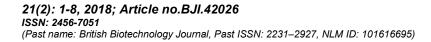
Biotechnology Journal International





Reproductive Performance of Carica papaya, Hibiscus rosa-sinensis and Ipomoea batatas on Female African Catfish (Clarias gariepinus)

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Authors' contributions

This work was carried out in collaboration between all authors. Author PBE designed the study, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript. Authors UUU and CMA managed the analyses of the study. Authors AJU and FOO managed the literature searches. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/BJI/2018/42026 <u>Editor(s):</u> (1) Chung-Jen Chiang, Department of Medical Laboratory Science and Biotechnology, China Medical University, Taiwan. <u>Reviewers:</u> (1) Arnold Ebuka Irabor, Delta State University, Nigeria. (2) Jimoh Wasiu Adeyemi, Federal College of Animal Health and Production Technology, Nigeria. (3) Martha de Oliveira Guerra, Federal University of Juiz de Fora, Brazil. (4) Yuli Andriani, Universitas Padjadjaran, Indonesia. Complete Peer review History: <u>http://www.sciencedomain.org/review-history/24960</u>

Original Research Article

Received 22nd March 2018 Accepted 31st May 2018 Published 4th June 2018

ABSTRACT

This study investigated the reproductive performance of pawpaw (*Carica papaya*) seeds, hibiscus plant (*Hibiscus rosa-sinensis*) leaves and sweet potato (*Ipomoea batatas*) leaves on some reproductive parameters (ovary weight, mean egg diameter and egg fecundity) in female African catfish (*Clarias gariepinus*). One hundred and twenty (120) juveniles of *C. gariepinus* were collected from the University of Calabar fish farm. The 120 fish were randomly divided into 12 experimental tanks measuring 80x80x80 cm (L x W x H) using a completely randomized design (CRD). Three grams (3 g) of each test plant were incorporated into 1 kg of Coppens feed (3 g/kg) and reformulated into four experimental diets; Treatment A- Control, B- pawpaw seed meal, C- Hibiscus

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leaf meal and D- sweet potato leaf meal. The experiment was done in three replications. The fish were fed twice daily for 6 months. Data obtained were analyzed using a one way analysis of variance (ANOVA). Results obtained revealed that the different test substances significantly (p<0.05) negatively affected the different reproductive parameters studied. The ovary weight, gonadosomatic index (GSI), egg diameter, fecundity as well as total weight significantly (p<0.05) decreased in all the treated fish when compared with that of the control. Pawpaw seed meal (PSM) had the highest effect on the reproductive parameters of the fish studied (ovary weight, GSI, fecundity and egg diameter values of 14.89 ± 5.51 , 0.82 ± 0.30 , 19371 ± 51.84 and 0.63 ± 0.07 , respectively) when compared to the other test plants. The findings of this study suggest that *C. papaya* seeds, hibiscus leaves and sweet potato leaves have the potential to impair reproductive performance in female African catfish. Therefore, holistic measures should always be taken when using these plants considering the effect it could exert on other aquatic inhabitants and systems.

Keywords: Pawpaw; hibiscus; sweet potato; reproductive performance; Clarias gariepinus.

1. INTRODUCTION

Fish farming has contributed immensely to the availability of food globally and in Nigeria. More so, product from fish farming are widely exported and traded by several developing countries to earn income. These products are very rich in nutrients which are important to the body. Statistically, they account for about 10 to 15 percent of dietary protein supply [1]. Fisheries sector employs over 44.5 million people and a lot of them are from developing countries. Also, industries engaged in the marketing, supply and distribution of fish product create job opportunities for over 150 million individuals [2].

The African catfish (Clarias gariepinus) is a species of catfish of the family Clariidae and comprise the most cultivated fish in Nigeria, and highly demanded freshwater fish all over the world due to its resistant to stress, ability to tolerate a wide range of environmental conditions, high stocking densities under culture conditions and relatively fast growth [3-4]. They are found throughout Africa and the Middle East, and live in freshwater lakes, rivers, and swamps, as well as human-made habitats, such as oxidation ponds or even urban sewage systems. Because of high demand of quality fish and fish proteins globally, there have been an increase in various researches in different ways to improve fish fertility to meet the demand and target productivity in aquaculture, with a dramatic movement from synthetic drugs to medicinal plants of natural importance. Medicinal plants that were once considered of no value are now being investigated, evaluated and developed into drugs with little or no side effects [5-7].

On the other hand, plant is one of the richest natural resources occurring in abundance in

Nigeria and the world over. It is a good source of antioxidants which are capable of preventing stress and potential diseases. It has some chemical substances which have the ability to nourish, heal, poison and improve different organisms [8]. Plants are used as a good source of natural medicine. Most leafy vegetables contain proteins, vitamins, carbohydrates, iron, fats, minerals and calcium [9]. However, some plants have been shown to have deleterious impact on aquatic organisms [10–13].

Sweet potato (Ipomoea batatas) belongs to the family convolvulaceae with nearly 1650 predominately tropical species. The genus Ipomoea makes up the largest number of species within the convolvulaceae family. The family is dominated by climbing or twinning woody or herbaceous plants that usually have heart-shaped leaves and funnel-shaped flowers [14]. I. batatas is a tuberous-rooted perennial plant mainly grown annually. The roots are adventitious, mostly located within the top 25 cm of the soil. Some of the roots produce elongated starchy tubers. Tuber flesh colour can be white, yellow, orange and purple while skin colour can be red, purple, brown or white. The stems are creeping slender vines, up to 4 m long. The leaves are green or purplish, cordate, palmate veined, borne on long petioles [15]. I. batatas leaves extracts have alterative, astringent, bactericide, aphrodisiac, laxative and tonic properties [16]. More so, a variety of white sweet potato is eaten raw to treat hypertension, anaemia and diabetes [17] while the root of Ipomoea species is used in the treatment of constipation [18]. Sweet potatoes and it derivatives are powerful antioxidant and may be potent in boosting the immune system and treating fever, asthma, bug bites, burns, catarrh, ciguatera, convalescence, diarrhoea, nausea,

stomach distress, tumors and whitlows. It has also been reported to affect fertility [14–15,19–20].

Hibiscus (Malvaceae) is a genus of herbs, shrubs and trees. Its 250 species are widely distributed in tropical and subtropical regions of the world and are reported to possess various medicinal properties viz; antitumor, antihypertensive, antioxidant, anti - ammonemic [21-25]. About 40 species are found in India. Hibiscus rosa sinensis Linn is a native of China and is a potent medicinal plant. It is a common Indian garden perennial shurb [26] and often planted as a hedge or fence plant. Traditionally, this drug is attributed to antifertility activity in Ayurvedic literature [27]. The flowers have been reported to possess anti-implantation and anti spermatogenic activities [28-29]. The petroleum ether extracts of the leaves and flowers have been shown to potentiate hair growth in vivo and in vitro [30]. Leaves and flowers also possess hypoglycemic activity [31-32]. The mucilage of the leaf has anti - complementary activity [33].

Carica papaya is a soft – wooded perennial plant that has a life span of 5 - 10 years although commercial plantations are usually replanted [34]. It normally grows a single – stemmed tree with a crown of large palmate leaves emerging from the apex of the trunk but plant strands may become multi stemmed when damaged. The fruit, seeds and leaves contained novel biologically active compounds which are potent as therapeutics [35]. C. papaya seeds have been reported to contain glycosides and polyphenols in excess among other compounds such as alkaloids, saponins, flavonoids and quinnones [35]. The seeds of the plant have been reported to possess antifertility potentials in animals [36-37].

This study therefore sought to investigate the reproductive performance of pawpaw (*Carica papaya*) seeds, hibiscus plant (*Hibiscus rosasinensis*) leaves and sweet potato (*Ipomoea batatas*) leaves on some reproductive parameters (ovary weight, mean egg diameter and egg fecundity) in female African catfish (*Clarias gariepinus*).

2. MATERIALS AND METHODS

2.1 Duration and Location of the Study

The study was conducted for six months at the University of Calabar fish farm.

2.2 Collection and Preparation of Plant Samples

The plant samples (seeds of *Carica papaya*, leaves of *Hibiscus rosa-sinensis* and *Ipomoea batatas*) were collected within the University of Calabar campus and authenticated in the herbarium unit of the Department of Botany, University of Calabar.

The samples were washed with clean water, air dried for three weeks, ground using electric blender (Qlink-Q15L40) to get the powdery form and extracted using Soxhlet method with 70 percent ethanol as solvent. The filtrate was obtained using rotary evaporator at 45°C, while the extract was reduced into pastes with hot-air oven at 60°C. The pastes obtained were stored in plastic screw capped bottles, labeled and stored in refrigerator for use.

2.3 Collection of Fish Samples

One hundred and twenty (120) juveniles of *C. gariepinus* were purchased from the University of Calabar fish farm. An average initial body weight of 46.3 g, and 17.7 cm length were obtained using weighing balance (Scout-pro; 3000 g), and a measurement meter, respectively at the time of stocking. The fish were acclimated for 7 days in tanks and the water parameters tested to be ideal, before feeding with commercial feed (Coppens) twice daily (morning and evening) throughout the period of the experiment.

2.4 Experimental Design and Procedure

Twelve experimental tanks measuring; 80cm x 80cm x 80cm (L x W x H) were constructed with an outlet and inlet pipe in the University of Calabar fish farm hatchery complex with each tank was filled with clean water. The 120 fish were randomly divided into four experimental groups using a completely randomized design (CRD) in three replicates with each treatment containing 10 female fish. Sex determination was done through visual examination of the gonad. Three grams (3 g) of each plant extract were incorporated into 1 kilogram of commercial feed (3 g/kg; Coppens). The plants extract made up 75% of each experimental diet. The extracts were dissolved in 5ml dimethylsulphoxide (DMSO) and made into solution with water, and mixed with fish feed homogeneously using a spreader and air dried for 48 hours. This procedure was repeated for each plant and the

prepared diets stored in airtight containers, labeled as follows; Treatment A- Control, Bpawpaw seed meal (PSM), C- Hibiscus leaf meal (HLM) and D- sweet potato leaf meal (SPLM). The physico-chemical parameters of the water were measured using the APHA [38] method of water quality assessment.

2.5 Examination of Reproductive Parameters

Three females fish samples were randomly taken from each treatment tank at table size, dissected under chloroform anesthesia using biological dissecting instruments, and ovaries removed and weighed using electronic weighing balance (Scout-Pro: 3000 g) for the Gonadosomatic Index (GSI).

2.6 Fecundity

Fecundity was determined at table size as the product of the number of eggs in 1 g of the eggmass and total weight of the ovary [39]. For each gravid fish sample, morphometric measurements [TL (cm) and total weight (g)] were taken before eggs were removed. The eggs were removed by dissecting the fish abdomen with a sharp scissors. The eggs were collected and washed in distilled water before weighing to the nearest 0.1 g. The collected eggs were preserved in Gilson fluid for 48 h for easy separation of the eggs from the ovarian tissues before estimation. The diameters of 30 eggs per fish were measured according to protocols outlined by Mesa et al. [40] using a stereomicroscope with an ocular micrometer eye piece.

2.7 Determination of Gonadosomatic Index (GSI)

Gonad developmental stages in female *C. gariepinus* were respectively classified according to Oldorf et al. [41]. Calculations were done using; gonad weight/ total body weight x total number of fish.

2.8 Statistical Analysis

All data collected on the gonad weight, gonadosomatic Index (GSI) and egg fecundity were subjected to analysis of variance (ANOVA) using predictive analysis software (PASW), version 18.0. Significant means were separated using the Least Significant Difference (LSD) at 5% probability level.

3. RESULTS

3.1 Total Weight, Ovary Weight and Gonadosomatic Index (GSI) of Female *C. gariepinus* Fed the Experimental Diets

Gonadosomatic index (GSI) results indicated that the control female C. gariepinus had the highest mean total weight of 1834.22 g with ovary weight and GSI of 26.67 g and 1.45%, respectively when compared with B, C and D. Fish species treated with feed B containing Pawpaw Seed Meal (PSM) had the lowest mean total weight. ovary weight and gonadosomatic index (total weight of 1799.78 g, ovary weight of 14.89 g and GSI of 0.82%, respectively) when compared to other treatments. Female fish fed with Feed C Hibiscus leaf meal (HLM) had average total weight, ovary weight and GSI of 1821.47 g, 21.22 g and 1.15% respectively while animals fed with feed D (SPLM) had mean total weight, ovary weight and GSI of 1810.67 g. 15.56 g and 0.86%, respectively as shown in Table 1.

3.2 Fecundity and Egg Diameter of *C. gariepinus* Fed with the Experimental Diets

Fecundity results of *C. gariepinus* fed with the different diets are presented in Table 2. The control had the highest mean fecundity of 36145 with egg diameter of 0.94 mm and when compared with other treatments. This was followed by fish samples fed with feed containing HLM with mean fecundity of 29340 and egg diameter of 0.89 mm. Fish samples fed feed containing PSM had the lowest mean fecundity of 19371 with egg diameter of 0.63mm while the fish samples fed SPLM had mean fecundity and egg diameter of 20462 and 0.83 mm, respectively.

4. DISCUSSION

The gonadal indices of female *C. gariepinus* fed with the three experimental diets (Table 1) showed that fecundity, mean egg diameter, female ovary weight, and gonadosomatic index (GSI) were significantly (P<0.05) decreased in those fed experimental diets when compared with the control. The total weight, ovary weight and the GSI significantly decreased in those fed experimental diets when compared with control (Table 1) which agrees with the findings of Eyo [42] and Jegede [43]. This may be due to the

Treatments	Total weight (g)	Ovary weight (g)	Gonadosomatic index (%)
А	1834.22 ^a	26.67 ^a ± 7.30	1.45 ^a ± 0.40
В	1799.78 ^d	14.89 ^d ± 5.51	$0.82^{\circ} \pm 0.30$
С	1821.47 ^b	21.22 ^b ± 9.96	1.16 ^b ± 0.53
D	1810.67 ^c	15.56c± 11.14	0.86c ± 0.21

Table 1. Mean total weight, ovary weight and Gonadosomatic index of female fish fed the experimental feed

*Means with different superscript letters along each horizontal array differ significantly (p= .05)

Table 2. Mean fecundity, ovary weight (g) and egg diameter (mm) of fish fed the experimental feed

Treatments	Mean fecundity	Egg diameter (mm)	Ovary weight (g)
А	36145 ^a ± 32.11	$0.94^{a} \pm 0.08$	26.67 ^a ± 7.30
В	19371 ^d ± 51.84	$0.63^{d} \pm 0.07$	14.89 ^d ± 5.51
С	29340 ^b ± 11.2	$0.89^{b} \pm 0.05$	21.22 ^b ± 9.96
D	20462 ^c ± 53.22	$0.83^{\circ} \pm 0.05$	15.56 ^c ± 11.14

*Means with different superscript letters along each horizontal array differ significantly (p= .05)

phytochemical constituent of the plants such as saponins, alkaloids, terpenoids, flavonoids, etc., which might have altered the biosynthetic processes growth underlying the and development of the fish and its ovaries in the treatment groups in comparison with the control. Moreover, among the different treatments, SPLM had the highest total length, ovary weight and GSI while fish samples fed with PSM had the least. This implies that the PSM had more effect on the reproductive indices studied than the other treatments. This can be attributed to type and/or the quantity of phytochemicals contained in pawpaw seeds that might have been responsible for the adverse effects on the fish when compared with the other two plants. This is corroborated by Kushwaha [44] and Ikpeme [36] who reported antifertility potential of pawpaw seeds in albino rats. This findings is also supported by Udoh and Kehinde [37] who also observed antifertility properties of pawpaw seeds in albino rats.

Results also revealed that the mean fecundity and egg diameter of the fish significantly decreased in the treatment groups when compared with the control (Table 2). Fecundity, defined as the number of eggs carried by a gravid female fish, is very important aspect of fish culture since it is concerned with the average reproductive characteristics of fish [42]. Results obtained on the fecundity and egg diameter of the fish corroborate results obtained on the total length, ovary weight and gonadosomatic index. This is in view of the fact that fecundity in fish has been reported to have positive significant correlation with total length and ovary weight [42]. This implies that a decrease in the ovary weight and egg diameter will result in decrease in fecundity. This assertion is supported by Phukon and Biswas [45] and Buragohain and Goswami [46].

5. CONCLUSION

Findings of this study provide evidence that seeds of *C. papaya* and leaves of *Hibiscus rosa* – *sinensis* and *I. batata* have effect on ovary weight, GSI, egg diameter and fecundity of African catfish. Therefore, holistic measures should always be taken when using these plants considering the effect it could exert on other aquatic inhabitants and systems.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history: The peer review history for this paper can be accessed here: http://www.sciencedomain.org/review-history/24960