Seasonal Effects of Selank on the Behavior of Hibernating Animals

T. P. Semenova, M. M. Kozlovskaya*, A. V. Zuikov, I. I. Kozlovskii*, and L. A. Andreeva**

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 140, No. 12, pp. 658-660, December, 2005
Original article submitted April 6, 2005

We studied the seasonal effects of peptide preparation Selank on orientation and exploratory activity in 36 arctic ground squirrels *Citellus undulatus* in the open-field and hole-board tests. Selank most significantly increased behavioral activity of hibernating animals in the spring and fall. The test peptide had no effect on locomotor activity of animals. The selective stimulatory effect of Selank on exploratory behavior of hibernating animals was season-dependent. The maximum effect was observed under conditions of seasonal depression-like state.

Key Words: *Selank; exploratory behavior; stimulatory component of activity spectrum; hibernating animals; seasonal effects*

Selank is an original neurotropic peptide preparation synthesized at the Institute of Molecular Genetics and studied at the Institute of Pharmacology. This synthetic heptapeptide consists of a tetrapeptide tuftsin (Thr-Lys-Pro-Arg) and 3 natural L-amino acids Pro-Gly-Pro. The presence of Pro-Gly-Pro group increased the resistance of the synthetic tuftsin derivative to adverse effects of external and internal factors [3]. Experiments on animals with genetically determined strategy of active or passive behavior in a stress situation demonstrated anxiolytic and nootropic properties of Selank [3, 4,6]. Single administration of Selank induced long-term shifts in the concentrations of norepinephrine, dopamine, serotonin, and their metabolites in the brain of Wistar rats [4,5]. Published data show that biogenic amines are involved in the regulation of biological rhythms [1]. Therefore, it is important to evaluate seasonal effects of Selank on integrative activity of the brain in animals.

Institute of Cell Biophysics, Russian Academy of Sciences, Pushchino; *V. V. Zakusov Institute of Pharmacology, Russian Academy of Medical Sciences; **Institute of Molecular Genetics, Russian Academy of Sciences, Moscow. ***Address for correspondence:*** tat_semenova@icb.psn.ru. T. P. Semenova

Hibernating animals serve as a unique natural model for studies of seasonal neuropsychotropic activity and mechanisms underlying the effect of pharmacological preparations. Hibernation involves various levels of organization from biochemical (seasonal changes in the concentration of neurotransmitters, peptides, and hormones) to behavioral [10,11]. The transition phase between wakefulness and hibernation lasts 1.5-2 months and begins from changes in integrative activity of the brain. It manifests in impairment of cognitive function and decrease in emotional resistance and orientation and exploratory and locomotor activity [8]. These changes in functional activity of the central nervous system (CNS) are typical of seasonal depression-like state.

Here we studied the seasonal stimulatory neurotropic effect of Selank on hibernating animals.

MATERIALS AND METHODS

Experiments were performed on 36 adult male and female arctic ground squirrels *Citellus undulatus* weighing 600-800 g. The animals were kept in individual cages (35×40×20 cm) and received dry feed, fresh vegetables, grass, and water *ad libitum*.

In the middle of October these animals were placed in a special room for hibernation (4°C) and maintained there until awakening (middle of April). The study was conducted in a laboratory room at constant temperature (15°C). We performed 3 series of experiments: series I, middle of March (period of awaking between bouts of sleep, end of hibernation); series II, end of June (maximum activity of animals); and series III, beginning of October (pre-hibernation period). Each series was performed on 2 animal groups. Control animals received physiological saline (control). Group 2 animals received intraperitoneal injections of 300 mg/kg Selank 30 min before the experiment.

Exploratory behavior was studied in the open-field and hole-board test. The open field was a brightly illuminated area (1 m^2) divided into 100 radial squares. The hole board was a diffusely illuminated arena with 16 holes in the floor (diameter 2.5 cm). We recorded horizontal (number of crossed squares) and vertical exploratory activity (number of rearing postures) in the open-field test. Vertical orientation and exploratory activity and the number of explored holed were determined in the hole-board test. The behavior of each animal was studied over 10 min (evening time, 18.00-22.00). Locomotor activity was recorded in an Animex device. The results were analyzed by Student's *t* test (Excel 97 and Sigma Plot 4.0 software).

RESULTS

Single administration of Selank in spring had a stimulatory effect on orientation and exploratory behavior of animals in the open field. After peptide treatment the number of crossed squares and vertical rearing postures increased by 2.3 and 13.7 times, respectively (Table 1). The number of rearing postures in the hole board test increased by 1.5 times (Table 2). Most parameters of exploratory activity remained practically unchanged in summer (Table 2). Administration of Selank in the fall was followed by a significant increase in horizontal and vertical exploratory activity of animals in the open field (by 3.1 and 3.7 times, respectively, Table 1). The test parameters in the hole board increased by 4.4 and 4.7 times, respectively (Table 2). The peptide had no effect on locomotor activity of animals.

The stimulatory effect of Selank on exploratory behavior of hibernating animals is manifested during the transitional seasons. The psychotropic effect of Selank on behavior of animals depended on the season. Functional activity of CNS in hibernating animals during spring and fall is characterized by an imbalance between monoaminergic systems [2]. Monoamines serotonin, norepinephrine, and dopamine play a key role in the regulation of selective attention, learning, memory, and emotional resistance of animals and human to stress [4,9]. Administration of this heptapeptide produced a tranquilizing and stimulatory effects, improved emotional behavior, and normalized the ratio between monoamines in brain structures of Wistar rats treated neonatally with 6-hydroxydopamine. The stimulatory effect of Selank on exploratory behavior of hibernating animals is most pronounced under conditions of an imbalance between monoaminergic systems in the brain (spring and fall). The results

TABLE 1. Seasonal Effects of Selank on Exploratory Behavior of Squirrels in the Open Field (n=6, $M\pm m$)

Season	Number of squares		Number of rearing postures	
	control	Selank	control	Selank
Spring	22.6±10.5	50.0±12.2*	1.0±0.6	2.70±0.42**
Summer	28.0±8.5	35.8±9.0	1.3±0.5	0.7±0.5
Fall	17.7±6.3	54.5±11.6*	1.0±0.4	3.7±1.6**

Note. Here and in Table 2: *p<0.05 and **p<0.01 compared to the control.

TABLE 2. Seasonal Effects of Selank on Exploratory Behavior of Squirrels in the Hole Board Test (n=6, $M\pm m$)

Season	Number of rearing postures		Number of explored holes	
	control	Selank	control	Selank
Spring	0.7±0.2	1.5±0.6*	1.2±0.7	3.2±0.4*
Summer	0.2±0.2	0.5±0.3	1.7±0.3	2.8±0.9
Fall	0.5±0.3	2.2±0.9*	1.0±0.8	4.7±1.5*

of studying functional activity of CNS and seasonal variations in the stimulatory effect of Selank on behavior of hibernating animals can be used for the development of new methods for pharmacological correction of seasonal changes in adaptive behavior.

This work was supported by the Russian Foundation for Basic Research (grant No. 04-04-49098).

REFERENCES

1. I. A. Anoshkina, T. P. Semenova, L. P. Dolgacheva, and S. G. Kolaeva, *Neirokhimiya*, **20**, 259-264 (2003).
2. A. I. Anufriev and A. K. Akhrimenko, *Ecological and Physiological Characteristics of Natural Hypometabolic States* [in Russian], Pushchino (1992), pp. 34-38.
3. E. B. Arushanyan, *Chronopharmacology* [in Russian], Stavropol (2000).
4. M. M. Kozlovskaya, I. I. Kozlovskii, E. A. Val'dman, and S. B. Seredenin, *Fiziol. Zh*., No. 4, 264-268 (2002).
5. T. P. Semenova, *Optimization of Learning and Memory* [in Russian], Pushchino (1992).
6. T. P. Semenova, *Zh. Vyssh. Nervn. Deyat*., **53**, No. 2, 358-361 (2002).
7. S. B. Seredenin, M. M. Kozlovskaya, Yu. A. Blednov, *et al.*, *Ibid*., **48**, No. 1, 153-160 (1998).
8. S. B. Seredenin, T. P. Semenova, M. M. Kozlovskaya, *et al*., *Khim.-Farm. Zh*., **30**, No. 5, 12-14 (1996).
9. E. A. Gromova, A. R. Chubakov, and T. P. Semenova, *Signal Molecules and Mechanisms of Animal Behavior*, Eds. W. Winlow *et al*., New York (1991), pp. 290-293 (1991).
10. F. Nurnberger, *Cell Tissue Res*., **281**, 391-412 (1995).
11. N. K. Popova, T. P. Voronova, and A. V. Kulikov, *Pharmacol. Biochem. Behav.*, **46**, 9-13 (1993).
12. T. P. Semenova, I. A. Anoshkina, B. M. Khomut, and S. G. Kolaeva, *Behav. Processes*, **56**, 195-200 (2001).