BIOGERONTOLOGY

Geroprotective Effect of Epithalamine (Pineal Gland Peptide Preparation) in Elderly Subjects with Accelerated Aging

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A 12-year randomized clinical study of epithalamine (pineal gland peptide preparation) was carried out in elderly patients with coronary disease and accelerated aging of the cardiovascular system. Long-term treatment with epithalamine decreased the functional age and degree of cardiovascular aging; exercise tolerance increased. After 12 years the number of elderly subjects dead in the group treated by epithalamine was 28% lower than in the control group, despite the same basic therapy. Cardiovascular mortality was 2-fold lower in patients treated by epithalamine; the incidence of cardiovascular failure and respiratory diseases was 2-fold lower in this group. Long-term treatment with epithalamine was associated with a geroprotective effect on the long-term life prognosis in elderly subjects with accelerated aging.

Key Words: *pineal gland peptides; epithalamine; accelerated aging; prevention*

The priority problem of gerontology is prevention of accelerated aging (AA) in order to prolong the mean life span, retain active longevity, and attain the species threshold lifespan of humans [1, 2,14].

Experimental study showed that aging is associated with reduced synthesis of regulatory peptides and changes in target cell sensitivity to these peptides [15]. The peptides regulate protein biosynthesis processes in cells by modulating gene expression [8]. Impairment of peptide regulation affects functional state of the cell, which manifests at the molecular level by decreased gene expression and protein synthesis. It is hypothesized that impaired peptide regulation of gene expression leads to gradual fading of body functions [7,8]. Age-specific changes in the peptide regulation suggest that the use of peptides derived from animal organs and tissues or constructed by purposeful synthesis is a perspective trend of AA prevention [4,5,9].

Epithalamine (ET) is one of the first peptide preparations in the world medical practice, which exhibited high geroprotective efficiency and safety in experimental and clinical studies. Epithalamine is a complex of polypeptides with molecular weights of 1-10 kDa isolated from the pineal gland and containing no melatonin [7,9]. The preparation stimulates the function of pinealocytes and increases melatonin production by these cells [3,11]. The effect of ET on the function of the pineal gland seems to be very important, because age-related wea-

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kening of the melatonin-producing function has a significant impact on the body in general: leads to age-specific disorganization of biological rhythms, disorders in wake-sleeping cycle, neuroendocrine regulation, immunity, reproductive function, stress resistance, and promotes carcinogenesis [1,2].

The geroprotective effect of ET was noted not once. Long-term treatment with pineal gland peptides led to prolongation of the mean and maximum lifespans in rats, mice, and Drosophilidae and reduces the incidence of malignant tumors [1,7, 12]. The geroprotective effect of ET is attributed to normalization of age-related disorders in neuroendocrine regulation, immunity, carbohydrate and lipid metabolism, ovarian cyclic function, and lost reproductive potential [1,7,8]. Epithalamine reduces the intensity of free radical oxidation and improves the antioxidant status of the body [1,7,10]. Recent experiments confirmed the hypothesis that the geroprotective effect of pineal gland peptides is realized at the genetic and chromosomal levels (through regulation of expression of certain genes and restoration of their structure) [13]. Pineal gland peptides induce telomerase activity and reduce the incidence of chromosome aberrations, which seems to promote slower realization of the genetic program of aging [6,8,13].

Detection of geroprotective effects of ET in experimental studies formed the basis for studies of its efficiency and safety in elderly people with AA, which became the object of our study.

MATERIALS AND METHODS

Clinical studies of the efficiency and safety of long-term treatment with ET were carried out in 1992-2004 by scientists of Institute of Gerontology, Academy of Medical Sciences of the Ukraine, within the framework of Agreement on Scientific Cooperation with St. Petersburg Institute of Bioregulation and Gerontology. The efficiency of ET was evaluated in a thoroughly selected population of rapidly aging elderly subjects, representing an adequate object for evaluation of geroprotective treatments. All patients gave informed consent to participation in the study.

Higher functional age in comparison with calendar age was considered as the criterion of AA [2]. The examined patients suffered mainly from the cardiovascular variant of AA. This was seen from a significantly greater (by 10 and more years) functional age of the cardiovascular system CVS) in comparison with actual age. Other manifestations (syndromes) of AA were also present: autonomic dysregulation, immune system dysfunction, decreased blood levels of sex hormones below the age-specific norm, reduction of physical and mental working capacity and of compactness and mineral saturation of bone tissue, disorders in lipid and carbohydrate metabolism, reduction of the detoxicating function of the liver, increased functional age of the CNS and of structural and functional age of bone tissue, *etc.* Examined subject presented with different combinations of AA, but changes in CVS and metabolic disorders were the most significant.

Seventy elderly patients with AA and coronary heart disease were observed for 12 years. The patients were divided into 2 groups: 36 patients (aged 65±2 years) treated with ET in 1992-95 (main group) and 34 patients (aged 64±2 years) receiving saline (controls).

The patients were examined before ET or placebo treatment, after the 1st course of treatment, every year during the first 3 years, and then 5, 8, 10, and 12 years after the starting of ET treatment.

The protocol of long-term treatment with ET suggested therapeutic courses every 6 months for 3 years. Each course consisted of 5 intramuscular injections of ET in a dose of 10 mg with 3-day intervals. The dose per course was 50 mg, total dose over 3 years was 300 mg. Epithalamine was injected at 10.00, as it was experimentally proven that injection of the drug at this time of the day best of all stimulated the melatonin-producing function of the pineal gland [11].

Elderly patients in the main and control groups received the same basic therapy, including daily preventive dose of aspirin (100-125 mg), daily nitrosorbide (20-30 mg), low-dose β-adrenoblocker (propranolol, 20-30 mg), and/or angiotensin-converting enzyme inhibitor (captopril, 25-50 mg).

RESULTS

All patients well tolerated ET treatment, no cases of individual intolerance of the drug or other side effects were recorded. Positive changes in the subjective status during the course of treatment were 5.4 times more often ($p<0.05$) observed in the main group (65%) than in controls (12%). Clinical manifestations of coronary disease and AA did not augment during 3-year observation in the main group patients, while in the control group the symptoms augmented in 53% patients ($p<0.05$).

Before ET treatment, functional age of CVS in elderly patients surpassed the actual age by more than 10 years, this indicating AA. After the first 3 courses of ET therapy, the functional age of CVS decreased by 3.2±1.5 years. Subsequent courses (4-6) promoted stabilization of the attained favorable effect (Fig. 1).

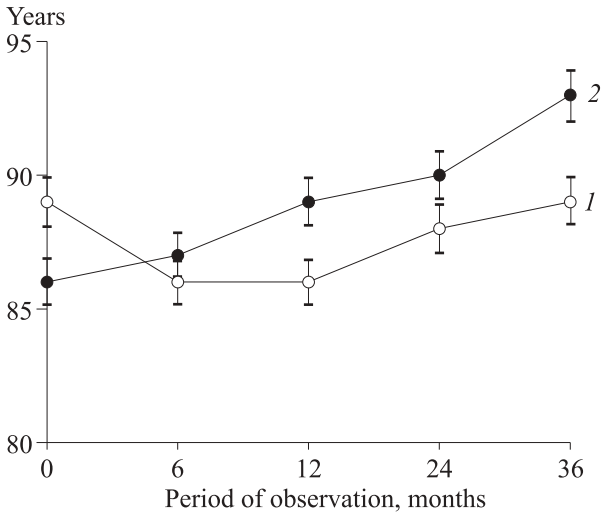

Fig. 1. Dynamics of functional age of CVS during ET treatment. Here and in Figs. 2, 3: *1*) patients treated by ET; *2*) control.

By the end of 3-year observation the functional age of CVS in the main group virtually did not differ from the initial values, despite the fact the actual age of the patients increased by 3 years during this period. In controls the functional age of the CVS increased by 7.2±3.5 years during the same period, indicating the progress of AA. The geroprotective effect of ET was more clearly seen in the dynamics of CVS aging. This parameter was determined as the difference between the functional and actual age. The degree of CVS aging decreased by 3.6±1.7 years over 3 years under the effect of ET, while in the control group it increased by 4.5± 2.2 years ($p<0.05$).

As early as after the first course of ET elderly subjects could walk a longer distance (by 254±73 m) along the terrain cure route without stopping and noted lesser fatigue during the exercise.

TABLE 1. Plasma Melatonin Concentration (ng/liter) in Elderly Patients at Different Time of the Day before and after a Course of ET (*M*±*m*)

Time o'clock	Term of measurements	Subgroup with hypo-function	Subgroup with retained function
09:00	Before treatment	4.9±1.1	15.5±6.1
	After treatment	12.1±5.8	20.6±12.3
15:00	Before treatment	4.1±1.1	6.7±1.8
	After treatment	4.9±1.0	6.8±1.2
21:00	Before treatment	15.3±3.3	66.1±20.5^{+}
	After treatment	16.3±4.8	30.3±11.5
03:00	Before treatment	24.2±5.1	149.6±42.2^{+}
	After treatment	59.0±12.6*	75.0±37.6

Note. $^{+}p<0.05$ compared to subgroup with pineal gland hypofunction; *$p<0.05$ compared to values before ET treatment.

The results of bicycle ergometry confirmed increased physical performance ability (Fig. 2). The threshold exercise tolerance increased by 21% under the effect of ET. This was paralleled by increase in the maximum O_2 consumption during performance of the threshold exercise (by 0.11± 0.05 ml/min), indicating increased functional potential of the oxygen transporting system. Positive effect of AA on physical working capacity was observed in 58% patients, *vs.* only 7% (8-fold less) in control group ($p<0.05$).

One of the mechanisms of favorable effect on physical working capacity was improvement of the efficiency of the CVS functioning (economic work). Heart rate and systolic blood pressure decreased significantly at the peak of dosed (25 W) exercise (by 7±3 bpm and by 9.4 mm Hg, respectively), while stroke volume increased by 12±4 ml, indicating decreased energy expenditure of the heart. Epithalamine had a favorable impact on the autonomic regulation. The treatment promoted a decrease in the low-frequency component and increase in the intensity of high-frequency component of heart rhythm variability, that is, decreased the sympathetic and increased the influences on CVs. This restructuring of autonomic regulation provides more effective functioning of the CVS during exercise.

The results indicate stable increase of exercise tolerance in elderly subjects with AA after regular (twice a year) ET treatment. Age-associated disorders in physical performance ability progressed in the controls. A 3-year observation witnessed a further decrease in the intensity of tolerated exercise.

The favorable impact of ET on aging organism is partially due to its normalizing effect on the melatonin-producing function of the pineal gland [3]. Epithalamine stimulated endogenous production of melatonin in patients with initially low function of pinealocytes: plasma hormone concentration moderately increased during the dark hours of the day (Table 1). Injection of ET to patients with retained melatonin-producing function of the pineal gland virtually did not suppress the production of endogenous melatonin. Therefore, prescription of ET does not require preliminary evaluation of the pineal gland function.

By the end of the period of observation 19 (55.9%) of 34 controls and 28 (77.7%) of 36 patients receiving ET were alive (Fig. 3), that is, the total mortality decreased by 28% ($p<0.05$). This indicates a favorable impact of ET on the long-term

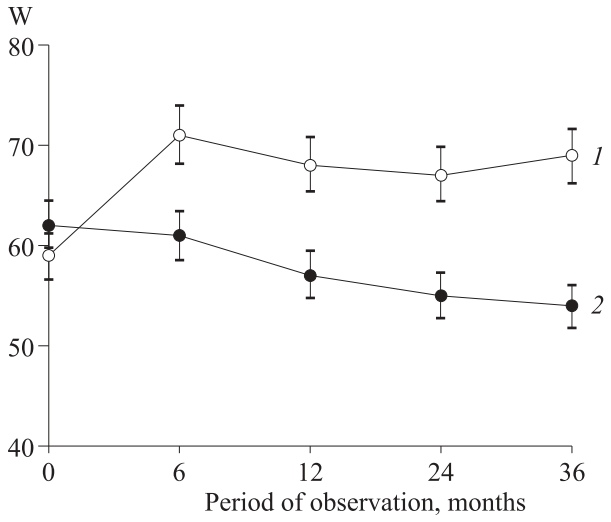

Fig. 2. Dynamics of threshold exercise intensity during ET therapy.

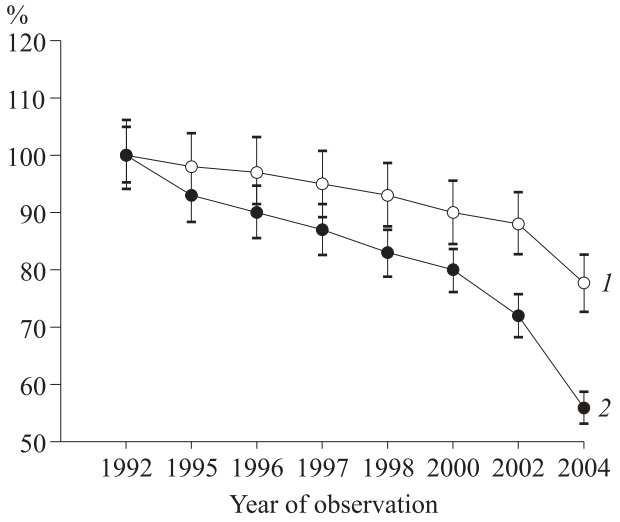

Fig. 3. Survival dynamics in coronary patients: 12-year observation.

prognosis in elderly patients with AA. In the control group 12 (35.3%) of 34 patients died from cardiovascular diseases, *vs.* only 6 (16.7%) of 36 in the main group. Hence, ET therapy reduced cardiovascular mortality virtually 2-fold ($p<0.05$).

The incidence of nonfatal events was also lower in elderly patients treated by ET. More severe course of coronary disease (frequent, lasting, and more intense attacks of angina pectoris, urgent hospitalization because of coronary disease exacerbation, development of myocardial infarction) was observed in 58.8% controls and 36.1% patients in the main group. Cardiac failure more often developed in controls (35.2% *vs.* 19.4% in the main group). Patients of the main group had 2.4 times less respiratory episodes during ET course in comparison with the controls. These results indicate a favorable effect of regular ET courses on the development of nonfatal cardiovascular and other events in elderly subjects with AA.

Experience gained in many-year treatment of elderly patients by ET indicates that pineal peptide preparations should be used in gerontology as a means preventing AA. In geriatrics the peptide preparations can be included in combined therapy of patients with cardiovascular, respiratory, CNS, and locomotor system diseases, for repair of immune system dysfunctions frequent in elderly and senile age, for correction of lipid and carbohydrate metabolism, for improving stress resistance, and for correction of disturbed 24-h biorhythms.

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