TIME COURSE OF THE ACTION OF TUFTSIN ON BEHAVIOR

A. A. Kamenskii, V. N. Kalikhevich,
and N. Yu. Sarycheva

UDC 615.276.4.015.4:612.821.1/.3

KEY WORDS: peptides; tuftsin; anxiety response

Tuftsin,* an endogenous stimulator of immunity, has been shown to possess neurotropic activity; in particular, injection of tuftsin in doses of 0.15 to 0.5 mg/kg body weight stimulates motor activity in rats, lowers the pain threshold, and increases aggressiveness [1, 5]. It has also been shown that tuftsin subsequently changes its direction of action. The neurotropic effect of tuftsin is not in dispute, but data on the character of the behavior changes, their times and durations, are contradictory.

The aim of this investigation was to study the time course of the action of tuftsin on behavioral responses.

EXPERIMENTAL METHOD

Experiments were carried out on noninbred male albino rats weighing 150-200 g. Tuftsin was synthesized in the Department of Chemistry of Natural Compounds, Leningrad University. The peptide was injected into the animals in aqueous solution intraperitoneally in a dose of 0.3 mg/kg body weight. Control animals received an injection of distilled water.

Motor activity was measured on Animex and Opto-Varimex apparatuses (USA) in darkness and under conditions of quietness.

To study the orienting-investigative activity of the animals and parameters reflecting emotional changes, the open field (OF) method [7] was used in two modifications: stressor (lights switched on, bells ringing) and nonstressor (quietness, red light). The results were subjected to statistical analysis by the Mann–Whitney–Wilcoxon test [4].

EXPERIMENTAL RESULTS

Changes in motor activity under the influence of tuftsin were studied on the "Animex" apparatus. The peptide was injected immediately before the experiment and 4, 12, 24, 48, 72, and 120 h previously. Enhancement of locomotion was found to be of short duration, lasting 20-30 min after injection (Fig. 1).

The observations made on the Animex apparatus were confirmed in the nonstressor modification of OF. An increase in the length of the run ($P < 0.05$), amounting to 155% of the control value, an increase in the number of standings, and a decrease in the number of defecations to 30% were observed 5 min after injection of tuftsin in a dose of 0.3 mg/kg. When tuftsin was injected 4, 12, or 24 h before the experiment, the length of the run was reduced by 20%. The experimental animals ventured into the center of OF 20% fewer times. These changes stopped 48 h after the injection.

When the stressor modification of OF was used tuftsin caused no changes in motor activity, but there was an increase in the number of groomings. The excitatory effect of tuftsin thus lasted about 30 min. This was followed by a small decrease in motor activity, which continued for 1 day.

Since the excitatory action of tuftsin was manifested more strongly in the nonstressor modification of OF, when the rats' behavior was determined to a greater degree by the orienting-investigative response, we postulated that tuftsin potentiates this response. To confirm this hypothesis the "Opto-Varimex" apparatus, which records horizontal and vertical activity

*Thr-Lys-Pro-Arg tetrapeptide.

Complex Laboratory, Faculty of Biology, M. V. Lomonosov, Moscow University. (Presented by Academician of the Academy of Medical Sciences of the USSR I. P. Ashmarin.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 101, No. 1, pp. 55-57, January, 1986. Original article submitted April 19, 1985.

 0007-4888/86/0001-0070$12.50

TABLE 1. Response of Rats to Frightening Stimuli after Injection of Tuftsin (0.3 mg/kg)

Experimental conditions	Number of animals	First stimulus		Second stimulus	
		Time, sec			
		Before startle	Startle	Before Startle	Startle
Control	7	2	54	2	111
Tuftsin (0.3 mg/kg)	7	20	37	18	25
				$P<0,05$	$P<0,05$

TABLE 2. Startle Response to Frightening Stimulus (in % of total number of animals in group)

Experimental conditions	Number of animals	Time after injection, min			
		30	35	40	45
Injection of water	13	17	31	17	31
Injection of tuftsin (0.3 mg/kg)	13	31	46	54	62
			$P<0,05$	$P<0,05$	$P<0,05$

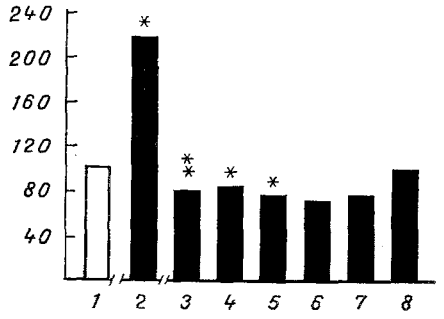

Fig. 1. Motor activity of rats measured by "Animex" apparatus for 30 min at different times after injection of tuftsin. Ordinate, activity (in % of control). 1) Control; 2-8) injection of tuftsin 5 min and 4, 12, 24, 48, 72, and 120 h before experiment. *P < 0.05, **P < 0.01 compared with control.

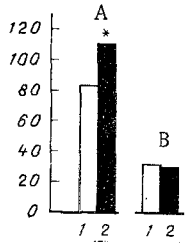

Fig. 2. Vertical motor activity of control rats (1) and immediately after injection of tuftsin (2), measured with the "Opto-Varimex" apparatus. Ordinate, activity (in relative units). A) Intact animals; B) after preliminary adaptation. *P < 0.05 compared with control.

separately, was used. Injection of tufstin caused an increase in the animals' vertical activity (number of standings), which continued for 30 min after injection ($P < 0.05$).

An experiment was carried out with the same apparatus on animals whose orienting-investigative response was extinguished beforehand. Extinction was induced by keeping the animal for 30 min under conditions identical with those of the experiment, followed by the experiment itself. It was found that horizontal activity of the pre-adapted rats remained at its previous level, whereas vertical activity, which reflects to a greater degree the orienting-investigative response, was considerably reduced. Injection of tuftsin into the adapted animals caused no changes in either type of motor activity (Fig. 2).

Injection of tuftsin thus potentiates the orienting-investigative response, but when that is extinguished beforehand, the peptide does not restore it. These observations are in agreement with the results of investigations which revealed excitation under the influence of tuftsin [1-3] and the effect of the peptide on orienting-investigative behavior when injected in a dose of 0.5 mg/kg [3].

It was shown previously that the action of tuftsin on behavior is linked with an increase of emotional tension during stress [2]; on a model of behavioral depression, in an unavoidable stressor situation, tuftsin also had an activating effect [3].

The tendency toward an increase in the length of run in the stressor modification of OF in response to injection of tuftsin is evidence of a shorter duration of starting of the rats,

i.e. possible weakening of anxiety-linked responses or of predominance of the active-defensive over the passive-defensive response. This explanation is supported by the reduction in the number of defecations in the animals receiving tuftsin. The action of the peptide on response associated with the emotion of anxiety also was demonstrated previously [1].

To confirm the view that tuftsin depresses the anxiety response, the response of rats to fright (bright light, ringing of bells) was studied in the Animex apparatus. The rats were adapted to the appparatus for 15 min, after which they were given tuftsin or water, and 5 min later they were exposed to frightening stimulation (a combination of bright flashes of light and the ringing of a bell), three times in succession, for 1 sec each time. The frightening stimulation was repeated after 5 min (Table 1).

Table 1 shows that the response to fright was considerably weaker after injection of tuftsin: In the experimental rats the period of motor activity preceeding the period of total immobility (startle) was longer, and the period of startle was shorter. In addition, injection of tuftsin caused disappearance of the startle response in three of seven animals.

After about 30 min the picture changed: Whereas the control animals responded only a little to frightening stimulation, the experimental rats continued to respond to fright, and the number of startled rats actually increased (Table 2).

Tuftsin thus has a biphasic action on the behavior of albino rats, which is revealed in a wide range of tests. The first phase lasts about 30 min and is characterized by intensification of investigative behavior and weakening of behavioral manifestations associated with anxiety, or predominance of active defensive responses to fright. Next follows a period of reduced motor activity and depression of the orienting reaction, which lasts at least 24 h. This phase is accompanied by intensification of responses which demonstrate enhancement of passive-defensive response. The second phase coincides in time with changes in activity of enzymes of energy metabolism in peripheral blood lymphocytes, reflecting the ability of these cells to take part in genesis of the immune response [6].

LITERATURE CITED

1. S. M. Andreev, L. V. Antonova, I. P. Ashmarin, et al., Dokl. Akad, Nauk SSSR, 233, No. 2, 498 (1980).
2. A. V. Val'dman, N. A. Bondarenko, M. M. Kozlovskaya, et al., Byull. Éksp. Biol. Med., No. 4, 49 (1982).
3. A. V. Val'dman, M. M. Kozlovskaya, I. P. Ashmarin, et al., Byull. Éksp. Biol. Med., No. 7, 31 (1981).
4. E. V. Gubler and A. A. Genkin, The Use of Nonparametric Statistical Tests in Medico-Biological Research [in Russian], Leningrad (1973).
5. A. A. Kamenskii, L. V. Antonova, N. A. Samoilova, et al., Byull. Éksp. Biol. Med., No. 7, 43 (1980).
6. M. B. Shishova, A. A. Kamenskii, V. V. Meretskov, and N. Yu. Sarycheva, Byull. Éksp. Biol. Med., No. 5, 55 (1983).
7. C. Hall, J. Comp. Psychol., 22, 345 (1936).