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EFFECTS OF OXYTOCIN (OXT/OT) ON EATING BEHAVIOUR OF ALBINO WISTAR RATS

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ABSTRACT: *Oxytocin (Oxt/OT) is a hormone, a neuropeptide, that is sometimes called curdle chemical hormone (The American Society of Health-System Pharmacists, 2015). It is one of the hormones produced in the hypothalamus just like gonadotropic and vasopressin hormones. The rats used for the study comprised twelve rats (6 males and 6 females). The rats were grouped into experimental and control groups. The drug used for the study was a drug concentration of oxytocin and a Non-Hydrated Intravenous Solution (NH IV Solution). NH IV solution was used in order to reduce the concentration of the oxytocin drug so that it wouldn't be harmful to the rats. Before presenting food to the rats, the drug was administered once a day (early in the morning). The rats were grouped into three categories (lower, normal and higher dosage groups). The drug administered to the rats was based on the grouping of the rats. The rats under lower dosage were given 0.5ml of drug concentration; the normal dosage was given 0.8ml of drug concentration, while the higher dosage was given 1ml of drug concentration. Weight checking was done before and after the study in order to examine possible side effects of oxytocin on the weight of animals based on their rate of eating. The results were analysed using different statistical methods depending on the hypothesis stated. Hypothesis 1 was analysed using factorial design, while hypothesis 2 was analysed using an independent t-test method of research. The results analysed revealed that oxytocin has some significant effects on the eating behaviour of the albino Wistar rats. The result proved that oxytocin has a negative/side effect on eating behaviour, causing a reduction in craving for food in the rats.*

KEYWORDS: Oxytocin hormone, eating behaviour, Albino Wistar rats, Eating disorder, Weight gain/loss.

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INTRODUCTION

Oxytocin (Oxt/OT) is a hormone and neuropeptide that is sometimes called the curdle chemical hormone (The American Society of Health-System Pharmacists, 2015). It is one of the hormones produced in the hypothalamus, just like gonadotropic and vasopressin hormones. After the secretion of oxytocin in the hypothalamus, it travels in the neuron itself to the posterior lobe of the pituitary gland. Its secretion depends on the electrical activity of neurons in the hypothalamus (Landgraf & Neumann, 2004). Oxytocin is a peptide of 9-amino acids (Cys-tyr-ile-gln-asn-lys-pro-leu-gly). Cys- Cysteine, Tyr-Tyrosine, Ile-Isoleucine, Gin-Glutamine, Asn- Asparagine, Pro- Proline, Lys- Lysine, Leu- Leucine and Gly- Glycine (Burtis, Ashwood & Bruns, 2012).

As a medication, it is used to cause the contraction of the uterus to start or enhance childbirth, increase the speed of childbirth (reducing the duration of childbirth), and stop bleeding after delivery. It plays a key role in social bonding and sexual reproduction in both sexes (genders) during and after childbirth (American Society of Health-System Pharmacists, June 2015).

Oxytocin is released into the bloodstream as a hormone in response to stretching of the cervix and uterus during childbirth (labour), stimulation of the nipples from breastfeeding, and fondling of the breast during sexual activity and during orgasm. About 10 years ago, it was found that infusing oxytocin into the brains of non-pregnant female rats rapidly induced maternal behaviour towards young pups. This was striking as a treatment that attempted to mimic the supposed endocrine conditions of late pregnancy (treatment with oestrogen and progesterone, sometimes with added prolactin) had proved relatively ineffective. Related findings were reported in ewes, which exhibit hostile attributes towards offspring other than their own. It appears that the central (behavioural) effects of oxytocin might resemble its peripheral actions and represent a chemical “code” synchronising behavioural and physiological mechanisms for the important adaptive role of maternity. (British Medical Journal, “309:891”, June 1994).

Animal research has demonstrated oxytocin’s unique role in parturition, milk let-down, pro-social behaviour (trust, generosity, decreasing fear and social attachment) anti-social behaviour (envy and gloating) protective aggression against intruders as well in aspects of and bonding between mother and infant and between mating pairs (sexually attracted pairs/genders) and play a unique role in human sexual response and orgasm. (British Medical Journal, “309:891”, June 1994).

Oxytocin receptors are transmitted/released by neurons in different parts of the brain and at the spinal cord, including the amygdala, ventromedial hypothalamus, septum, nucleus accumbens, and brainstem. This expression is caused by oxytocin receptor cells, leading to some physiological and psychosocial responses such as:

Let-down Reflex: In lactating mothers, oxytocin acts at the mammary glands, causing milk to be released into the sub-areolar sinuses of the breast, from where it can be extracted or sucked by the newborn baby. Suckling by infants at the nipple is relayed by the spinal nerves to the hypothalamus. Breast stimulation due to sucking causes neurons that make oxytocin to fire its action potentials in intermittent bursts. The bursts result in the secretion of pulses of oxytocin from the neuro-secretory nerve terminals of the pituitary gland. (Takayanagi, et. al. 2005).

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Uterine wall contraction: Most importantly for cervical dilation before childbirth, oxytocin is said to be used in enhancing contractions of the uterine wall during the second and third stages of labour. Oxytocin is also said to assist the uterus in clotting the placental attachment point postpartum (Takayanagi et al., 2005).

Possible Relationship Between Oxytocin and Eating Behaviour

Oxytocin as a hormone or as medication has some basic effects (s) on the eating behaviour of humans. These effects are either positive or negative. This influence is somewhat gender-based (the effect it has on males is sometimes different from that on females). Eating behaviour in the context of this study/research will be viewed from the perspective of an eating disorder. Eating disorders are in two basic forms, it's either the individual eats too much, i.e., binge eating (bulimia nervosa), or does not want/willing to eat at all (anorexia nervosa).

Binge eating disorder (BED) is a mental disorder characterised by recurrent consumption of an unusually huge/large amount of food in a discrete period, accompanied by a sense of lack of control, without inappropriate compensatory behaviours typical of bulimia. (American Psychiatric Association, 2013). Consequently, binge eating disorder is often associated with obesity, and obese individuals with binge eating disorder have a higher concern for body weight and body-shape dissatisfaction than an obese individual without binge eating disorder. (Amianto, Ottone, Abbate Daga & Fassino, 2015). A study by York University researcher Caroline Davis and her colleagues at the Centre for Addiction and Mental Health (CAMH) is the first to demonstrate that variants of the Oxytocin Receptor (OXTR) gene contribute to why people over-eat or engage in episodic binge eating. They investigated how the OXTR gene encourages craving, food preferences, food consumption, and personality risk traits associated with the brain reward system/mechanisms. It encourages many survival behaviours, including those used to manage stress, according to Dr Caroline Davis, the lead researcher of the study.

Anorexia Nervosa (AN) is a very serious mental illness with high morbidity; it is also notoriously difficult to treat. Research has shown that oxytocin levels have been found to be low in patients with anorexia nervosa. Oxytocin increases attention and sensitivity to emotional expressions, and some evidence suggests that patients with anorexia nervosa struggle with recognising emotions. Research has shown that there is some significant difference in the oxytocin levels of animals suffering from anorexia nervosa and bulimia nervosa. There are low levels of oxytocin in animals with anorexia nervosa, while the level of oxytocin in patients/individuals with bulimia nervosa is sometimes the same as that of a normal animal.

Statement of Problem

Oxytocin is generally known to have some basic influence/effect on the labour process in women and also influences social and interpersonal relationships among people (MacDonald & MacDonald, 2010). Also, it's the most common medication used to cause vagina contractions due to delays in childbirth.

Apart from the medical use of oxytocin and its ability to induce labour and cause contractions in the uterine walls during childbirth, research by Nauert (2015) posits that oxytocin must be carefully used in order to avoid over-dosage because over-dosage use of oxytocin in a healthy young adult could lead to over-sensitivity/hypersensitivity to the emotions of people around.

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The main problem to be investigated in this study is to ascertain the extent to which the use of oxytocin as a medication will affect the rate of eating.

Therefore, based on the problem stated, this experiment will investigate the usage of oxytocin as a medication on three different levels normal dosage: dosage per body weight (0.503ml – 0.703ml/150 – 250g), lower dosage: dosage per body weight (0.402ml – 0.501ml/150 – 250g) and higher dosage: dosage per body weight (0.705ml - 0.803ml/150 – 250g) for fourteen days, to check the oxytocin drug will have a similar or different effect(s) on the eating behaviour of the experimental subject (Wistar rats).

Hypothesis Tested

The following hypothesis was tested in the study.

- Oxytocin drug will have a significant effect on the eating behaviour of albino Wistar rats.
- There will be a significant difference in weight gain between the albino Wistar rats of the experimental group and the control group.

METHODS

Twelve Wistar rats were used for the experiment. The weight of the Wistar rats ranged between 150g – 256g for both males and females. Based on the literature reviewed, an average body weight of a mature Wistar rat falls in the range between 150g – 300g. The Wistar rats were housed in groups based on the number of variables (eating behaviour) tested, having both experimental and control groups per variable. The researcher made sure the Wistar rats were housed under normal day and night atmospheric conditions.

Drug

The drug used for the study was Oxytocin Injection BP by Rotex Media, Trittau, Germany. The drug comes in packs depending on the country's drug regulations. The drug pack used comes in an international unit of 10IU per ampoule, with a total of ten ampules, making it 100IU ampules. The dosage used ranged from 0.5ml to 1ml, depending on the body weight of the rats. A non-hydrated Intravenous Solution (NH IV Solution) was used to dilute the oxytocin drug to reduce its concentration for it would not be harmful to the rats.

Experimental Procedure

Twelve rats were used for the study, comprising six males and six females. The weight of the rats were put into consideration before selecting them for the study. The body weight of the rats was in the range of 150g to 256g, and an average body weight value was deduced after summing up the total body weight of the rats and dividing it by twelve, which is the total number of rats used for the study. The average body weight of the rats used was 196g. The time used for the study was 3 weeks. The first week was for acclimatisation of the rats and familiarity with the researcher and the laboratory environment. The remaining two weeks were used for the research; there was weight checking before grouping the animals into control and experimental groups, this was done to avoid sampling or grouping error. A daily weight check was done throughout the period of the experiment to monitor weight gain or weight loss in the

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rats. The average weight of food (153g/day) was maintained throughout the experiment. The food presented/given to the rats was a product of Afrimash Hybrid feeds. The feed comes in powdered form, but the researcher added water to the feed to make it a little bit solid or sticky for the rats so that the powdered feed won't be sniffed into their nostrils while eating. The food presented to the rats was measured before presenting it to the rats, and the leftovers were weighed the next morning to monitor their rate of eating. The weight of the rats was checked after the experiment to know if the effect of oxytocin on the weight of the rats was based on their rates of eating. The drug administered to the rats was a concentration of oxytocin drug mixed/diluted with a Non-Hydrated Intravenous Solution (NH IV Solution). The rats were tested on three levels of drug dosage (lower dosage of 0.5ml, normal dosage of 0.8ml and higher dosage of 1ml) of oxytocin concentration. The laboratory room temperature was monitored using the AccuWeather app, designed by Google Application Company. Throughout the experiment, the laboratory temperature was between 23^0 and 26^0 Celsius.

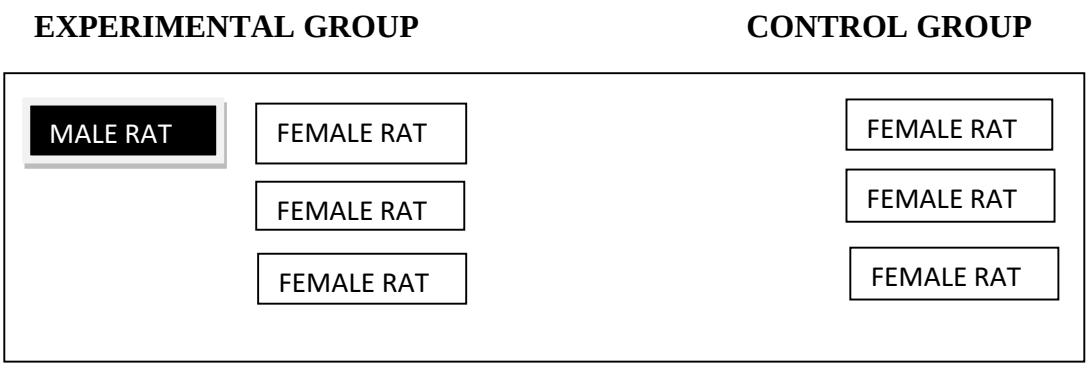
Experimental Design and Data Analysis

The research was a laboratory experiment studying the effect of oxytocin drugs on the eating behaviour of albino Wistar rats. Twelve rats, comprising six males and six females, were used for the study. The rats were tested on three levels of drug dosage administration (lower dosage of 0.5ml, normal dosage of 0.8ml, and higher dosage of 1ml) of oxytocin concentration.

The rats were divided into two main groups, the experimental and control groups, but later divided into three subgroups: lower dosage, normal dosage, and higher dosage. Each subgroup had a male and female in different cages/cells, making the cells twelve cells for both the control and experimental groups. Cage labelling/naming was introduced to properly identify cells from one another.

The study adopted an independent group and factorial design. The results were analysed using different statistical methods depending on the hypothesis stated. Hypothesis 1 was analysed using a factorial design, while hypothesis 2 was analysed using an independent t-test method of research.

Table 1: Descriptive table showing the grouping of the rats used for the study.



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RESULT

Table 1: Descriptive Analysis showing differences in eating behaviour (consumed food) between the control and experimental groups.

Experimental Group

Days	Lower Dosage Male	Lower Dosage Female	Normal Dosage Male	Normal Dosage Female	Higher Dosage Male	Higher Dosage Female
Day 1	34g	76g	53g	42g	42g	55g
Day 2	72g	54g	48g	23g	65g	41g
Day 3	33g	44g	50g	36g	76g	33g
Day 4	30g	25g	44g	37g	77g	37g
Day 5	34g	12g	34g	28g	68g	61g
Day 6	38g	32g	26g	39g	56g	44g
Day 7	57g	29g	48g	37g	65g	52g
Day 8	50g	35g	54g	47g	77g	57g
Day 9	72g	54g	62g	79g	90g	99g
Day 10	63g	72g	80g	75g	74g	69g
Day 11	61g	54g	48g	74g	50g	95g
Day 12	58g	50g	65g	68g	74g	65g
Day 13	50g	35g	54g	47g	77g	57g
Day 14	52g	45g	65g	78g	85g	80g

Control Group

Days	Lower Dosage Male	Lower Dosage Female	Normal Dosage Male	Normal Dosage Female	Higher Dosage Male	Higher Dosage Female
Day 1	88g	63g	71g	62g	93g	70g
Day 2	64g	36g	61g	84g	87g	73g
Day 3	61g	54g	74g	48g	50g	95g
Day 4	89g	17g	76g	40g	91g	74g
Day 5	20g	27g	36g	20g	54g	44g
Day 6	57g	32g	55g	68g	81g	64g
Day 7	70g	41g	77g	55g	73g	74g
Day 8	69g	57g	76g	60g	81g	64g
Day 9	63g	82g	73g	70g	84g	59g
Day 10	62g	64g	72g	79g	85g	90g
Day 11	69g	57g	76g	60g	81g	84g
Day 12	75g	68g	73g	60g	81g	84g
Day 13	69g	57g	76g	60g	81g	64g
Day 14	78g	70g	89g	79g	88g	85g

NOTE: The fixed weight of food given to the Wistar rats per day = 153g/day.

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Table 2: One-way ANOVA table showing the influence of drug dosage on the eating behaviour of the Wistar rats.

Source	Type III square	Mean Square	df	F
Corrected Model	3386.917	483.845		6.366
Intercept	30510.15	30510.15		401.49
Dosage	93.750	93.750	1	7.97*
Error	304.00	76.00	4	

The analysed result revealed that drug dosage has a significant effect on the eating behaviour of the albino Wistar rats. [F (1, 4) = 7.97, P<.05)]. This shows that oxytocin drug has a significant effect on the eating behaviour of albino Wistar rats. It enhances eating behaviour in Wistar rats. Based on this, the hypothesis earlier stated, which says "oxytocin drug will have a significant effect on eating behaviour of albino Wistar rats," is hereby accepted.

Table 3: Independent t-test table showing the weight comparison in eating behaviour of Wistar rats in experimental and control groups.

	N	Mean	df	t	Sig
Experimental group	6	381.7857	10	17.89**	.000
Control group	6	393.5714			

**P < 0.001

From the table above, comparing the mean scores of the two groups (experimental and control group), the result revealed that there is a significant difference in the weight gain of the Wistar rats in the experimental group (Mean=381.7857, N=6) and control group (Mean = 393.57, N=6). This assertion shows that the oxytocin drug has a significant side effect on the weight gain of the Wistar rats.

DISCUSSION AND CONCLUSION

The results presented in the table above revealed that oxytocin has significant effects on eating and weight gain in the Wistar rats. The result in Table 2 revealed that the oxytocin drug has a significant effect on the eating behaviour of the Wistar rats. The result is in support of the study done by Gold (2013) on eating disorders (Eds), where he stated that eating disorders can lead to changes in brain function, which are associated with the chronicity of the illness and formation of permanent unhelpful habits, such as overeating (Gold & Avena, 2013); (Janet Treasure & Ulrike Schmidt, 2013). These changes in brain function have a profoundly detrimental impact on social, psychological, and physical health (Kessler et al., 2013). Oxytocin has been found to play an important role in controlling appetite, stress, and social functioning. In other words, if there is a deficiency or over-secretion of oxytocin, it will result in changes in brain function, which might, in turn, affect the regulation of appetite, stress, and basic social functioning.

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The result is also in accordance with the study done by Davies and her colleagues (2016) at the Centre for Addiction and Mental Health (CAMH). The study was the first to demonstrate that variants in the Oxytocin Receptor (OXTR) gene contribute to why some individuals overeat or engage in episodic binge eating. They investigated how the OXTR gene encourages craving, food preferences, food consumption and personality risk traits associated with the brain reward system/mechanisms. It influences many survival behaviours, including those used to manage stress, according to Davies, the lead researcher in the study. "For example, oxytocin enhances pro-social and related behaviours. On the other hand, an increase in oxytocin tends to decrease cravings – especially the consumption of sweet carbohydrates.

Also, the result for the second hypothesis revealed that oxytocin had a significant effect in predicting weight gain when the Wistar rats in the experimental group were compared to the Wistar rats in the control group. The results made it known that oxytocin has a significant side effect on weight gain in the Wistar rats that were injected with the drug, which means the oxytocin drug inhibits weight gain in Wistar rats. The findings of this study correlate with the study done by Lawson. According to Lawson, chronic oxytocin administration leads to continuous weight loss due to a reduction in food intake, increasing energy dissipation, and inducing lipolysis. Lawson also went further to state that translational human studies are beginning to describe the effects of oxytocin on eating behaviour and metabolism and need to come up with other therapeutic potentials of oxytocin-based drugs for the treatment of metabolic disorders, such as obesity and diabetes mellitus (Lawson, 2017).

CONCLUSION

Based on the findings of this study, the following conclusions are hereby made.

i. Oxytocin drugs can help in treating eating behaviour such as binge eating.

ii. The oxytocin drug will enhance eating behaviour in Wistar rats that are not willing to eat.

iii. Oxytocin has a significant effect on weight gain, which means that the drug will inhibit weight gain in albino Wistar rats. Administration of the drug caused weight reduction in the Wistar rats used for the study. This was discovered when the weights of rats in the experimental group were compared with the weights of rats in the control group.

RECOMMENDATIONS

The results obtained from the study revealed that oxytocin has a physiological effect on the eating behaviour of the Wistar rats. Aside from the medical and physiological usefulness of Oxytocin in aiding safe child delivery, this study has revealed some other medical usefulness of Oxytocin and based on this, some recommendations are hereby made.

i. Sensitisation and public awareness need to be made on other usefulness of Oxytocin aside from inducing childbirth.

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ii. The results of this study revealed a relationship between Oxytocin and body weight. Therefore, the populace needs to be educated on Oxytocin's effect on reducing body weight.

iii. Medical authorities need to be informed of the usefulness of Oxytocin other than inducing labour in pregnant women. This study revealed that the Oxytocin drug could be used to enhance eating behaviour in any individual suffering from anorexia nervosa.

iv. Likewise, drug enforcement agencies should be educated and informed on how to sensitise citizens on the possible side effects of overdose usage of Oxytocin, which could probably lead to bulimia nervosa, which is an eating disorder.

v. Medical authorities/practitioners need to be cautioned/guided not to over-administer Oxytocin drugs to their patients because of its overtime usage side effects.

vi. Further research should be done to ascertain the results of this study, and further investigations should be conducted into the physiological effects of Oxytocin on eating behaviour.

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