EFFECT OF SYNTHETIC KISSPEPTIN-10 ALONE AND IN COMBINATION WITH DOMPERIDONE ON INDUCED BREEDING OF CIRRHINUS MRIGALA

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INTRODUCTION

As an additional source of human diet and as a component of various animal feeds, there is now a greater emphasis on fish production. The Mrigal is one of the most widely farmed species among the IMCs which has a good demand and market value. The knowledge of artificial breeding is a key aspect as it permits intensive production of a given species in controlled conditions. This allows continue production of juveniles for restocking natural or artificial water bodies (Montchowui et al., 2011). It has greatly contributed for the rapid development of carp culture in India without having to depend heavily on rive rine spawn collection (Nandeesh et al., 1990).

The Mrigal is one of the most widely farmed species among the Indian Major Carps which has a good market value. Mrigal do not breed in confined water hence hormone-induced spawning is the only reliable method to induce reproduction in these fishes. Mrigal have been successfully induced with various hormones like hCG, Carp Pituitary Extract (CPE), ovopel, ovaprim, ovatide (Chaudhuri and Alikunhi, 1957; Vardia, 1954; More et al., 2010; Nandeesh et al., 1990).

Although several hormones have been tried in India with varying degrees of success (Tripathi and Khan, 1990), none has gained acceptance at the farmer’s level either due to cost, non-availability or procedural difficulties. Recently it has been demonstrated that the kisspeptin system plays an essential role in the neuroendocrine control of puberty and reproduction by stimulating the GnRH neurons and subsequently releasing gonadotropin hormones. (Parhar et al., 2004; d’ Anglemont et al., 2007; Nocillado et al., 2007; Popa et al., 2008). In the present study, an attempt has been made to study the effect of synthetic kisspeptin-10 on induced breeding of Mrigal, Cirrhinus mrigala. The results of this study will provide information for the development of new induced agent for the breeding.

MATERIALS AND METHODS

All the induced breeding experiments were carried out at National Fish Seed Farm, Manimuthar (Tamil Nadu). Healthy, disease free, fully mature ripe fishes were chosen for the induced breeding.

Hormone treatment

In the first treatment-1 fishes were induced with Synthetic Kisspeptin alone at three concentrations viz., 50, 100, 150 μg/kg body weight and in the second treatment-2 they were induced with kisspeptin along with domperidone (at 10mg/kg body weight). Kisspeptin-10 was injected into the fish breeders through intramuscular injections just below the doransal fin. Hypodermic 2 ml syringe having 0.1 ml graduations with a needle no. 22 was used. Injections were given during the evening between 4-5pm. The injected breeders were released into the hapa for breeding. Next day the spent brooders were collected and transferred to the other pond.

Number of eggs produced

The numbers of eggs were measured by volumetric method. A beaker was used in which number of eggs was counted in
triplicate and the average of the three estimations was taken to know the numbers of eggs per beaker. Multiplying this with the number of beaker measured, total number of eggs was calculated.

Fertilization Rate:
The temperature of water used for incubation was kept around 27-30 °C. The fertilization rate of eggs laid by induced fish in each set, under different treatments was estimated. For estimation, three sub samples of water harden eggs were taken from each set and the number of fertilized egg (n) out of total eggs produced (N) in each sub samples was counted. The fertilized egg percentage was then calculated by the following formula (Hogendoorn, 1979) and their mean was determined

\[
\text{Fertilization } \% = \frac{n}{N} \times 100
\]

Hatching rate
Hatched eggs are collected and the hatching rate was calculated using the following formula (Olubiyi, et al. 2005)

\[
\text{Hatching } \% = \frac{\text{No. of hatchet fry}}{\text{Total no. of fertilized eggs}} \times 100
\]

Statistical analysis
The results found in the experiment were subjected to statistical analysis, ANOVA, (one way) that showed the significance (P<0.05) level of differences between the treatments. This statistical analysis was performed with the aid of the computer software MS Excel program.

RESULTS
The total number of eggs produced in case of treatment-1 and treatment-2 are shown respectively in Table-1 and Table-2. In Treatment-1, fishes induced with 100 µg/kg of Kisspeptin produced higher number of eggs (160557). On the contrary, in treatment-2, the higher numbers of eggs were produced by fishes induced with 150µg/kg+10mg/kg (181026). In both the treatments lowest number of eggs were produced by fishes induced with 50µg/kg of kisspeptin

Fertilization rate
In the present study, the fertilization rate ranged between 85.115-95% in all the treatment and are presented in Table 1. Maximum fertilization rate was obtained from the synthetic kisspeptin injected fish at 150 µg/kg body weight followed by 100µg/kg and 50µg/kg body weight. Highest fertilization rate of 94.49% was obtained from fishes induced with combination of Kisspeptin and domperidone at the rate of 100µg/kg + 10mg respectively.

Hatching rate
In the present study, least hatching percentage of 82.125% and a maximum hatching rate of 86.29% were observed from fish induced with 50µg/kg body weight of synthetic kisspeptin-10 (Table 1). The highest hatching percentage of 92.015 was recorded from treatment fish induced with combination of synthetic kisspeptin and domperidone at the rate of 100µg/kg + 10mg body weight followed by groups induced with 100µg/kg + 10mg and 50µg/kg + 10mg.

DISCUSSION
Overall, the results of this experiment revealed that there remained non-significant difference between the 2 treatments. In all the fishes induced with synthetic kisspeptin-10 alone and in combination with domperidone, fertilization rate, hatching rate had little variation. This variation was seen probably because they were treated in different doses of hormones. Although, some variations may arise due to the physiological differences in the pair of fishes and experimental error. Similar results were obtained by Basavaraja et al. (2007) where a combination of buserelin (an analogue of mammalian LHRH) and domperidone was used for breeding of Mrigal. The result obtained from m-LHRHa+ domperidone was better than the result obtained from S-GnRHa+ domperidone. Determination of hatching rate of fish is important for various aspects. It can determine the status of how many fry can be produced from a number of fish and how many are lost and why. It helps to improve the hatchery product and thereby production. Various works carried out on mrigal using different hormone like ovatide, ovaprim pituitary extract gave different hatching rate. (Mishra et al., 2001; Saini et al., 2001; More et al., 2010)

Recently Synthetic kisspeptin has been widely used to get gonadal maturation in fishes (Francis et al., 2011; Selvaraj et al., 2013; Unniappan et al., 2011; Benjamin et al., 2012) Even though studies are available on the effect of kisspeptin-10 (natural) on the maturity of fish; efficiency of synthetic kisspeptin-10 is very limited. In the present study an attempt was made to assess the effect of synthetic kisspeptin-10 alone

<table>
<thead>
<tr>
<th>Treatment-1</th>
<th>Number of eggs produced</th>
<th>Fertilization Rate (%)</th>
<th>Hatching Rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>50µg/kg body weight</td>
<td>125455 ± 2404.5</td>
<td>85.115 ± 0.79</td>
<td>82.125 ± 0.59</td>
</tr>
<tr>
<td>100µg/kg body weight</td>
<td>160557 ± 8983.1</td>
<td>89.495 ± 0.86</td>
<td>85.725 ± 0.28</td>
</tr>
<tr>
<td>150µg/kg body weight</td>
<td>159696 ± 2436.4</td>
<td>91.705 ± 0.45</td>
<td>86.29 ± 0.93</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Treatment-2</th>
<th>Number of eggs produced, Fertilization rate (%)</th>
<th>Hatching rate%</th>
</tr>
</thead>
<tbody>
<tr>
<td>50µg/kg + 10mg/kg body weight</td>
<td>135278 ± 2397.29</td>
<td>87.525 ± 1.01</td>
</tr>
<tr>
<td>100µg/kg body 10mg/kg weight</td>
<td>171706 ± 10596.62</td>
<td>94.49 ± 0.65195</td>
</tr>
<tr>
<td>150µg/kg body10mg/kg weight</td>
<td>181026 ± 7026.93</td>
<td>92.05 ± 1.07</td>
</tr>
</tbody>
</table>
and in combination with domperidone on induced breeding of mrigal. The addition of domperidone did not show any significant changes in the breeding performance of *C. mrigala*. Hence the present study concludes that kisspeptin alone can be used successfully for the induced breeding of mrigal.

**ACKNOWLEDGEMENT**

The authors express their gratitude to the Dean, Fisheries College and Research Institute, Tamilnadu Fisheries University, Thoothukudi, National Fish Seed Farm Manimuthar, India for providing the facilities for the conduct of the present study.

**REFERENCES**


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