



Study of Thyroid gland treated with Oxytocin in relation to the reproduction in the fish *Cyprinus carpio*.

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ABSTRACT

Oxytocin is neurohypophysial hormone primarily stimulates the uterine contractions during labour and release of breast milk. This hormone also plays a role in mating behavior and sexual response. In the present study, oxytocin was injected 1.5ml/kg body weight once in a week for four weeks in the fish, *Cyprinus carpio*. It has been observed that oxytocin stimulates the sexual behavior leading into breeding. Histological studies showed that oxytocin stimulate the estrogen secretion in the female and increased the number of follicular cells. Thus it had been concluded that thyroid gland treated with oxytocin is associated with induction of spawning reflexes in the fish *Cyprinus carpio*.

Keywords: Estrogen, Neurohypophysial hpomone Oxytocin, spawning reflexes.

INTRODUCTION

Oxytocin is made in magnocellular neurosecretary cells in the supraoptic nucleus and paraventricular nucleus of the hypothalamus and is released into the blood from the posterior lobe of the pituitary gland. Oxytocin is also produced in the corpus luteum of the buffalo and cow (Thomus, 2004). Oxytocin may enhance both sperm and egg transport thereby promoting reproductive success. In rhodents, oxytocin increased the sperm numbers in the ejaculate which would be beneficial to reproductive techniques (Milen Hess, 2002). In addition to effect in sexual behavior, it has been also shown to increase cervical contractions, modulations of sperm production and transport, Bales *et al.*, (2004). Recently, it has been shown that central isotocin has different effect from vasotocin with regard to the neural activity that drives humming behavior in the plainfin midshipman fish (Goodson and Bass, 2000). Oxytocin has also been shown to promote female sexual behavior in rat and other species ovarian hormones affect the levels of neurohypophysial hormone in the pituitary of the rat (Bale *et al.*, 2001).

Scientists gave an elementary account of thyroid of fishes, although the thyroid gland of teleost has attracted the attention of many investigators (Edwin *et al.*, 2007; Blanton Specker, 2007). Thyroid follicle is a hollow ball made up of a single layer of epithelial cells enclosing a fluid filled space. The thyroid cells are capable of accumulating iodine from the blood and storing it in the form of a thyroid hormone inside the lumen of the follicle under the influence of TSH from pituitary gland (Blanton and Specker, 2007). This hormone can be enzymatically released later into the blood when there is a demand for it. The thyroid hormone affects the general cellular metabolism in the animal. Frisen and Frisen (1967) believe that the two kinds of thyroid are equivalent in function and quantitative differences can be Sultan, explained on the basis of differences in total number of follicles in the pharynx and kidney (Raine and Latherland, 2009; Raine *et al.*, 20050). Thus, studies on the efficacy of oxytocin on reproduction in relation to thyroid glands in the fish, *Cyprinus carpio* (L.) are meagre. The present work is an attempt in this direction to assess the oxytocin treated thyroid gland as a suitability of substitute inducer for spawning reflex.

MATERIALS AND METHODS

Cyprinus carpio were collected, reared in a fibre glass tank and acclimatized to the laboratory conditions. The experiment was carried out for four week. Control and experimental groups were formed. Fishes from control groups were injected with 1.5ml of distilled water and experimental groups were injected with 1.5ml of oxytocin intramuscularly at every alternate day for four week. At the end of the experiment, fishes from control and experimental groups were sacrificed to study the histomorphological changes in the thyroid gland.

1. Control group: At the end of the experiment, the survival rate of control fish was 100%. The average weight length and girth of male and female were 612gm, 26cm, 28m and 655gm, 28cm and 27cm, respectively.

Thyroid gland

It is heterotropic structure and consisted of follicles distributed around blood vessels in pharyngeal region. They were small and mostly oval in outline. They were surrounded by epithelial cell lining.

The ovary was in the maturing form and histologically, reorganized into young oocytes, early maturing oocytes. The ovaries were yellowish in colour. Histologically oocyte showed small clear yolk vesicles and nucleus with undulated nuclear membrane.

Testis were opaque and in the maturing phase. In a section, a large number of primary, secondary spermatocytes and spermatids were visible. After injecting distilled water to the control group, no sign of courtship was observed even after the second dose

2. Experimental groups: Weight of testis and ovary was 75gm and 128gm, respectively. After 4 weeks, GSI of male and female was 14.56 and 22.69 respectively.

At the end of the experiment, the ovaries were in prespawning phase. The entire body cavity was occupied by the ovaries which were turgid. Histologically, large number of oocytes and ripe eggs were seen in a section with clear yolk vesicles and nucleus with undulated nuclear membrane.

At the end of the experiment, there was considerable increase in the weight and volume of testis which becomes turgid. Histologically, the seminiferous tubules were increased in number with spermatids and sperms.

Thyroid gland

External morphology:

The thyroid gland has a form of small follicles distributed around the ventral aorta as well as the roots of the afferent branchial arteries. They are densely accumulated on the dorsal side of the ventral aorta.

Histomorphology:

The thyroid gland is composed of a number of follicles, blood capillaries and connective tissue. The nuclei are elongated or oval shaped or round. Each nucleus contains 1 to 2 nucleoli. Sometimes, there appear large vacuoles in the epithelial cells and the nuclei are pushed towards one side. The nucleus stains red with azocarmine of Dawson and Friedgood's stain. The cytoplasm of these cells stains red after the application of this technique. The staining reactions of the colloid in the follicles presents many variations. In some follicles, it is homogeneously blue, in others it is homogeneously red after the application of trichrome stains. In still others, a part of the colloid is stained red and the remaining part is stained blue. Some follicles

show resorption vacuoles in the colloid towards its peripheral area. The follicular cells of thyroid gland were fully loaded with colloid and follicular size were increased and follicular number were also increased. Some thyroid follicles with thin epithelial lining were filled with colloid.

At the end of the experiment male and female were kept together (2:1). After 3 hrs of 2nd injection, the brooders started swimming actively, became excited and restless. Female was chased by the males pushing her with snout and after 4 hrs of 2nd injection, spawning occurred.

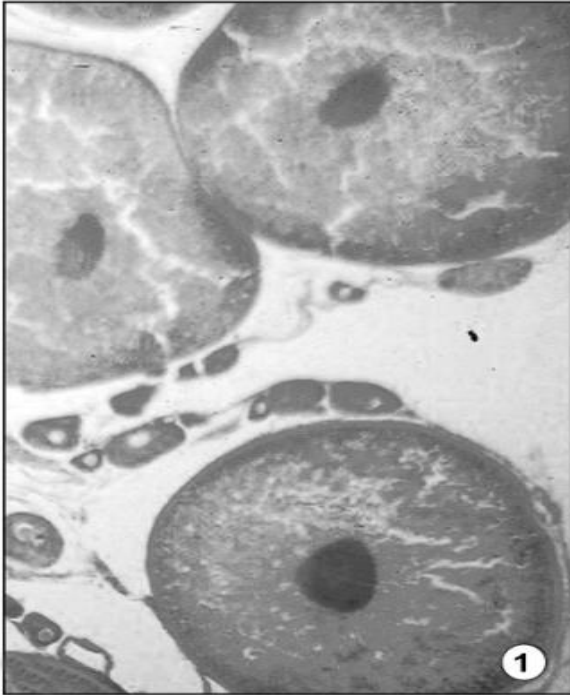


Fig. 1: Section of ovary showing maturing follicles with nucleus.

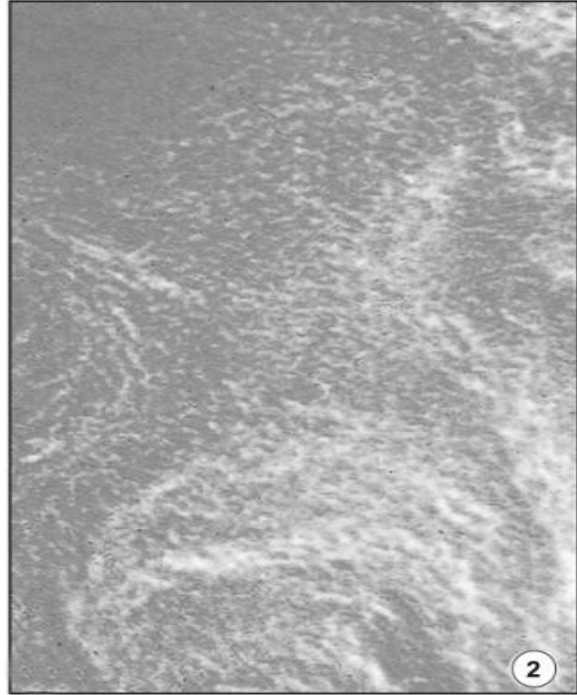


Fig. 2: Section of testis showing spermatocyte and spermatids.

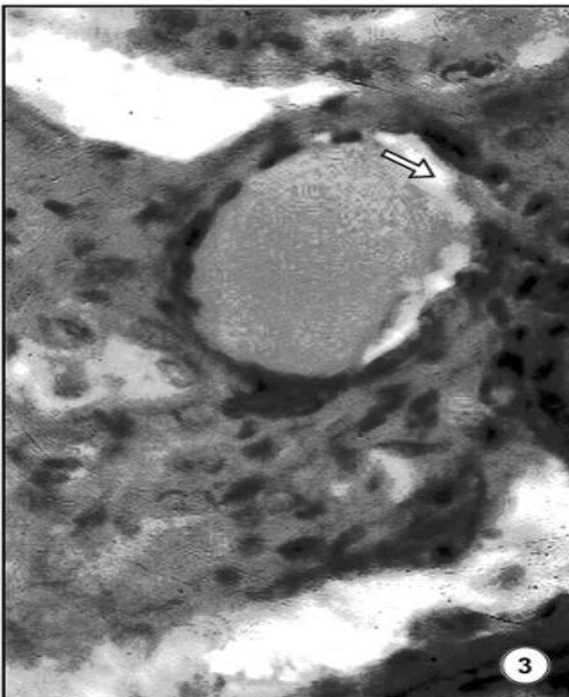


Fig. 3: Magnified view of thyroid showing large thyroid follicle with resorption vacuoles (→) along the epithelial lining.

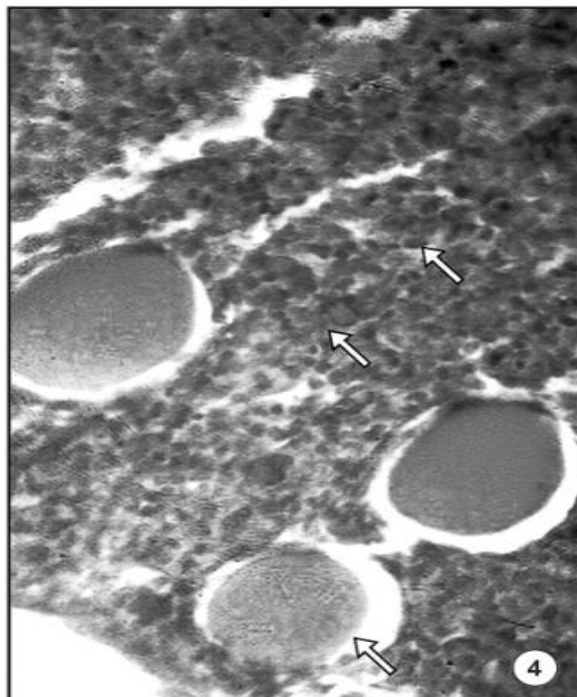


Fig. 4: Magnified view of adrenal gland showing chromaffin cells (→) interrenal cells (→) and thyroid follicle (→).

DISCUSSION

Recent studies have demonstrated that odours emanating from an individual have encoded messages of its race, age, sex and biological status. But our knowledge in this field is still fragmentary. In mammals, oxytocin is known to play an important role in almost all aspects of reproduction, including social recognition (Dluzenm *et al.*, 2000). Oxytocin also stimulates the sexual receptivity and reproductive behavior. In rodents oxytocin stimulates the spermatogenesis to increase the sperm number in the ejaculate, improves sperm quality and quantity (Huseyin, 2005) which would benefit all assisted reproductive techniques used in animals and also initiate female sexual behavior (Millen Hess, 2002). Very little information is available on the role of oxytocin in fish reproduction. Michael Black *et al.*, 2004 stated that socially induced sex change regulates forebrain isotocin in *Lythrypnus dalli*. He also reported that the reproductive behaviour of *Lythrypnus dalli* changes from female to male following the removal of a dominant male from a social group. The teleostean thyroid in general, consists of diffused follicles or groups of follicles clustered round the ventral aorta and afferent branchial arteries, but in *Ophiocephalus* and *Heteropneustes* it is compact and encapsulated. Interestingly, 13th of the 27th fish species in which heterotopic thyroid follicles have been described belong to the family of carp and minnows which are interspersed resulting into metastases. Thyroid hyperplasia and neoplasia have been described in teleostean fish (Fournie *et al.*, 2005; Leatherland and Down, 2001). The colloid in thyroid follicle of kidney tissue is the smallest and sub pharyngeal follicle appear to be more round than follicle of renal tissue and which is supported by the findings of Edwin *et al.*, (2007) in *Cyprinus carpio*. The staining reaction of the colloid varies in the same follicle. It stains basophilically depending upon the degree of secretory activity of the cells and also depending on the presence of resorption vacuoles in the follicle. They suggested that the acidophilic and basophilic affinity of the colloid is an indication of inactivity and activity of the gland.

On the basis of the above literature, the present investigation is an attempt to establish the role of thyroid gland treated with oxytocin in breeding of the fish *Cyprinus carpio*. The results of the present study related with endocrine glands like thyroid, show

inductive effect in maturation. Increase in GSI and fecundity is observed. Hence thyroid gland treated with oxytocin plays a definite role in fish reproduction.

SUMMARY AND CONCLUSION

The objective of the present study was to evaluate the study of thyroid gland treated with oxytocin on the reproduction in the fish, *Cyprinus carpio*. Oxytocin used in the present study showed early maturity, increased fecundity, increased rate of seed production and survival. In the present study, gonadosomatic index (GSI) were studied and recorded in the control and experimental groups. The effect of oxytocin on thyroid glands showed stimulatory effect. Particularly, the gonads were excessively stimulated resulted into early maturity. Thus, the results obtained from the present study, it can be concluded that oxytocin treated thyroid gland in the fish breeding technology will be effective and can be used as a substitute or stimulator.

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