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STRIATAL DOPAMINE, SEXUAL ACTIVITY AND LIFESPAN. LONGEVITY OF RATS TREATED WITH (-)DEPRENYL

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Summary

The influence of longterm deprenyl treatment on the sexual performance and lifespan of male rats was studied. One hundred and thirty two rats were treated from the end of their 2nd year of life either with saline (1 ml/kg, s.c.) (n=66) or with deprenyl (0.25 mg/kg, s.c.) (n=66) three times a week until death. Whereas none of the two-year-old saline-treated rats displayed full scale sexual activity, this appeared in 64 out of 66 rats on deprenyl. The longest living rat in the saline-treated group lived 164 weeks. The lifespan of the group was 147.05±0.56 weeks. The shortest living animal in the (-)deprenyl-treated group lived 171 weeks and the longest living rat died during the 226th week of its life. The lifespan was 191.91±2.31 weeks. This is the first instance that a well aimed medication prolonged lifespan of members of a species beyond their maximum age of death (182 weeks in the rat). A close relation between sexual activity and lifespan was detected.

In the human striatum dopamine content and number of neurons are known to decline rapidly beyond the age of 45 years (1). Dopamine itself is with high probability the culpable substance for these changes (2). The complex autooxidation of the high amounts of dopa and dopamine in the striatum, continuously generating substantial quantities of toxic free radicals and highly reactive quinones, creates a permanent danger for the nigrostriatal dopaminergic neuron, which has to mobilize its natural defensive measures to protect the neuron from the deleterious effect of these toxic by-products. Neuromelanin, which is generated via the polymerization of oxidative products of dopamine with the evident aim of finally depositing waste products, is in the substantia nigra the visible sign of successful self-defense of the neuron against the free radicals and quinones originating from dopamine metabolism. The sluggish depositing of neuromelanin in the human substantia nigra (3) is in excellent agreement with this view.

Aging of the nigrostriatal dopaminergic neuron is essentially similar in rodents as in human (4). Direct biochemical evidence to age-related decline of striatal dopaminergic function in the rat is

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the loss of striatal D_2-receptors in the aging rat brain (5,6,7).

It seems reasonable to develop drugs which provide protection against the self-produced neurotoxins and slow down the age-related changes in the nigrostriatal dopaminergic neuron. (-)Deprenyl, the selective inhibitor of B-type MAO (8,9) prevents dopamine breakdown, facilitates the activity of the nigrostriatal neuron with high selectivity (10,11) and protects this neuron from the neurotoxicity of 6-hydroxydopamine (6-OHDA) (12) and 1-methyl-4-phenyl-1,2,5,6-tetrahydropyridine (MPTP) (13).

Given concurrently with levodopa and a peripheral decarboxylase inhibitor (-)deprenyl is now widely used in Parkinson's disease (1), it is an effective antidepressant (14,15) and its benefical effect in Alzheimer's disease was recently demonstrated (16). Parkinsonian patients receiving this drug may survive longer than those who do not (17). Such a finding was adumbrated in 1981 (18,19) and a subsequent pilot study (20) provided prima facie evidence that (-)deprenyl increases life expectancy in the rat. We now provide data showing that (-)deprenyl-treated senescent rats live substantially longer and display considerably enhanced sexual activity compared with saline-treated controls.

Methods

Measurement of sexual activity. Sexually inexperienced male albino rats (first generation Wistar males x Logan females) were used. The rats were housed in a well-ventillated animal house in plastic cages (35x35x25 cm) each containing 6 rats. They were kept under 12:12 h light-dark cycle, fed with standard diet, allowing food and water ad libitum. Experiments were carried out in the light phase.

Ovariectomized females were used as stimulus objects and kept under the same conditions as the males. The females were brought into heat by the subcutaneous injection of 30 µg estradiol monoproprionate followed 48 h later by 0.5 mg progesterone and were used 4-7 h after progesterone injection. Indicator males, which achieved intromission with high frequency, were used for the selection of receptive females. Only females showing high receptivity were brought together with the males.

Copulatory tests were performed in dimly lit environment. The male rat was transferred from the home cage to an ovoid-shaped observation cage (40x40x60 cm) and then allowed 10-min adaptation three times a week. At the fourth occasion the stimulus female was put into this cage 5-min after the adaptation of the male. During 30 min copulatory patterns of the male were recorded, according to Beach (21), by an experimenter.

The experiments were performed on 132 randomly selected sexually inexperienced male rats which completed the 23rd month of their life. Copulatory activity of the rats was first tested in four consecutive weekly mating tests (screening period).

The copulatory patterns: mounting, intromission, ejaculation were followed during the 30-min observation period. According to their copulatory behavior in the screening tests, rats were classified as follows: (1) sexually inactive males, non-copulators, which

failed to mount at any occasion during the 4 weeks of the screening test period; (2) males which mounted only; (3) males which displayed mounting and intromission, but failed to ejaculate, 'sexually sluggish rats'; and (4) sexually active males with full-scale sexual activity.

Results

Sexual activity shows an age-related decline in the rat (19). In agreement with our previous findings none of the 132 rats used in this experiment displayed ejaculation as they completed their 2nd year. Of the total in the present experiment, 44 were sexually sluggish (mounting and intromission, but no ejaculation), 42 showed mounting only and neither pattern appeared in 46 ('non-copulators'). Each of the three categories was divided into equal groups, which were treated subcutaneously with saline (0.1 ml/100 g) or (-)deprenyl (0.25 mg/kg), respectively, three times a week. Sexual activity was assessed once a week.

In the saline-treated group (n=66), sexual activity decreased rapidly, the last intromission being displayed at the 23rd week of treatment and the last mounting at the 33rd week. These control rats began to die at the 36th week of treatment and the last died at the 60th week (Fig. 1). Thus, the shortest-lived animal in the saline-treated group died at age 32 months (140 weeks), 50% were dead by the end of the 34th month (147th week) and the last died during his 38th month (164th week). This is in good agreement with our previous experience.

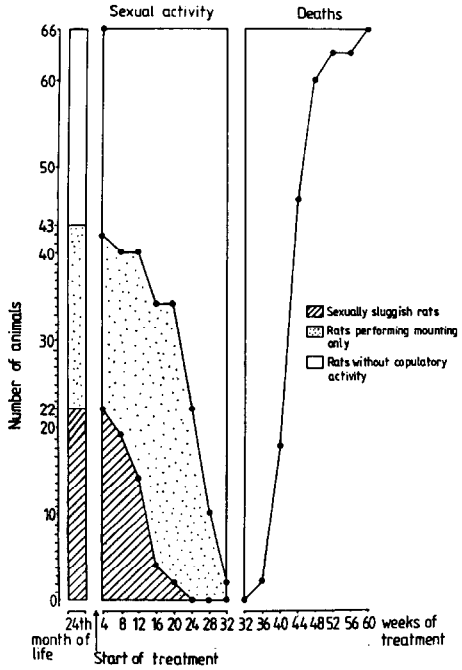

FIG. 1

Death-rate and age-related decay of sexual activity in saline-treated senescent male rats (n=66)
Sexual function prior to saline treatment was first tested during the 24th month of life. After four consecutive weekly mating tests, continuous saline treatment (0.1 mg/100 g subcutaneously on Mondays, Wednesdays and Fridays) commenced and sexual activity was subsequently tested weekly. Left: distribution according to sexual activity before and during saline treatment, illustrating decline in sexual performance up to the 32nd week of treatment. Right: death rate.

In the deprenyl-treated group (n=66), sexual potency increased gradually, reaching a maximum between the 28th and 36th week of treatment. Full scale sexual activity manifested in 64 out of 66 rats. The first rat in this group died at the 67th week of treatment (171st week of age) and the longest lived animal died at the 122nd week of treatment (226th week of age)(Fig. 2). Thus, in the (-)deprenyl-treated group, the shortest lived animal lived longer than the longest-lived animal of the saline-treated group, 50% were dead by the end of the 45th month of their life, living 9 months (32%) longer than their saline-treated peers and 31 rats completed 4 years of life.

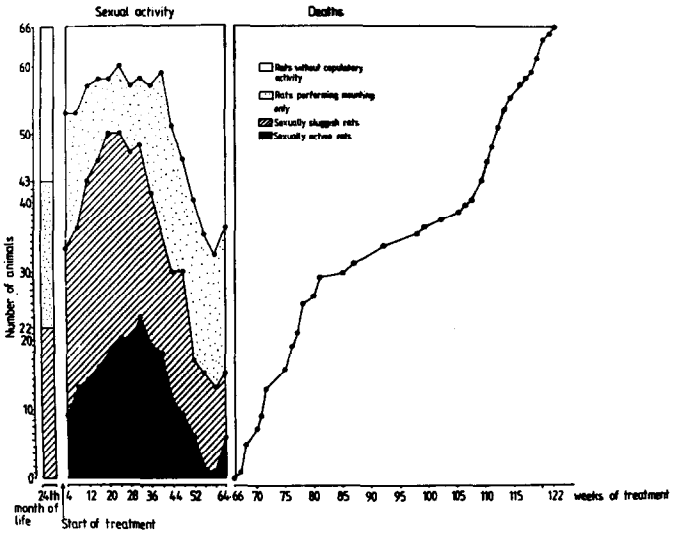

FIG. 2

Influence of (-)deprenyl treatment on death-rate and sexual activity in senescent male rats (n=66)
Course of experiment corresponds with control experiment (Fig. 1), the only difference being that (-)deprenyl (0.25 mg/kg) was administered subcutaneously as 0.025% solution in saline (0.1 ml/100 g) instead of saline alone. Changes in sexual activity illustrated up till 64th week of treatment. Three weeks later the first death in the group occurred. (-)Deprenyl-treated animals lived longer and their sexual activity was increased and of longer duration compared with saline-treated peers (see Fig. 1). The differences are so great that any statistical analysis would be supererogatory.

The shape of the death-rate curve of the (-)deprenyl-treated rats clearly shows two distinct populations, one which died out much earlier than the other. A close correlation between sexual activity and lifespan in the male rats explains this phenomenon. As shown in Fig. 2, treatment with the drug greatly increases sexual performance. Out of 66 rats, there were only two which never manifested ejaculation. A subgroup ejaculated only on 0 to 5 occasions and

were designated low performers (LP). 35 ejaculated in 6 to 15 tests and a further group of high performers (HP) in more than 15 tests. Fig. 3 shows the striking difference in the lifespan between LP (n=14) and HP (n=17). The last animal of the LP died on the 85th week of treatment, i.e. was 189 weeks old, whereas, the first animal in the HP group died during the 98th week of treatment (was 202 weeks old) and the last one during the 122nd week of treatment (was 226 weeks old).

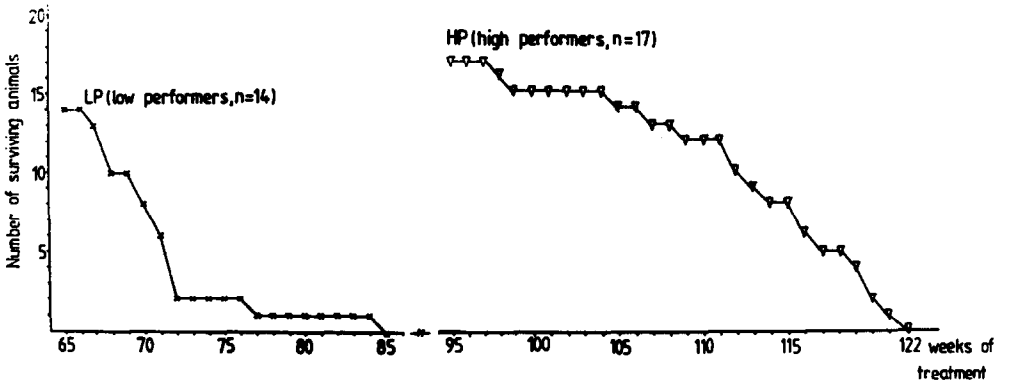

FIG. 3

Correlation between duration of life and sexual activity in (-)deprenyl-treated senescent male rats
LP (n=14): death-rate of sexually least active animals.
HP (n=17): death-rate of sexually most active animals.

Table I shows the lifespan of rats grouped according to their sexual performance displayed during the 24th month of their life. The better performers, either in the saline- or in the (-)deprenyl-treated groups, lived significantly longer than their less active peers. The difference in the duration of life between saline- and (-)deprenyl-treated rats is dramatic.

Discussion

We found two papers in literature dealing with the relation between dopaminergic activity and life expectancy in rodents. Cotzias et al (22) demonstrated that levodopa administered to mice in their diet significantly increased life expectancy, but no change in the maximum age of death was detected. Clemens et al (23) showed that, compared to their untreated peers, higher percentage of female rats treated with the dopamine agonist, lergotrile mesilate, were alive

at the end of their second year. These data are in essential agreement with our findings with (-)deprenyl.

TABLE I. Duration of Life of Rats Treated with Saline (n=66) and (-)Deprenyl (n=66), Respectively

Classification of the groups according to sexual performance before treatment	No. of animals	Average lifespan (weeks) saline-treated		(-)deprenyl-treated	
Non-copulators	23	142.74±0.38	A	187.90±3.27	D
Mounting rats	21	146.95±0.42	B	191.95±3.59	E
Sluggish rats	22	152.00±0.92	C	214.05±3.07	F
Total	66	147.05±0.56 (median: 148)	G	191.91±2.31 (median: 182)	H

Significances according to the Student's t test for two means:
A:B $p < 0.001$; A:C $p < 0.001$; B:C $p < 0.001$; A:D $p < 0.001$;
B:E $p < 0.001$; C:F $p < 0.001$; D:E $p > 0.05$; D:F $p < 0.001$;
E:F $p < 0.001$; G:H $p < 0.001$

According to multiple analysis of variance (ANOVA), too, the difference between the saline- versus (-)deprenyl-treated groups ($F= 257.2$ /df 1.42/ $p < 0.001$), as well as the difference between different sex groups (non copulators, mounting rats, sexually sluggish rats: $F= 128.58$ /df 2.82/ $p < 0.001$) and the interaction ($F= 40.23$ /df 2.82/ $p < 0.001$) were found to be highly significant.

In contrast to levodopa and to the postsynaptic dopamine agonists, the long-term administration of which has serious side effects, (-)deprenyl is a safe drug. Due to its safeness and utmost selectivity to the nigrostriatal dopaminergic neurons, (-)deprenyl is a unique tool for studying the physiological significance of the nigrostriatal dopaminergic system.

The fact that we succeeded to keep alive a male rat population beyond the maximum age of death (182 weeks in the rat) by the aid of long term small dose administration of (-)deprenyl, is unprecedented. This is the first instance that, due to a well-aimed medication, members of a species reached, as an average lifespan, that age, which is taken as the maximum age of death or technical lifespan (TLS) of the species. To the relevance of this finding the important notion of Hayflick (24) should be quoted: "it is probable that only by increasing lifespan, or maximum age of death, of members of a species, will important insights be made into the aging process".

According to a retrospective study (17), parkinsonian patients with supplementation of (-)deprenyl (n=564) lived 15.3 months ($p < 0.001$) longer than those on Madopar alone (n=377). The possibility of increasing life expectation, improving the quality of life in senescent human population and decrease the incidence of such age-related mental illnesses like Parkinson's disease and Alzheimer's disease, by the continuous small dose administration of (-)deprenyl

(2-3 tablets a week) from the 5th decade of life is now open for clinical scrutiny.

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