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J. MROWIEC [1], A. PLECH [1], B. RZESZOTARSKA [2],
F. RYSZKA [3], R. BRUS [1], E. MASIUKIEWICZ [2]

BEHAVIOURAL EFFECTS OF LUTEINIZING HORMONE-RELEASING HORMONE (LHRH) IN RATS

[1] Department of Pharmacology of Medical Faculty in Zabrze, Silesian Medical Academy of Katowice,
[2] Department of Organic Chemistry, Pedagogical University of Opole,
[3] Department of Applied Pharmacy and Drug Technology of Pharmaceutical Faculty in Sosnowiec, Silesian Medical Academy of Katowice.

Behavioural effects of intracerebroventricularly-injected (icv) LHRH were studied in female rats. Locomotor and exploratory activities as well as irritability were determined. A pronounced inhibitory effect of 10 μg doses of LHRH was found. At 100 μg doses of LHRH, barrel behaviour was observed. We conclude that LHRH can modify the activity of central serotonergic receptors in rats.

Key words: *LHRH, rat, behavioural effects of LHRH, behaviour, exploratory activity, locomotion, irritability, barrel behaviour.*

INTRODUCTION

Luiteinizing hormone-releasing hormone (LHRH), isolated from the porcine hypothalamus is of a key importance in the regulation of the synthesis and release of gonadotropins LH and FSH. This decapeptide hormone is synthesized in hypothalamic neurosecretory cells and released into hypothalamo-pituitary portal vessels (1, 2). LHRH fibres were found within the forebrain in both hypothalamic and extrahypothalamic regions: the medial and lateral preoptic and anterior hypothalamic areas, diagonal band of Broca, septum and within circumventricular organs: the organum vasculosum of the lamina terminalis also in the subfornical organ in olfactory bulb and in the hippocampus. Moreover, some projections from these structures to the median eminence have been identified (3—6).

LHRH in addition to well-defined endocrine role affects CNS-controlled functions. LHRH has been known long time to induce sexual behaviour of hypophysectomized and ovariectomized female rats (7, 8) and a great deal of effort has been devoted to the elucidation of the regulation of this behaviour (9, 10). Recently has LHRH been observed to impair conditioned avoidance behaviour (11). Several lines of evidence indicate the interaction of LHRH with dopamine, noradrenaline, and histamine (11—16, see also 9). There are also premises showing such an interaction with serotonine (5—HT) (12, 16).

In the present study we examined the effects of central administration of LHRH on locomotor and exploratory activities as well as irritability of rats. In our previous work (16), the increase in 5-HIAA level was found in all the investigated parts of rat's brain after intracerebroventricularly-injected (icv) LHRH. It has been attributed to the accelerated 5-HT metabolism resulted from the increased activity of central serotonergic neurons. Therefore, we also examined the effects of the icv LHRH preceded by pizotifen (PI), an effective blocker of central postsynaptic serotonergic receptors (17, 18). For preliminary results see ref. 19.

MATERIAL AND METHODS

Subjects

Wistar female rats (190—200 g), obtained from Animal Farm of Silesian Medical Academy were housed in a reverse 12 h : 12 h light-dark cycle (light from 6 am to 6 pm) with free access to food and water. 24 h before the experiment, rats were prepared for intracerebroventricular injections according to Herman (20) by drilling a hole under light ether-anaesthesia in the skull 2 mm to right from sagittal suture and 2 mm posterior from coronary suture.

Molecular tools

LHRH was synthesized in Department of Organic Chemistry of Pedagogical University of Opole, Opole and its characteristic is given in ref. 21. Pizotifen came from Sandoz.

Treatment

LHRH either at a 10 μg or 100 μg dose, dissolved in 10 μl of 0.9% NaCl was injected to non-anaesthetized rats always at the same time of day between 9 am and 1 pm by means of a Hamilton microsyringe introduced through the

hole into the right lateral cerebral ventricle (icv) on a depth of 4 mm from the surface of the skull. The entire injection lasted 2 min and subsequently the empty syringe was still kept in the brain for 1 min. Pretreatment with 0.5 mg/kg dose of pizotifen was made ip, 30 min before the administration of LHRH. Control animals receiving only 10 μl of 0.9% NaCl were handled in the same way as experimental groups.

The examination of rat's behaviour

The collecting of data was started 1, 15, 30 and 60 min post injection. A locomor activity was determined in the open field text according to Janssen et al (22) by recording a number of episodes of ambulation per 3 min. Simulatneously, an exploratory activity was measured by recording a number of peepings. Afterwards, the exploratory activity was examined in the hole text following File (23, 24) and recording a number of head dips into a board hole per 3 min. After finishing the hole text, an irritability was investigated by means of the score of Nakamura and Thoenen (25). At the end of an experiment, animals were anaesthetized by hexobarbital (150 mg/kg, ip) and methylene blue solution was icv injected.

Statistics

Results were elaborated according to Dunnett and significant ANOVA.

RESULTS

LHRH at 10 μg doses did not change rat's behaviour measured in the open field test, 1 min post injection (Fig. 1 a), while it significantly decreased the incidences of peeping at 15, 30 and 60 min, (Fig. 1 b—d) and the incidences of ambulation at 30 and 60 min (Fig. 1 c and d). Pretreatment of animals with PI (0.5 mg/kg, ip, 30 min before the administration of LHRH) did not prevent the decrease of a number of those episodes (Fig. 1 a and d).

LHRH at 100 μg doses produced, immediately after administration, atypical effect, viz., barrel rotatory behaviour lasting 15 min and followed by catalepsy and immobility for 20 min. This phenomenon was prevented by PI at 0.5 mg/kg dose injected ip 30 min before LHRH. Moreover, the considerable decrease of the incidences of ambulation and peeping was observed at 60 min when rat's rotatory behaviour completely disappeared and those latter effects were not blocked by PI (Fig. 1 e).

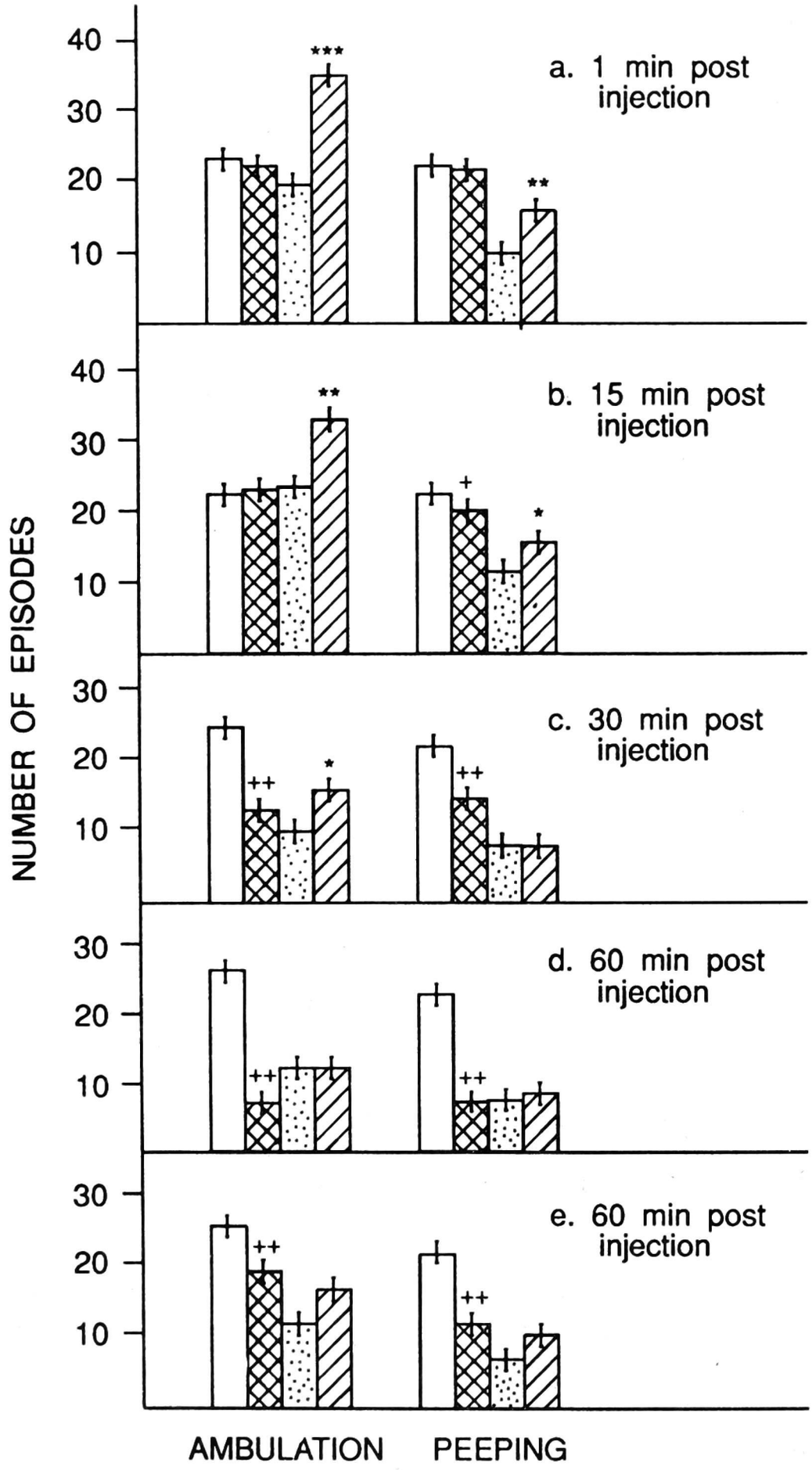

Fig. 1. Effect of icv LHRH on rats's behaviour measured in the open field test (22). ☐ icv 0.9% NaCl (n = 8) ▨ icv LHRH (a—d 10 μg; e 100 μg) (n 8) ▩ ip PI 0.5 mg/kg and after 30 min icv 0.9% NaCl (n = 10) ▨ ip PI 0.5 mg/kg an after 30 min icv LHRH (a—d 10 μg; e 100 μg) (n = 10). +$p<0.05$ and ++$p<0.001$ vs. saline treated controls; *$p<0.05$, **$p<0.01$, and ***$p<0.001$ vs LHRH treated group.

LHRH at 10 μg doses did not change the exploratory activity measured in the hole test, 1 and 15 min post injection (data not shown), while the significant decrease of the activity occured at 30 and 60 min and this effect was not blocked by PI (Fig. 2a and b). Neither did 100 μg dose of LHRH afect rat's exploratory activity recorded 60 min post injection (data not shown).

LHRH at 10 μg doses was inert for rat's irritability, 1, 15 and 30 min post injection (data not shown). It took 60 min before the considerable decrease of irritability appeared and this phenomenon was blocked by PI (Fig. 2c). However, LHRH at a high dose of 100 μg did not change animal's irritability measured 60 min post injection.

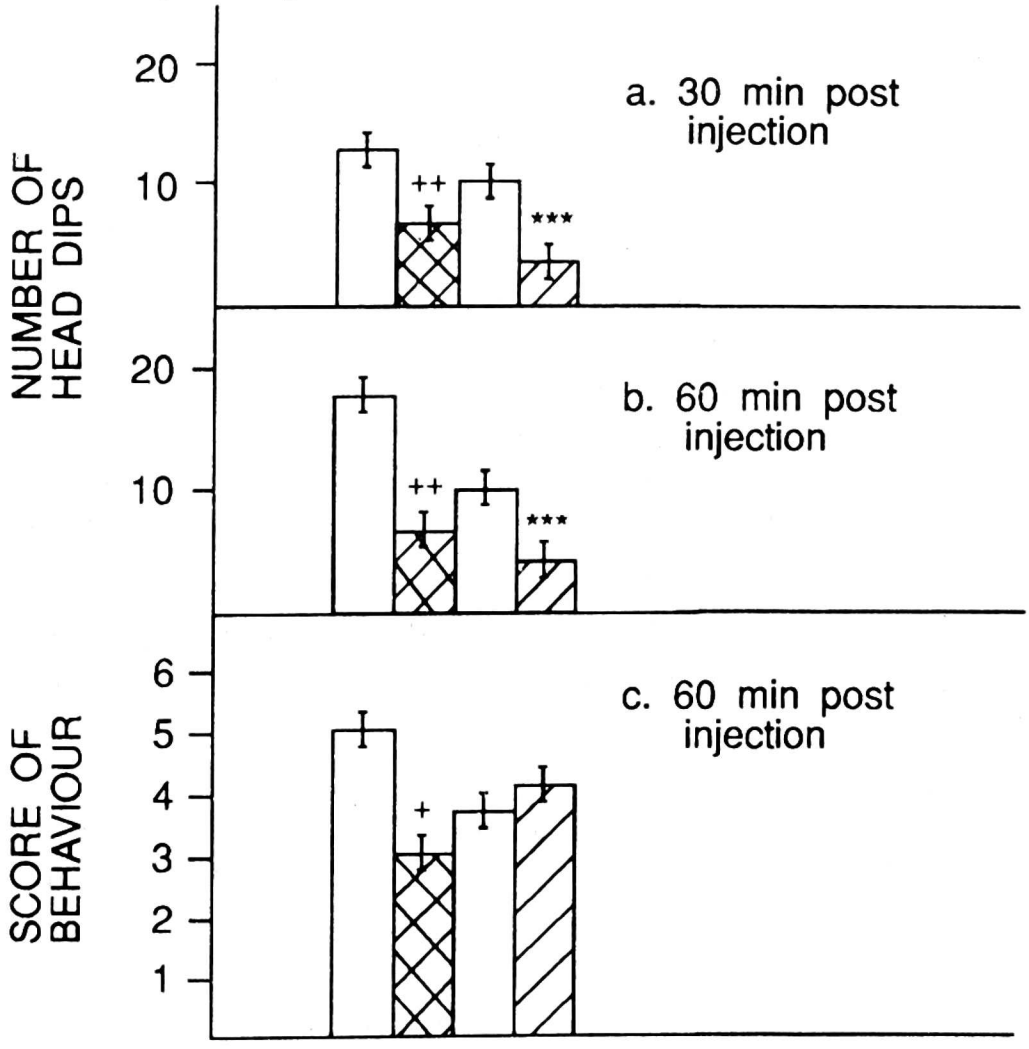

Fig. 2. Effect of icv LHRH on rat's behaviour measured in the hole test (23, 24) (a and b) and by means of Nakamura and Thoenen score (25) (c). For the legend see Fig. 1.

DISCUSSION

Obtained data show that LHRH affects rat's CNS, resulting in changes of behaviour. Viz., at 10 μg dose, the decrease of locomotor and exploratory activities as well as irritability is observed and at 100 μg dose, the induction of barrel behaviour takes place.

The pronounced tendency for depressing the listed-above forms of rat's behaviour is in a line with results of Nasello et al as they observed LHRH-induced inhibition of rat's conditioned avoidance behaviour (11). The latter seems to be, at least in part, brought about by the inhibition of exploratory and locomotor activities. Wolny and Herman previously found the increased exploratory and locomotor activities in a phase of proestrus and estrus in a sexual cycle of intact female rats (26). The increased portal plasma level of LHRH and blood plasma levels of LH, estradiol-17 β and progesterone have been determined in this phase (27). Our results may imply that not only gonadal hormones and gonadotropins but also LHRH takes part in the regulation of behaviour of female rats in the course of a sexual cycle.

A 100 μg LHRH dose causes immediately barrel behaviour. We consider it a toxic effect. A similar barrel behaviour occured in rats also after icv injections of some other neuropeptides dynorphin, tuftsin, and vasopressin and was connected, as blocked by haloperidol, with the stimulation of central dopaminergic receptors (28—30). The LHRH-induced barrel behaviour is associated with the stimulation of central serotonergic receptors as it is blocked by pizotifen, an effective blocker of central postsynaptic serotonergic receptors (17, 18). Moreover, the same ivc (100 μg) LHRH doses were found in our earlier study to increase significantly the level of 5-HIAA, the main cerebral metabolite of 5-HT, in the all investigated parts of the brain, cortex, striatum, hypothalamus, and medulla oblongata (16).

In conclusion, the present results indicate that LHRH adinistered icv to rats affects their CNS and can modify the activity of the central serotonergic system.

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Author's address: A. Plech, Department of Pharmacology, Medical Faculty in Zabrze, Silesian Medical Academy of Katowice, K. Marksa Str. 38, 41-808 Zabrze, Poland