monoamine oxidase in the brain: A drug strategy to improve the quality of life in senescence. In: *Strategy in Drug Research.* Keverling Buisman JA, ed. Amsterdam: Elsevier, 1982:107–135.
30. Knoll J. Deprenyl (selegiline). The history of its development and pharmacological action. *Acta Neurol Scand* 1983;59(Suppl):57–80.
31. Knoll J. The striatal dopamine dependency of lifespan in male rats. Longevity study with (–)deprenyl. *Mech Ageing Dev* 1988;46:237–262.
32. Knoll J. The pharmacology of selegiline/(–)deprenyl. *Acta Neurol Scand* 1989;126:83–91.
33. Knoll J. Nigrostriatal dopaminergic activity, deprenyl treatment, and longevity. *Adv Neurol* 1990;53: 425–429.

34. Knoll J. Pharmacological basis of the therapeutic effect of (–)deprenyl in age-related neurological diseases. *Med Res Rev* 1992;12:505–524.
35. Knoll J. Memories of my 45 years in research. *Pharmacol Toxicol* 1994;75:65–72.
36. Knoll J. Rationale for (–)deprenyl (selegiline) medication in Parkinson's disease and in prevention of age-related nigral changes. *Biomed Pharmacother* 1995;49:187–195.
37. Knoll J. (–)Deprenyl (selegiline), a catecholaminergic activity enhancer (CAE) substance acting in the brain. *Pharmacol Toxicol* 1998;82:57–66.
38. Knoll J, Magyar K. Some puzzling effects of monoamine oxidase inhibitors. *Adv Bioch Psychopharmacol* 1972;5:393–408.
39. Knoll J, Miklya I. Enhanced catecholaminergic and serotoninergic activity in rat brain from weaning to sexual maturity. Rationale for prophylactic (–)deprenyl (selegiline) medication. *Life Sci* 1995;56:611–620.
40. Knoll J, Ecseri Z, Kelemen K, Nievel J, Knoll B. Phenylisopropylmethyl- propinylamine (E-250), a new psychic energizer. *Arch Int Pharmacodyn Ther* 1965;155:154–164.
41. Knoll J, Vizi ES, Somogyi G. Phenylisopropylmethylpropinylamine (E-250), a monoamine oxidase inhibitor antagonizing the effects of tyramine. *Arzneimittelforschung* 1968;18:109–112.
42. Knoll J, Yen TT, Dalló J. Long-lasting, true aphrodisiac effect of (–)deprenyl in sexually sluggish old male rats. *Mod Prob Pharmacopsychiatry* 1983;19:135–153.
43. Knoll J, Dalló J, Yen TT. Striatal dopamine, sexual activity and lifespan. Longevity of rats treated with (–)deprenyl. *Life Sci* 1989;45:525–531.
44. Knoll J, Knoll B, Török Z, Timár J, Yasar S. The pharmacology of 1-phenyl-2-propylaminopentane (PPAP), a deprenyl-derived new spectrum psychostimulant. *Arch Int Pharmacodyn Ther* 1992;316:5–29.
45. Knoll J, Tóth V, Kummert M, Sugár J. (–)Deprenyl and (–)parafluorodeprenyl- treatment prevents age-related pigment changes in the substantia nigra. A TV-image analysis of neuromelanin. *Mech Ageing Dev* 1992;63:157–163.
46. Knoll J, Yen TT, Miklya I. Sexually low performing male rats die earlier than their high performing peers and (–)deprenyl treatment eliminates this difference. *Life Sci* 1994;54:1047–1057.
47. Knoll J, Miklya I, Knoll B, Markó R, Kelemen K. (–)Deprenyl and (–)1-phenyl-2-propylaminopentane, [(–)PPAP], act primarily as potent stimulants of action potential-transmitter release coupling in the catecholaminergic neurons. *Life Sci* 1996;58:817–827.
48. Knoll J, Miklya I, Knoll B, Markó R, Rácz D. Phenylethylamine and tyramine are mixed-acting sympathomimetic amines in the brain. *Life Sci* 1996;58:2101–2114.
49. Knoll J, Yoneda F, Knoll B, Ohde H, Miklya I. (–)1-(Benzofuran-2-yl)-2-propylaminopentane, [(–)BPAP], a selective enhancer of the impulse propagation mediated release of catecholamines and serotonin in the brain. *Br J Pharmacol* 1999;128:1723–1732.
50. Knoll J, Miklya I, Knoll B, Dalló J. Sexual hormones terminate in the rat the significantly enhanced catecholaminergic/serotoninergic tone in the brain characteristic to the post-weaning period. *Life Sci* 2000;67: 765–773.
51. Larsen JP, Boas J, Erdal JE. Does selegiline modify the progression of early Parkinson's disease? Results from a five-year study. The Norwegian-Danish Study Group. *Eur J Neurol* 1999;6:539–547.
52. Lees AJ. Selegiline hydrochloride and cognition. *Acta Neurol Scand* 1991;136(Suppl):91–94.
53. Lees AJ. Comparison of therapeutic effects and mortality data of levodopa and levodopa combined with selegiline in patients with early, mild Parkinson's disease. *Br Med J* 1995;311:1602–1607.
54. Mangoni A, Grassi MP, Frattola L, Piolti R, Bassi S, Motta A. Effects of a MAO-B inhibitor in the treatment of Alzheimer disease. *Eur Neurol* 1991;31:100–107.
55. Mann JJ, Gershon S. A selective monoamine oxidase-B inhibitor in endogenous depression. *Life Sci* 1980;26:877–882.
56. Martin C. Sexual activity in the aging male. In: *Handbook of Sexology.* Money J, Musaph H, eds. Amsterdam: Elsevier, 1977:813–824.
57. Martini E, Pataky I, Szilágyi K, Venter V. Brief information on an early phase-II study with (–)deprenyl in demented patients. *Pharmacopsychiatry* 1987;20:256–257.
58. Milgram MW, Racine RJ, Nellis P, Mendoca A, Ivy GO. Maintenance on L-(–)deprenyl prolongs life in aged male rats. *Life Sci* 1990;47:415–420.
59. Monteverde A, Gnemmi P, Rossi F, Monteverde A, Finali GC. Selegiline in the treatment of mild to moderate Alzheimer-type dementia. *Clin Ther* 1990;12:315–322.
60. Myttyla VV, Sotaniemi KA, Vourinen JA, Heinonen EH. Selegiline as initial treatment in *de novo* parkinsonian patients. *Neurology* 1992;42:339–343.
61. Parkinson Study Group. Effect of (–)deprenyl on the progression disability in early Parkinson's disease. *New Engl J Med* 1989;321:1364–1371.

62. Parkinson Study Group. Effect to tocopherol and (–)deprenyl on the progression of disability in early Parkinson's disease. *New Engl J Med* 1993;328:176–183.
63. Parkinson Study Group. Impact of deprenyl and tocopherol treatment of Parkinson's disease in DATATOP patients requiring levodopa. *Ann Neurol* 1996;39:37–45.
64. Phillips SL. Amphetamine, p-hydroxyamphetamine and β-phenylethylamine in mouse brain and urine after (–)- and (+)-deprenyl administration. *J Pharm Pharmacol* 1981;31:739–741.
65. Piccinin GL, Finali GC, Piccirilli M. Neuropsychological effects of L-deprenyl in Alzheimer's type dementia. *Clin Neuropharmacol* 1990;13:147–163.
66. Sandler M, Glover V, Ashford A, Stern GM. Absence of "cheese effect" during deprenyl therapy: Some recent studies. *J Neural Transm* 1978;43:209–215.
67. Reynolds GP, Elsworth JD, Blau K, Sandler M, Lees AJ, Stern GM. Deprenyl is metabolized to methamphetamine and amphetamine in man. *Br J Clin Pharmacol* 1978;6:542–544.
68. Rinne JO, Röyttä M, Paljärvi L, Rummukainen J, Rinne UK. Selegiline (deprenyl) treatment and death of nigral neurons in Parkinson's disease. *Neurology* 1991;41:859–861.
69. Ruehl WW, Entriken TL, Muggenberg BA, Bruyette DS, Griffith WG, Hahn FF. Treatment with L-deprenyl prolongs life in elderly dogs. *Life Sci* 1997;61:1037–1044.
70. Sano M, Ernesto C, Klauber MR, and Members of the Alzheimer's disease Cooperative Study. Rationale and design of a multicenter study of selegiline and α-tocopherol in the treatment of Alzheimer disease using novel clinical outcomes. *Alzheimer Dis Assoc Disord* 1996;10:132–140.
71. Sano M, Ernesto C, Thomas RG, et al. A controlled trial of selegiline, alpha-tocopherol, or both as treatment for Alzheimer's disease. *New Engl J Med* 1997;336:1216–1222.
72. 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.
73. Stoll S, Hafner U, Kranzlin B, Muller WE. Chronic treatment of Syrian hamsters with low-dose selegiline increases lifespan in females but not males. *Neurobiol Aging* 1997;18:205–211.
74. Suuronen T, Kolehmainen P, Salminen A. Protective effect of L-deprenyl against apoptosis induced by okadaic acid in cultured neuronal cells. *Biochem Pharmacol* 2000;59:1589–1595.
75. Tariot PN, Cohen RM, Sunderland T, Newhouse PA, Yount D, Mellow AM. L-(–)deprenyl in Alzheimer's disease. *Arch Gen Psychiatry* 1987;44:427–433.
76. Tatton WG. Apoptotic mechanisms in neurodegeneration: Possible relevance to glaucoma. *Eur J Ophthalmol* 1999;9(Suppl):S22–S29.
77. Tetrud JW, Langston JW. The effect of (–)deprenyl (selegiline) on the natural history of Parkinson's disease. *Science* 1989;245:519–522.
78. Tringer L, Haits G, Varga E. The effect of L-E-250 (-L-phenyl-isopropylmethyl-propinyl-amine HCl) in depression. In: V. *Conferentia Hungarica pro Therapia et Investigatione in Pharmacologia*. Leszkovszky G, Ed. Budapest: Publishing House of the Hungarian Academy of *Science*s, 1971:111–114.
79. ThyagaRajan S, Meites J, Quadri SK. Deprenyl reinitiates estrous cycles, reduces serum prolactin, and decrease the incidence of mammary and pituitary tumors in old acyclic rats. *Endocrinology* 1995;136: 1103–1110.
80. ThyagaRajan S, Felten SY, Felten DL. Antitumor effct of L-deprenyl in rats with carcinogen-induced mammary tumors. *Cancer Lett* 1998;123:177–183.
81. Varga E. Vorläufiger Bericht über die Wirkung des Präparates E-250 (phenyl- isopropyl-methyl-propinyl-amine-chlorhydrat) In: *III Conferentia Hungarica pro Therapia et Investigatione in Pharmacologia*. Dumbovich B, ed. Budapest: Publishing House of the Hungarian Academy of Sciences, 1965:197–201.
82. Varga E, Tringer L. Clinical trial of a new type of promptly acting psychoenergetic agent (phenyl-isopropyl-methyl-propinylamine HCl, E-250). *Acta Med Acad Sci Hung* 1967;23:289–295.
83. Vizuete ML, Steffen V, Ayala A, Cano J, Machado A. Protective effect of deprenyl against 1-methyl-4-phenylpyridinium neurotoxicity in rat striatum. *Neurosci Lett* 1993;152:113–116.
84. Wu RM, Chiuech CC, Pert A, Murphy DL. Apparent antioxidant effect of l-deprenyl on hydroxyl radical formation and nigral injury elicited by MPP^+ *in vivo*. *Eur J Pharmacol* 1993;243:241–247.
85. Yasar S, Winger G, Nickel B, Schulze G, Goldberg SR. Preclinical evaluation of l-deprenyl: Lack of amphetamine-like abuse potential. In: *Inhibitors of Monoamine Oxidase B*. Szelenyi I, Ed. Basel: Birkhäuser Verlag, 1993:215–233.
86. Yen TT, Knoll J. Extension of lifespan in mice treated with Dinh lang (Policias fruticosum L) and (–)deprenyl. *Acta Physiol Hung* 1992;79:119–124.