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ORIGINAL ARTICLE

Possible Central Role of Oxytocin in Neurobehaviour of Male Rats and Mice

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ABSTRACT

Oxytocin a nonapeptide of molecular mass 1007 daltons secreted in the paraventricular and supraoptic nuclei of the hypothalamus. Its receptors are widely distributed in the central nervous system. However, the behavioral effects of centrally administered oxytocin have received little attention because of availability of less information about its role in male psychobehavior. In the present study, we tested the effects of ICV injections of oxytocin on exploratory behavior in male rats and mice adopting well established open field test. The effects were studied by considering latency time to explore, grooming, rearing, horizontal movement, defecation and urination as parameters in exploratory behavioral studies. Administration of 2ml per animal graded doses of oxytocin (1mg/ml, 2mg/ml and 4mg/ml) provoked significant ($P<0.01$) increase in exploratory behavior by both rats and mice in open field test. Taken together, the present study demonstrates that central injection of oxytocin in male rats and mice in doses (1mg/ml, 2mg/ml and 4mg/ml) enhance exploration and improves social interaction, suggesting that oxytocin may be involved in some aspects of male psychosocial behavior.

Key words: oxytocin, i.c.v, exploration, male behavior, open field test.

Introduction

Oxytocin was the first peptide hormone to have its structure determined and the first to be chemically synthesized in biologically active form[4]. The actions of oxytocin range from the modulation of neuroendocrine reflexes to the establishment of complex social and bonding behaviors related to the reproduction and care of the offspring[7]. In response to variety of stimuli such as suckling, parturition or certain kinds of stress the processed oxytocin peptide is released from the posterior pituitary into the systemic circulation[15]. Such stimuli also lead to an intranuclear release of oxytocin. Oxytocinergic neurons display wide spread projection throughout the central nervous system, it's central and peripheral actions are mediated through the receptors called as 'OT' receptors which are typically class I G protein coupled receptors coupled viz Gq and G11α. Oxytocin has long been considered to be restricted to stimulation of uterine contractions during labor and milk ejection during lactation[9,20]. Over the past decade the central actions of oxytocin has been intensively studied revealing the profound regulation by steroids. The regulation by gonadal and adrenal steroids is one of the most remarkable features of the oxytocin system and is unfortunately the least understood[13].

An equivalent concentration of oxytocin is found in the neurohypophysis and plasma of both sexes[26] suggests that oxytocin has further physiological functions. In all species oxytocin and vasopressin are on the same chromosomal locus but are transcribed in opposite directions[21,10]. In rats, OT receptors

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are abundantly present in several brain regions, i.e., some cortical areas, the olfactory system, the basal ganglia, the limbic system, the thalamus, the hypothalamus, the brain stem, and the spinal cord. In the adult rat, a high density of OT receptors is found in the dorsal peduncular cortex, the anterior olfactory nucleus, the islands of Calleja and ventral pallidum cell groups, the limbic system (bed nucleus of the stria terminalis, central amygdaloid nucleus, ventral subiculum), and the hypothalamic ventromedial nucleus[2,32]. OT receptor mRNA was detected in brain areas mostly coinciding with the occurrence of OT binding sites[34]. OT receptors were detectable in all spinal segments, but in low amounts and restricted to the superficial layers of the dorsal horn[31]. No major differences in receptor distribution were observed between male and female brains. The systemic oxytocin hormone could act peripherally stimulating smooth muscle cells of the male reproductive tract but could also reflect central effect in the brain in modulating social behavior. Oxytocin is identified in the testis from various mammalian species concerning the localization of the oxcytocinergic system in the male reproductive system. It has also been found in prostate gland, epididymis, Leydig cells and involved in the synthesis of testosterone, regulation of somniferous tubule contractility and modulation of steroidogensis[24]. The role oxytocin has been studied in various activity mediate centrally as well as peripherally including analgesic, male sexual behavior, female sexual behavior, hypertension, milk ejection, hemorrhage, stress and natriuretic[17,19]. Recently, it has been reported that centrally administered oxytocin can induced or modify several forms of behavior together with associated motor sequences[3]. It has also been extensively studied for its role in aggressiveness, anxiety, fear, and stress in lactating mothers. However its role in important male psychobehaviors viz anxiety, fear, curiosity and stress have not been studied till the date. Inspite of equal distribution of oxytocin receptor in male and female brains, hence this study was visioned.

It is well known that oxytocin is secreted in neurohypophysis in both male and female, have a significant role in control of behavior but its role in male behavior has not been studied in detail as much as studied in the behavior of females. In male oxytocin is made locally within the testis and also possibly in epididymis and present in a interstitial Leydig cells as a main source of testosterone[14,23]. It has been proven that central administration of oxytocin causes spontaneous erection in rats, increased mounting behavior and parental behavior by acting through its receptors[26,9]. However, its role in social interaction, understanding, anxiety, stress and fear has not been well established. Therefore the present study was designed to evaluate and to find out the central importance of oxytocin in combating the most common male psychological disorders anxiety, fear, stress and exploratory behavior by using well established hole board test, elevated plus maze, open field test in preclinical models. This study not only expands the existing knowledge on oxytocin but also explores possibility of its use in clinical psychobehavior abnormalities.

The present study was carried out with an objective to unveil central role of oxytocin in male rat and mice behavior. This study will expand the knowledge existing on oxytocin and its role in male behavior including anxiety, stress, fear and exploration.

Materials and methods

Materials

Oxytocin was purchased from Fluka, biochemia, product of Switzerland and Unitek Hamilton syringe (10 µl) was purchased from Unitek Scientific Corporation; Mumbai, Hole-board, Elevated plus maze and Open-field instruments were fabricated locally based on earlier standard literatures[6,16,22].
Animal selection

Male wistar albino rats weighing 180 - 220 g, male swiss albino mice weighing 25 - 30 g were used in this study. The animals were maintained under suitable nutritional and environmental conditions throughout the experiment. All the pharmacological experimental protocols were approved by the Institutional animals ethics committee, (REG NO: 821/01/a/CPCSEA, dated: 6th AUG 2004) H.S.K. College of Pharmacy, Bagalkot - Karnataka

The animals were divided into five groups of seven animals each, for each model,

Group 1: serves as normal (without i.c.v. administration)
Group 2: serves as control (i.c.v. administration of saline 2 µl / animal)
Group 3: oxytocin administrated by i.c.v. (2 µg / animal)
Group 4: oxytocin administrated by i.c.v. (4 µg / animal)
Group 5: oxytocin administrated by i.c.v. (8 µg / animal)

Methods

Intracerebroventricular injections

Intracerebroventricular injection (2 µl / mouse) of vehicle or oxytocin was made free-handedly in the left ventricle, according to the procedure of Haley

and McCormick, using a hamilton microsyringe (10 µl) with a needle (diameter 0.5 mm), the level of which protruded by only 3 mm from a guard, for rat and 2 mm for mice limiting its penetration into the brain. The injection in manually immoblised mice lasted for approximately 3 sec. and success of the injections was observed using a methylene blue dye (after sacrifice and frontal brain sectioning) that the injection was successful in trials. It was also verified in several animals, after sacrifice, that the mark of the needle puncture on the parietal bone was located at least 1.5 mm behind the bregma and at least 2.5 mm before the lambda, with laterality between 1 and 2 mm relative to the brain median line.10 min after the intracerebroventricular administration of saline / oxytocin the observations was recorded for hole board, elevated plus maze and open-field test for 5 min[6].

Hole Board Test in male wistar rats and swiss albino mice

Hole board test used to assess number of pokings in the holes by animals during 5 min time. Exploratory behavior was assessed using the hole-board test, as described by Boissier and Simon. The apparatus is fabricated according to standard procedure consist of a square plate (40 x 40 cm 1cm thick, with 16 holes- diameter of 2 cm), which was evenly distributed on the surface at 3.5 cm from the edges. The apparatus was elevated to a height of 100 cm in a dimly illuminated room. Ten minutes after the i.c.v. administration; mice were placed in the center of the plate, and the number of head dips were immediately counted for 5 min[6].

Elevated plus maze test in male wistar rats and swiss albino mice

The elevated plus- maze is a novel test for the selective identification of anxiogenic and anxiolytic drug effects in rodents. The test is principally based on the observations of Montgomery showing that exposure of animals to an elevated (open) maze alley evokes an approach-avoidance conflict that is considerably stronger than that evoked by exposure to an open maze alley. Exposure of rats to novel stimuli can evoke both exploratory drive and fear drive and approach-avoidance conflict response. Elevation of the maze causes greater fear and more avoidance conflict. The plus-maze apparatus consists of two open (16 x 5 x 12 cm for mice and 50 x 10 cm for rats) and two closed arms (16 x 5 cm x 12 cm for mice and 50 x 10 x 40 cm for rats), and an open roof with the entire maze elevated (25 cm for mice and 50 cm for rats) from the floor. The animals were placed individually at the center of the elevated plus-maze with their head facing open arm. During the 5-min test, the preference of the animal for the first entry, the number of entries into the open or closed arms and the time spent in each arm of the maze were recorded. Each animal were used only once and the tests were carried out during a fixed time of the day. The rationale was that the open arms are more fear-provoking and that the ratio of either time spent on open:closed arms or entries into open-closed arms reflect the relative "safety" of closed arms compared with the relative "fearfulness" of open arms. Anxiolytics would be expected to increase the proportion of entries into and time spent on open arms[16].

Open field test for male wistar rats and swiss albino mice

Open field test was used to assess exploratory behavior of animals during 5 min time. Animals were kept under laboratory condition 1hour prior to OF test. Briefly, rat / mouse were placed in an open field in the sound-attenuated room. The floor was white polyvinyl with a black grid dividing OF into 64 squares (10 x 10 cm) for rat and fabricated OF for mice consists of 64 squares (5 x 5 cm). Illumination was provided by a bulb (60 W) placed above the center of the field, while the rest of the room was in darkness. The rat / mouse was placed in the center of the field and observed for 5 min. in this test latency time to start to explore the open field (seconds), horizontal locomotor activity (grid lines crossed), vertical locomotor activity (rearing), grooming (rubbing the nose its forepaws and preening), and instance of defecation (number of boluses) were recorded. Between the trials the box was cleaned with wet sponge and paper tissue. The results of the seven animals OF tests were summed and presented as total OF activity[22].

Statistical Analysis

All the data collected in the present study are expressed as mean ± standard error of mean (SEM) and analysed by students 't' test for coming to conclusion. P value less than 0.05 was considered as significant.

Results and discussion

Results

Hole Board Test in male Swiss albino mice

i.c.v. administration of normal saline reduced total number of poking from 11.57±0.17 to 8.57±1.08 and decrease was 25.92 %. Oxytocin at 2µg increased total number of pokings to 12.42±0.71 and increase was 44.9% which was almost equivalent to

Table 1: Effect of i.c.v. administered saline/oxytocin on Behavior of male swiss albino mice in hole board test

Treatment	Total number of pokings	% Decrease in number of pokings	% Increase in number of pokings
Normal	11.57± 0.1 7	--	--
Saline	8.57±1.08	25.92	--
Oxytocin 1mg /ml	12.42±0.71*	--	44.9
Oxytocin 2mg /ml	9.57±0.71	---	11.66
Oxytocin 4mg /ml	7± 0.90	18.31	---

Male mice were administered saline/oxytocin of volume 2ml /animal prior to testing 10 minutes after the i.c.v. administration of drug, animals were kept on hole board and number of pokings were counted for 5 minutes. Results are expressed as mean ± SEM and percent change in Behavior. Results obtained were analysed stastically by students 't' test by comparing saline treated group with normal and oxytocin treated group with saline *P<0.05.

Table 2: Effect of *i.c.v.* administered saline/oxytocin on Behavior of male wistar rat in hole board test

Treatment	Total number of pokings	% Decrease in number of pokings	% Increase in number of pokings
Normal	5.71 ± 0.68	--	--
Saline	4.38 ± 1.95	23.29	--
Oxytocin 1mg /ml	4.85 ±0.40	--	10.7
Oxytocin 2mg /ml	4.28 ± 0.52	2.28	--
Oxytocin 4mg /ml	4.14 ± 0.40	5.47	--

Male wistar rat were administered saline/oxytocin of volume 2ml /animal prior to testing 10 minutes after the i.c.v. administration of drug, animals were kept on hole board and number of pokings were counted for 5minutes. Results are expressed as mean ± SEM and percent change in Behavior. Results obtained were analysed stastically by students 't' test by comparing saline treated group with normal and oxytocin treated group with saline.

Table 3: Effect of *i.c.v.* administered saline/oxytocin on Behavior of male swiss albino mice in elevated plus maze model

Treatment	No of entries in open arm	No of entries in closed arm	Total duration of time spent in open arm (sec)	Total duration of time spent in closed arm (sec)
Normal	1 ± 0.30	5.57 ± 1.23	5.57 ± 1.66	158.1 ± 25.55
Saline	0.85 ± 0.26	8.85 ± 0 .70	4.85 ± 10.78	194 ± 10.78
Oxytocin 1mg/ml	0.28 ± 0.28	7.57 ± 1.28	1.14 ± 1.14	210.5 ± 16.48
Oxytocin 2 mg/ml	3.85 ± 1.50	7.71 ± 0.86	27.28 ± 26.38	185.42 ± 26.38
Oxytocin 4 mg/ml	1.28 ± 0.28	10.42 ± 0.86	5 ± 12.40	185.1 ± 12.40

Male mice were administered saline/oxytocin of volume 2ml /animal prior to testing 10 minutes after the i.c.v. administration of drug, animals were placed on elevated plus maze and duration of time spent in open & closed arm is noted for 5minutes. Results are expressed as mean ± SEM and percent change in Behavior. Results obtained were analysed stastically by students 't' test by comparing saline treated group with normal and oxytocin treated group with saline

Table 4: Effect of *i.c.v.* administered saline/oxytocin on Behavior of male wistar rat in elevated plus maze model

Treatment	No of entries in open arm	No of entries in closed arm	Total duration of time spent in open arm (sec)	Total duration of time spent in closed arm (sec)
Normal	0.14 ± 0.14	1.85 ± 0.26	1.71 ± 1.70	288.2 ± 4
Saline	0.14 ± 0.14	2.28 ± 0.35	0.85 ± 0.85	253.8 ± 10.5*
Oxytocin 1mg /ml	2 ± 0.30**	5 ± 0.43**	17.85 ± 2.61***	248.2 ± 6.63
Oxytocin 2mg /ml	4.71 ± 0.47***	6.71 ± 0.60***	80.28 ± 10.33***	189.28 ± 16.68*
Oxytocin 4mg /ml	4.85 ± 0.26***	6.71 ± 0.64***	39.85 ± 3.20***	205 ± 8.70*

Male wistar rat were administered saline/oxytocin of volume 2ml /animal prior to testing 10 minutes after the i.c.v. administration of drug, animals were placed on elevated plus maze and duration of time spent in open & closed arm is noted for 5minutes. Results are expressed as mean ± SEM and percent change in Behavior. Results obtained were analysed stastically by students 't' test by comparing saline treated group with normal and oxytocin treated group with saline. *P<0.05, **P<0.01, ***P <0.001.

Table 5: Effect of *i.c.v.* administered saline/oxytocin on Behavior of male swiss albino mice in Open-field test

Treatment	Latency time to explore (sec)	Vertical movement (rearing)	Horizontal movement	Grooming	Defecation	Urination
Normal	6.57 ± 1.98	1.85 ± 0.76	18.14 ± 1.35	2.42 ± 0.29	1.42 ± 0.52	0.28 ± 0.18
Saline	6.14 ± 1.05	8.14 ± 0.73***	72.28 ± 3.68***	8.71 ± 1.9*	2 ± 0.21	0.28 ± 0.18
Oxytocin 1mg /ml	22.82 ± 8.65	5 ± 2.04	95.57 ± 5.57*	4.85 ± 1.82	1 ± 0.30*	0.14 ± 0.14
Oxytocin 2mg /ml	17.28 ± 8.6	9.28 ± 2.56	178 ± 27.42**	6.57 ± 2.14	1.14 ± 0.45	0.14 ± 0,14
Oxytocin 4mg /ml	5.85 ± 1.58	17.85 ± 3.5*	205.5 ± 12.8***	10.85 ± 2.87	0.14 ± 0.14***	0.14 ± 0.142

Male mice were administered saline/oxytocin of volume 2ml /animal prior to testing 10 minutes after the i.c.v. administration of drug, animals were kept on open field and number of horizontal and vertical movements, grooming, defecation & urination along with latency time to explore (sec) were recorded in 5minutes. Results are expressed as mean ± SEM and percent change in Behavior. Results obtained were analysed stastically by students 't' test by comparing saline treated group with normal and oxytocin treated group with saline. *P<0.05, **P<0.01, ***P <0.001.

Table 6: Effect of *i.c.v.* administered saline/oxytocin on Behavior of male wistar rat in Open-field test

Treatment	Latency time to explore (sec)	Vertical movement (rearing)	Horizontal movement	Grooming	Defecation	Urination
Normal	4.57 ± 0.84	6.14 ± 0.76	54.57 ± 4.38	5.28 ± 0.68	2.71 ± 0.28	0.28 ± 0.18
Saline	3.71 ± 0.52	2 ± 0.48**	52.85 ± 4.6	3.42 ± 0.60	3.42 ± 0.62	0.14 ± 0.60
Oxytocin 1mg /ml	6.14 ± 1.72	7.14 ± 1.18	74.57 ± 12	3.71 ± 0.86	2.28 ± 0.52	0.14 ± 0.14
Oxytocin 2mg /ml	2.57 ± 0.48	5 ± 0.61**	107.4 ± 7.07**	3.14 ± 0.59	2 ± 0.53	0.14 ± 0.14
Oxytocin 4mg /ml	2.71 ± 0.71	6.42 ± 0.48***	113.1 ± 6.42***	5 ± 0.57	2.57*	0.14 ± 0.14

Male wistar rat were administered saline/oxytocin of volume 2ml /animal prior to testing 10 minutes after the i.c.v. administration of drug, animals were kept on open field and number of horizontal and vertical movements, grooming, defecation & urination along with latency time to explore (sec) were recorded in 5minutes. Results are expressed as mean ± SEM and percent change in Behavior. Results obtained were analysed stastically by students 't' test by comparing saline treated group with normal and oxytocin treated group with saline. *P<0.05, **P<0.01, ***P <0.001.

normal. Oxytocin at 4µg slightly increased number of pokings (9.57±0.71) and increase was 11.66%. However, higher dose of oxytocin does not produce any significant effect on total number of pokings and decrease was 18.31%.

Hole Board Test in male wistar rats

Intracerebroventricular administration of saline 2µl reduced total number of pokings from 5.71±0.68 to 4.38±1.95 and percent decrease was 23.29% when compared to non i.c.v. saline administration. No significant effect was observed with all three doses of oxytocin in male rats and percent change in behavior was 10.7, 2.28 and 5.47 with respect to 2µg, 4µg and 8µg.

Elevated plus maze test for male Swiss albino mice

i.c.v. administration of saline and oxytocin does not produce any significant change in behavior of male mice when compared to normal and oxytocin treated group to i.c.v. saline.

Elevated plus maze test in male wistar rats

i.c.v. administration of saline produced no significant change in anxiety and fear when compared to normal male rats. 2µg dose of oxytocin significantly ($p<0.01$) increased total number of entries in open arm from 0.74±0.14 to 2±0.3 and total duration of time spent in open arm was 17.85±2.61 ($p<0.001$) when compared to i.c.v. saline, at the same time total number of entries in closed arm were also increased significantly to 5±0.43 from 2.28±0.35. intracerebroventricularly administered oxytocin at doses 4µg and 8µg significantly ($p<0.001$) increased total number of entries in open arm and duration of time spent in open arm with significant increase in total number of entries in closed arm.

Open field test for male Swiss albino mice

Central administration of saline significantly increased vertical and horizontal movement without altering latency time to explore, grooming, defecation and urination behavior when compared to normal. Small dose of oxytocin 2µg does not produce any significant change in behavior when compared to i.c.v. administered saline. Oxytocin 4µg, increased total number of horizontal movements but not other behaviors high dose of oxytocin 8µg significantly ($p<0.001$) increased total number of horizontal movements and decreased ($p<0.001$) total number of defecation.

Open field test for male wistar rats

Moderate dose (4µg) and high dose (8µg) of i.c.v. oxytocin significantly ($p<0.01$ and $p<0.001$) increased total number of vertical movement and horizontal movement, when compared to i.c.v. administrated saline, without affecting other parameters considered in this study. No significant change in behavior was observed with small dose of oxytocin (2µg) in male rat when compared to i.c.v. saline group.

Discussion

In the present study we observed that central administration of oxytocin in male mice increased number of head dippings, confirming anxiolytic property of oxytocin at small dose. However, in this study the anxiolytic property was reduced with respect to increase in dose of i.c.v. oxytocin and highest dose of oxytocin produces anxiogenic effect. This study also confirms the central role of oxytocin in curiosity behavior of male mice. One possible reason for this effect may be through its overactivation of HPA axis. When the same kind of study carried out in wistar rat no significant change was observed with all doses of oxytocin supporting the earlier studies on variation in level of oxytocin in oxytocinergic neurons and oxytocin receptors distribution from species to species[33,5,12].

Well validated complementary model for the assessment of anxiety levels the elevated plus maze test was used [16]. i.c.v. administration of oxytocin in male mice has no significant effect on behaviors; however more behavioral changes were seen in wistar rats. Increasing doses of i.c.v. oxytocin increased the preference of rats for open arm sections. Thus oxytocin increased the number of entries, the time spent and total distance traveled in the open arm and the central area which may be

interpreted as decrease in anxiety. Oxytocin decrease time spent in the closed arm confirming the anxiolytic effect of oxytocin in male rats. This may be due to excess level of oxytocinergic receptors present in rat than mice[33].

Defecation and urination are the parameters which can be considered for studying stress and fear. These parameters were considered as a part of exploratory nature of animals and associated behaviors. The open field test was carried out on the black and white compartment device based on the conflict between inherient tendency of albino mice to explore a novel environment and their natural avoidance of brightly lighted open fields[22]. Intracerebroventricular injection of oxytocin significantly increased vertical and horizontal movements without altering latency time to explore, grooming, defecation and urination. This also increased the percentage of white compartment confirming its significant role in male swiss albino mice and wistar rats, exploratory behavior and curiosity and lack of fear.

The pronounced grooming is often related to dearousal following stress exposure in the open field test, it could be viewed as an early expression post stress behavior in rats. This behavior is elicited by exposure to mild stress and chemical stimulation of paraventricular nucleus of the hypothalamus at levels that are known to activate the HPA axis. This indicates the involvement of HPA axis in grooming behavior of rats. The present results are in consistent with previous behavior.

Activation of the oxytocin receptor might occur by the opening of a solvent exposed site in the cytosolic domains[12,11]. OT receptors are functionally coupled to $G_{q/11}\alpha$ class GTP binding proteins that stimulate together with $G\beta\gamma$ the activity of phospholipase C-β isoforms. This leads to the generation of inositol trisphosphate and 1,2-diacylglycerol. Inositol trisphosphate triggers Ca^{2+} release from intracellular stores, whereas diacylglycerol stimulates protein kinase C, which phosphorylates unidentified target proteins. Finally, in response to an increase of intracellular $[Ca^{2+}]$, a variety of cellular events are initiated. For example, the forming Ca^{2+}-calmodulin complexes trigger activation of neuronal and endothelial isoforms of nitric oxide (NO) synthase[8]. This will lead to increase in level of nitric oxide and help in overcoming from the behavior of aggression and anxiety. NO in turn stimulates the soluble guanylate cyclase to produce cGMP. In neurosecretory cells, rising Ca^{2+} levels control cellular excitability, modulate their firing patterns, and lead to control transmitter release.

The paraventricular nucleus of the hypothalamus contains the cell bodies of a group of oxytocinergic neurons projecting to extrahypothalamic brain areas and to the spinal cord. In male rats these neurons can be activated by dopamine, excitatory aminoacids, nitric oxide and oxytocin to induce sexual responses. These are apparently mediated by the activation of NO synthase[29].

Recently it has been reported that dopaminergic agonist induces behavioral responses by releasing oxytocin in the central nervous system. The electrophysiologic study has shown that oxytocin is able to activate several neuronal population in different rat brain areas including the hypothalamic supraoptic nucleus, paraventricular nucleus, hippocampus and the dorsal motor nucleus of the vagus nerve[28]. In the CSF, OT is normally present at concentrations of 10-50 pM, and its half-life is much longer (28 min) than in the blood (1-2 min). In humans and in monkeys, a circadian rhythm in the OT concentrations in the CSF has been found with peak values at midday.

Glucocorticoids have also been reported to modulate cerebral OT receptor varies across species not only in its distribution but also in its regional regulation by gonadal steroids. For example, estrogen increased OT receptor binding in the rat brain but reduced OT receptor bonding in the homologous regions of the mouse brain[1,25,33,18].

On the other hand, in spite of the relationships frequently observed between these psychic disorders and stress, involving the hypothalamic-pituitary-adrenocortical axis oxytocin appears to reduce corticosterone/cortisone level, and increases CCK level secretion[30]. As a matter of fact, oxytocin centrally administered in male mice/rats affect the plasma corticosterone level induced by i.c.v. injection; this modality of injection likely corresponding to a strong stress.

Taken together, the behavioral responses to i.c.v. injections of oxytocin described in the present study give some functional significance to the presence of oxytocin and its receptor in the central nervous system. Several studies have localized oxytocin binding sites in the cerebral cortex, amygdala, hippocampus, nucleus accumbens, thalamus, and striatum these regions are known to be involved in mood disorders such as anxiety and depression. Oxytocin is also reported to be present in hypothalamus plays a role in the control of feeding, thirst and several studies have identified the presence of OT receptor in specific nuclei of spinal cord ventral horn associated with motor function. Oxytocin receptors are also predominantly expressed in motor neurons of the brain stem and spinal cord indicating a possible role of oxytocin in the control of neuromuscular functions and locomotor activity.

On the basis obtained from this study and previous study we propose that i.c.v. administration of oxytocin, will acts on OT receptors present in hypothalamus, limbic system, basal ganglia, brain stem and spinal cord and activates central oxytocinergic system as well mimic dopaminergic system and follows IP_3/ DAG pathway, releasing calcium and activating nitric acid system, HPA axis,

central transmitter release, and modulates neuronal firing at the doses selected in this study, this may be involved centrally in regulating behavior of poking, ambulatory movement in male mice and anxiolytic, fearless behavior in male rats. At higher level of oxytocin in CSF may be involved in controlled ambulatory movement suggesting hyperactivation of HPA axis, dopaminergic system, limbic system and signalneurons. This study supports the earlierstudies on involvement of oxytocinergic system in behavior regulation.

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