

Gonadosomatic and Hepatosomatic Indices of *Clarias gariepinus* fed *Moringa oleifera* Leaf Meal Finisher Diets

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ABSTRACT

The gonadosomatic index, GSI which expresses gonad weight as a percentage of total body weight, is an important indicator of reproductive development, gonadal maturation, and reproductive readiness in fish. The HSI reflects liver participation in vitellogenesis. The aim of this study was to investigate the effects of partial replacement of fishmeal with *Moringa* leaf meal, MLM on liver and gonads of *Clarias gariepinus* when used in finisher diet. Four diets were formulated to contain 35% crude protein. MLM was included at 0 % (control), 6.5 %, 13 % and 19.5 % in the diets. *Clarias gariepinus* juveniles of average weight 96.0784 ± 5.0 g and length 19.60 - 22.51cm were used for the 90 days feeding trial. Feeding was to satiation twice daily. The highest mean GSI in males was observed in fish fed 6.5% MLM, followed by the control group (0.50 ± 0.13). Female *C. gariepinus* exhibited considerably higher GSI values than males. Female GSI ranged from 7.14 ± 1.95 to 14.39 ± 1.49 , with significant difference observed among treatments ($P < 0.05$). Fish fed 0% MLM (13.66 ± 3.46) and 6.5% MLM (14.39 ± 1.49) were not significantly different ($P > 0.05$). HSI did not follow a dose-dependent pattern in males and remained within normal physiological range with the highest value (1.33 ± 0.34) obtained at 6.5% MLM inclusion. Female *C. gariepinus* exhibited HSI values ranging from 1.03 ± 0.08 to 1.51 ± 0.03 . The control diet produced the highest HSI (1.51 ± 0.03) as females showed a reduction in HSI with increasing MLM inclusion. Findings posit that 6.5 % dietary inclusion of MLM in finisher-diet supports efficient nutrient partitioning between hepatic metabolism and reproductive development while maintaining normal liver physiology.

Keywords: Finisher-diet; Reproductive development; Gonadal maturation; Hepatic stress; Liver function.

INTRODUCTION

Feed is formulated from an array of complementary feed materials to effectively meet fish nutrient needs, for optimum physiological function, including protecting their natural defense systems, growth, and reproduction (Helmiati and Isnansetyo, 2020). Hence blending local feed sources with complementary amino acid profiles is a useful strategy to improve the quality of the protein. Use of alternative feed resources potentially boosts fish culture having merits in reduced cost, environmental sustainability and foreign exchange saving. Alternative feed resources are feed items sourced from the farmers immediate environment or not previously used that contain nutrients necessary for the targeted growth and production of cultured fish without negative effect on the environment. *Moringa oleifera* is an alternative plant protein source in aqua-feeds for its rich nutrient composition (Alphonse *et al.*, 2021), and good digestibility (Kasiga and Lochmann, 2014).

African catfish *Clarias gariepinus* is an important aquaculture species being the most important commercially farmed freshwater fish in Africa (Ogunji and Wuertz, 2023), a good species for peri-urban aquaculture, and most popular fish on the Nigeria local market.

C. gariepinus exhibit obvious sexual dimorphism at maturity. The males have an obvious tube-like genital papilla. Mature females can be recognized under visual examination because the belly of brood fish ready for spawning is soft, relatively broad and rounded. The genital opening is swollen, reddish and round-ended. They mature between the ages of 8 – 10 months The weight of the ovaries relative to body weight (gonadosomatic index, GSI) can reach 25 percent of body weight and may occupy as much as 80% of the abdominal cavity.

Aquaculture practices, including proper feeding, management, and reduced exposure to environmental stressors, influence gonadal development and liver function in fish (Villasante *et al.*, 2025).

Organo-somatic indices can be used as indices of change in nutritional and energy status (Maxwell and Dutta, 2005) and as well linked to effects of chemical on target organs like the gills, liver and kidney (Vighi, 2010). It is used to assess the general health, and the influence of environmental factors on fish (Ariweriokuma *et al.*, 2011; Dekić *et al.*, 2016). Gonadosomatic and hepatosomatic indices are organosomatic components which can infer the reproductive development as well as preparedness in fish.

Gonadosomatic index (GSI)

Gonadosomatic index is defined as the ratio of the fish gonad weight to body weight expressed in percentage (%) (Reno Irawan *et al.*, 2023). In fisheries science and aquaculture, it is an important tool that is useful in the evaluation of reproductive condition. The GSI is used to detect hydrated ovaries in females as well as testis in males and therefore it helps in detection of the reproductive period from weight increase. High GSI value is indicative of mature stage of gonads in fish. The average weight gain of female fish gonads will increase by 10–25% of their body weight, while in male fish it will increase by 5–10% of their body weight (Reno Irawan *et al.*, 2023). GSI is a critical metric for assessing fish reproductive health, maturity stages, and energy allocation. It reflects whether a fish has adequate nutrition, clean habitat conditions, and proper endocrine function for breeding. GSI rises during gonad maturation and peaks right before spawning, dropping sharply after gametes release. Higher GSI values in *Clarias gariepinus* indicate better readiness for artificial propagation. Females typically display much higher GSI peak than males due to larger ovary and egg mass.

Environmental and nutritional stress such as exposure to heavy metals or industrial toxicants, suboptimal temperatures, poor dissolved oxygen level, inadequate protein or restricted rations, suppresses gonadal development, lowering GSI values (Gbaaondo *et al.*, 2025). Findings documented by different researchers have indicated that the quality of aqua feed affects fecundity and gonad development in fish (Ekanem *et al.*, 2012; Ekanem *et al.*, 2013 and Bassey *et al.*, 2018). Limited studies documented on the effect of *M. oleifera* leaf meal on the fecundity and gonado-somatic index of the African catfish. Ibiyo *et al.* (2022) reported improved gonadal development when *M. oleifera* leaf meal was incorporation in the diet of *H. longifilis* at 15% level.

Hepatosomatic index (HSI)

The hepatosomatic index (HSI) is the ratio of liver weight to total body weight. It acts as a key biomarker for fish health, reflecting energy storage, nutritional status, and response to environmental stress or water pollution. It is an important aspect of the development of gonadal maturity because it can describe the energy reserves in the fish body when the fish is experiencing the development of gonadal maturity. The liver condition will change due to the accumulation of energy in the form of fat in the liver, where this energy will be used for the growth and development of gonads (Lodeiros *et al.*, 2001). Information on the general health condition of a fish can be obtained through hepato-somatic index determination. The size or weight of the liver and spleen relative to body weight may assist in signifying the overall health of the fish (Idam, 2022). Examining of hepatosomatic indices is crucial in understanding fish metabolism, particularly in relation to digestion, nutrient absorption, the synthesis and secretion of digestive enzymes (Samad *et al.*, 2025). A normal HSI shows the fish has enough food and good energy reserves. Low HSI means the fish is lacking proper nutrients, forcing it to use up liver energy stores. Poor or unbalanced feed can alter liver size and metabolism in culture fish. Ibiyo *et al.* (2022) observed no significant difference in the hepatosomatic index when they included 15% moringa leaf meal in *C. gariepinus* diet. There was direct relationship between liver and carcass weight in juvenile salmon fed diet with a higher carbohydrate-to-lipid ratio Li *et al.* (2015). Ayo-Olalusi *et al.* (2021) also recorded no negative effect on the liver, intestine and gonad when fish meal was replaced with sweet potato leaf meal up to 2.5 – 7.5% in *C. gariepinus* diets. However, Villasante *et al.* (2025) recorded significantly lower HSI in fish fed high plant meal diet. Exposure to heavy metals or toxins causes liver stress leading to abnormal liver enlargement (hypertrophy) or shrinking (atrophy), altering the HSI (Fiaz *et al.*, 2024). Poor water conditions lower the overall health of the fish and reflect directly as abnormal HSI. The liver grows active in producing vitellogenin (yolk protein) before

spawning. Stored liver fats and proteins are moved to the developing eggs, which temporarily reduces the HSI during active spawning.

M. oleifera has been commonly used in diet of cultured fish species, but its effect on liver and gonadal development has not been commonly examined. This study investigated the effects of partial replacement of fishmeal with *Moringa oleifera* leaf meal on liver and gonads of *Clarias gariepinus* when used in finisher diet.

MATERIALS AND METHODS

Sourcing of *Moringa oleifera* leaf meal and other feed items

The research was conducted at the teaching and research farm of the Department of Fisheries and aquaculture, Delta State University Abraka, Nigeria. Fresh moringa leaves were sourced locally in the Delta State University Abraka campus environ. The leaves were soaked in water overnight to remove saponin and other soluble anti-nutritional factors according to Hussain *et al.*, (2018) and Rawat and Saini (2022). Thereafter, water was drained and the leaves were air-dried at ambient temperature under shade, away from direct sunlight to prevent loss of vitamins through photodynamic damage or oxidation (Kamran *et al.*, 2020). The dried leaves were threshed from stalks, ground with hammer mill to obtain moringa leaf meal (MLM), and stored in plastic bags at room temperature. Other feed ingredients were sourced from feed outlets.

Diet formulation and feed preparation

The feed ingredients were ground and sieved to fine particle size. Four diets (Table 1) were formulated to contain 35% crude protein (35 g.100 g⁻¹). MLM was included at 0 % (control), 6.5 %, 13 % and 19.5 % in the diets. The choice of these nutrient source and levels, particularly protein, is intended to depict the practical farm-made diets for grow-out. During compounding of diets, the relevant proportions of the ingredients was weighed and then mixed properly by hand. The mixture was extruded with a 3 mm die and the resulting pellets dried, packed in polythene bags and stored.

Table 1: Gross ingredient composition of experimental diets

Ingredients	Moringa leaf meal inclusion			
	0%	6.50%	13%	19.50%
	(Control)	(Treatment 1)	(Treatment 2)	(Treatment 3)
Fish trash meal 47.4%	35	32.5	30	27.5
MLM 22.5%	-	6.5	13	19.5
Soybean 40.12%	25.26	26.64	27.41	28.19
GNC 39.68%	12.63	13.32	13.71	14.09
Maize 8.93%	13.4	9.36	5.91	2.47
Wheat offal 9%	6.7	4.68	2.96	1.24
Vitamin premix	1	1	1	1
Mineral premix	1	1	1	1
Wheat flour	2	2	2	2
Palm oil	2	2	2	2
Bone meal	0.5	0.5	0.5	0.5
Total (g)	100	100	100	100
Calculated CP (%)	35	35	35	35
Analyzed CP (%)	33.52	34.07	34.34	34.6

MLM- moringa leaf meal, GNC- groundnut cake, D1 - D4 – Experimental diets

Experimental procedure and feeding

The experiment was laid out in a completely randomized design consisting of four moringa leaf meal (MLM)

inclusion levels as treatments. One hundred and eighty (180) *Clarias gariepinus* juveniles of average weight 96.0784 ± 5.0 g and length 19.60 - 28.51 cm were used for the 90 days feeding trial. The initial body weight and total length of fish were taken according to Ekelemu *et al.* (2022) using electronic weighing balance for weight to the nearest gram and metre rule for length (individual fish length) to the nearest 0.1 cm. The fish, 15 juveniles were placed into each of 12 tarpaulin tanks measuring 1 m x 1 m x 1 m. The tanks containing fish were then randomly assigned to the diets and grouped in threes to give four treatments replicated three times. Fish were fed by hand to satiation twice daily at 09:00 h and 16:00 h. Sampling for weight and length measurements were done once in two weeks during which weight of feed consumed was also recorded.

Water quality measurement

Water from the bore hole in the farm was used. Culture water was renewed thrice weekly. The water quality parameters such as temperature, pH, Ammonia, Nitrite-N, Nitrate-N and Dissolved Oxygen concentration were measured. Temperature was measured using mercury in glass thermometer; pH was measured with a digital pH meter and DO, Ammonia, Nitrite-N and Nitrate-N were estimated following standard procedures (APHA, 2012).

Harvesting of gonads and liver

A sample of 12 matured females and males respectively from each treatment, four from each replicate tank, were randomly selected and their mean body weight recorded. Selected males had distinct pointed papilla while the females had rounded urogenital opening with slightly swollen abdomen. Percussion stunning was applied on the skull of each fish to make them unconscious. The samples were dissected and the gonads (Ovaries and milt sac), and liver taken out intact.

Gonadosomatic index (GSI)

Weight of gonads were taken and mean GSI values calculated. This was done as a percentage of the mean gonads weight (GW) relative to the mean body weight (BW) of the fish (Afonso-Dias *et al.*, 2005; Jan and Ahmed, 2016) for each tank.

$$GSI = \frac{GW}{BW} \times \frac{100}{1}$$

Hepatosomatic index (HSI)

The HSI was calculated according to the method of (Jan and Ahmed, 2016) by determining the mean weight of hepatopancreas as a percentage of the mean body weight of the fish for each replicate tank. Where LW and BW are liver weight and body weight in both sexes, respectively.

$$HSI = \frac{LW}{BW} \times \frac{100}{1}$$

Data analysis

One-way ANOVA was used to analyse replicate tank data collected to test for variation among treatment means. Duncan's multiple range test was used to determine significant differences among the treatments means at $P \leq 0.05$ using IBM SPSS Statistical software (version 26.0).

RESULTS AND DISCUSSION

Water quality parameters

The range of water quality parameters recorded throughout the feeding trial involving *Clarias gariepinus* fed graded levels of *Moringa oleifera* leaf meal (MLM) finisher diet is given in table 2.

Table 2: Water quality parameters during the trial

Parameter	Observed range
Temperature (°C)	25.30 - 29.41
pH	6.60 - 7.60
Dissolved oxygen (mgL ⁻¹)	5.70 - 6.30
Amonia (mgL ⁻¹)	0.04
Nitrite (mgL ⁻¹)	0.00 - 0.01
Nitrate (mgL ⁻¹)	47.00 - 49.00

Water temperature ranged from 25.30 to 29.41°C during the experimental period. This temperature range falls within the optimum for *Clarias gariepinus* culture which generally thrives between 25 and 30°C. The relatively stable temperature observed provided favourable conditions for normal metabolic activities and minimized environmental stress on the fish. The pH values ranged from 6.60 to 7.60, indicating slightly acidic to neutral water conditions. This range is considered suitable for the culture of *C. gariepinus*, which tolerates pH values between 6.5 and 8.5. Dissolved oxygen concentrations ranged from 5.70 to 6.30 mg L⁻¹ throughout the study. These values exceed the minimum oxygen requirement for African catfish culture, above 5 mg L⁻¹ (Bulbul Ali and Mishra, 2022). Dissolved oxygen is critical for aerobic respiration, energy production, nutrient metabolism, and feed utilization. The high dissolved oxygen concentrations indicates that oxygen shortage did not interfere with the dietary responses observed. Ammonia concentration remained at 0.04 mg L⁻¹ throughout the experiment. This value is considerably below concentrations known to impair growth or induce toxicity in cultured fish. Nitrite concentrations were negligible ranging from 0.00 to 0.01 mg L⁻¹, during the experiment. Bian *et al.* (2025) reported that for freshwater aquaculture species, safe nitrite concentrations is below 0.1 mg L⁻¹. The concentration of nitrate ranged from 47.00 to 49.00 mg L⁻¹. Excessive accumulation over prolonged periods may contribute to physiological stress and reduced reproductive performance. The concentrations recorded were within acceptable limits (below 50–100 mg L⁻¹) for freshwater aquaculture (Schram *et al.*, 2014).

Gonadosomatic index (GSI)

The gonadosomatic index is an important indicator of reproductive development, gonadal maturation, and reproductive readiness in fish. Table 3 presents the gonadosomatic index (GSI) of male and female *Clarias gariepinus* fed graded levels of *Moringa oleifera* leaf meal in finisher-diet.

Male Gonadosomatic index (GSI)

In males, the GSI values ranged from **0.40 ± 0.09** in treatment 3 (19.5% MLM) to **0.56 ± 0.12** in treatment 1 (6.5% MLM). The highest mean GSI was observed in fish fed 6.5% MLM, followed by the control group (**0.50 ± 0.13**), while lower values were recorded in fish fed 13% and 19.5% MLM. Although a gradual decline was observed beyond the 6.5% inclusion level, but the observed differences were not statistically significant ($P > 0.05$). This indicates that dietary MLM exerted little influence on male gonadal development within the inclusion levels evaluated. The relatively stable male GSI values suggest that spermatogenesis and testicular development were maintained despite increasing dietary MLM. Similar findings have been reported by Rwodzi (2014) in goats; Khan *et al.* (2024) and Nyingwa *et al.* (2026) in bucks. They observed that moderate dietary supplementation with phytogenic feed additives generally does not adversely affect male reproductive performance because spermatogenesis requires comparatively lower energy investment than oogenesis. Findings in this study corroborates Nyingwa *et al.* (2026) who stated that the bioactive components of moringa, including flavonoids, alkaloids, tannins, and saponins, have antioxidant, anti-inflammatory, and hormone-modulating effects that collectively improve reproductive outcomes. In contrast, Ampofo-Yeboah, (2013) observed disruption in gonad function, differentiation and sexual maturation of Mozambique tilapia fed moringa and pawpaw seeds based diets.

Female Gonadosomatic index (GSI)

Female *C. gariepinus* exhibited considerably higher GSI values than males, reflecting the naturally greater reproductive readiness associated with ovarian development and egg production. Female GSI ranged from 7.14 ± 1.95 to 14.39 ± 1.49 , with statistically significant differences observed among treatments ($P < 0.05$). Fish fed the control diet (13.66 ± 3.46) and treatment 1 (14.39 ± 1.49) were not significantly different ($P > 0.05$) indicating that the inclusion of 6.5% MLM maintained ovarian development comparable to that of the control group. Treatment 2 (13% MLM) produced an intermediate GSI value of 9.04 ± 3.92 , which was not significantly different from either the higher or lower groups. However, treatment 3 (19.5% MLM) recorded the lowest female GSI (7.14 ± 1.95), which indicates a significant reduction. These results demonstrate that high dietary inclusion of MLM reduced ovarian development and reproductive readiness in female *C. gariepinus*.

The superior female GSI observed in the control and 6.5% MLM diets suggests that moderate inclusion of *Moringa oleifera* leaf meal does not impair reproductive development and may provide sufficient nutrients to support gonadal maturation. Moringa leaves are rich in high-quality protein, essential amino acids, vitamins (particularly vitamins A, C, and E), minerals, carotenoids, and antioxidants that contribute to reproductive health by protecting reproductive tissues against oxidative stress and supporting steroid hormone synthesis (Zeeshan *et al.*, 2024; Ali and Khatun, 2025 and Mohai Ud Din *et al.*, 2026). Antioxidants such as flavonoids and phenolic compounds help preserve ovarian integrity by reducing lipid peroxidation and oxidative damage during oocyte development (Nyingwa *et al.*, 2026). According to Saleh *et al.* (2025), phytogenic feed additives enhance reproductive performance by improving antioxidant status, nutrient utilization, and endocrine function in cultured fish. Similarly, Sumana *et al.* (2025) reported that moderate dietary supplementation with *Moringa oleifera* improved physiological performance and reproductive potential.

The decline in female GSI observed at 13% and 19.5% MLM inclusion suggests excessive dietary incorporation which compromised ovarian development. This possibly resulted from increased dietary fibre and concentration or synergistic effect of anti-nutritional compounds such as tannins, saponins, phytates, and alkaloids associated with higher MLM inclusion. These compounds may reduce nutrient digestibility, impair amino acid absorption, and decrease the availability of essential nutrients required for vitellogenesis and ovarian growth. Corroborating, Adetunji *et al.* (2025) reported that excessive inclusion of plant-derived feed ingredients may impair reproductive performance by reducing nutrient digestibility and limiting energy allocation to gonadal development.

Table 3: Gonadosomatic indices (GSI) of <i>C. gariepinus</i> fed MLM finisher diet (Mean $\pm$ SD)		
Treatment diet	GSI (male)	GSI (female)
Control (0%)	0.50 $\pm$ 0.13	13.66 $\pm$ 3.46 ^a
Treatment 1 (6.5%)	0.56 $\pm$ 0.12	14.39 $\pm$ 1.49 ^a
Treatment 2 (13%)	0.43 $\pm$ 0.07	9.04 $\pm$ 3.92 ^{ab}
Treatment 3 (19.5%)	0.40 $\pm$ 0.09	7.14 $\pm$ 1.95 ^b
Means in the same column with same superscript are not significantly different at $P > 0.05$		

The increased dietary fibre associated with higher MLM inclusion also reduced metabolizable energy available for reproduction. Reproductive processes, especially oocyte maturation, require substantial energy reserves. When dietary energy becomes limiting, fish preferentially allocate nutrients to maintenance and survival rather than reproduction, resulting in lower GSI values. Balanced dietary protein and energy are essential for efficient nutrient partitioning between somatic growth and reproductive development (Radhakrishnan *et al.*, 2020; Rajendran *et al.*, 2022). Likewise, Nyingwa *et al.* (2026) noted that female fish exhibit greater sensitivity to nutritional deficiencies because ovarian maturation represents one of the most energy-demanding physiological processes.

The observed sex-related differences in GSI are consistent with the reproductive biology of teleost fishes. Female fish naturally exhibit much higher GSI values because ovaries occupy a substantial proportion of body weight during maturation, whereas testes remain comparatively small throughout the reproductive cycle.

Consequently, dietary effects are generally more pronounced in females than in males. The present findings therefore indicate that while male reproductive development remained relatively stable across dietary treatments,

female ovarian development was significantly influenced by increasing MLM inclusion, particularly at the highest dietary level.

The significant reduction in female GSI at 19.5% MLM may also reflect the cumulative physiological effects of anti-nutritional factors present in moringa leaves. Although these compounds occur at relatively low concentrations after feed formulation, prolonged consumption at higher dietary inclusion levels may reduce mineral bioavailability, inhibit digestive enzymes, alter endocrine regulation, or interfere with reproductive hormone synthesis. Abdel-Huang *et al.* (2024) reported that processing significantly enhances the nutritional utilization of plant-based ingredients while minimizing their adverse effects on reproductive performance.

Hepatosomatic index (HSI)

The hepatosomatic index is a widely used physiological indicator of liver condition, energy reserve status, nutrient metabolism, and overall fish health. Table 4 presents the hepatosomatic index (HSI) of male and female *Clarias gariepinus* fed graded levels of *Moringa oleifera* leaf meal (MLM) in finisher diets.

Male Hepatosomatic index (HSI)

In the male *Clarias gariepinus*, the control diet recorded an HSI of 1.09 ± 0.11 , while treatment 1 (6.5% MLM) produced the highest value (1.33 ± 0.34). The HSI subsequently decreased to 1.01 ± 0.12 in treatment 2 (13% MLM) before increasing slightly to 1.23 ± 0.13 in treatment 3 (19.5% MLM). Although these fluctuations did not follow a clear dose-dependent pattern, they remained within the normal physiological range reported for healthy African catfish, 0.8 to 3.5 % (Adebambo *et al.*, 2026). The slightly elevated HSI observed in Treatment 1 may indicate greater hepatic energy storage resulting from efficient nutrient utilization and active metabolic processes. At moderate dietary inclusion levels, *Moringa oleifera* contains bioactive compounds, antioxidants, vitamins, and essential nutrients that may improve hepatic metabolism and protect liver tissues from oxidative stress. Similar findings were reported by Owais *et al.* (2023), who demonstrated that phytogenic feed additives enhance liver function through antioxidant activity and improved metabolic efficiency. Likewise, Emam *et al.* (2024) reported that moderate dietary supplementation with moringa improved physiological performance without inducing hepatic abnormalities in tilapia.

The reduction in male HSI observed in Treatment 2 may indicate greater mobilization of hepatic energy reserves to support growth and metabolism rather than accumulation within the liver. Fish exhibiting efficient somatic growth often utilize stored hepatic glycogen and lipids for tissue synthesis, resulting in relatively smaller liver indices despite satisfactory physiological performance. This agrees with Khormi,(2026), who noted that lower HSI values are not necessarily indicative of impaired liver function but may instead reflect efficient nutrient partitioning toward muscle growth.

Table 4: Hepatosomatic indices (HSI) of male and female *C. gariepinus* fed MLM in finisher diet (Mean $\pm$ SD)

Treatment diet	HSI (male)	HSI (female)
Control (0%)	1.09 $\pm$ 0.11	1.51 $\pm$ 0.03
Treatment 1 (6.5%)	1.33 $\pm$ 0.34	1.03 $\pm$ 0.42
Treatment 2 (13%)	1.01 $\pm$ 0.12	1.09 $\pm$ 0.23
Treatment 3 (19.5%)	1.23 $\pm$ 0.13	1.03 $\pm$ 0.08

The slight increase in HSI observed in Treatment 3 suggests that the highest MLM inclusion level did not produce severe hepatic enlargement or degeneration despite the increased dietary fibre and phytochemical contents.

Female Hepatosomatic index (HSI)

Female fish exhibited HSI values ranging from 1.03 ± 0.08 to 1.51 ± 0.03 . The control diet produced the highest HSI (1.51 ± 0.03), whereas Treatments 1 and 3 recorded identical values of 1.03, and Treatment 2 produced an

intermediate value of 1.09 ± 0.23 . Unlike males, females showed a reduction in HSI following MLM inclusion. This difference may be associated with sex-related physiological variation in nutrient allocation and reproductive metabolism. Female fish generally possess greater hepatic activity because the liver synthesizes vitellogenin and other yolk precursor proteins required for ovarian development. Consequently, larger liver size is often observed in females, particularly when reproductive maturation is progressing. The relatively higher HSI recorded in the female control group may therefore reflect greater hepatic nutrient storage rather than any pathological condition. According to Qiao *et al.* (2016), sex-related differences in HSI are common in teleost fish because female liver metabolism is closely linked to reproductive physiology.

The lower HSI values observed in females receiving MLM diets may indicate more efficient utilization of hepatic nutrient reserves rather than reduced liver function. The antioxidant compounds present in *Moringa oleifera*, including flavonoids and phenolic compounds, may improve hepatic metabolic efficiency and reduce oxidative damage without promoting excessive liver enlargement. Owais *et al.* (2023) reported that phytogetic feed additives improve hepatic antioxidant status and cellular integrity, thereby enhancing liver efficiency while maintaining normal organ size. Similarly, Okon *et al.*, (2025) and Sumana *et al.*, (2025) observed that phytogetic compounds stimulate metabolic regulation and reduce physiological stress in cultured fish through improved antioxidant defence mechanisms.

The absence of a consistent increase in HSI with increasing dietary MLM suggests that liver enlargement did not occur despite the presence of phytochemicals and moderate levels of anti-nutritional compounds in the diets. Excessive dietary anti-nutritional factors such as tannins, saponins, and phytates may induce hepatic stress, increase detoxification activity, and result in enlarged livers. However, the relatively stable HSI values obtained in this study indicate that the inclusion levels of MLM did not produce adverse hepatic responses. This observation agrees with Miryan *et al.* (2025), who reported that moderate inclusion of plant-derived feed ingredients does not compromise liver health when diets remain nutritionally balanced. Conversely, several investigators have reported that excessively high inclusion of unprocessed plant leaf meals may increase HSI because the liver must metabolize greater quantities of phytochemicals and detoxify anti-nutritional compounds (Huang and Bu, 2022; Adetunji *et al.*, 2025). The present findings therefore suggest that the dietary inclusion levels used in this study remained within the physiological tolerance of adult *C. gariepinus*.

The relatively low standard deviations observed in most treatments further indicate that liver responses were fairly uniform among individual fish, although greater variation occurred in females receiving treatment 1 (± 0.42) and males receiving treatment 1 (± 0.34). This variability may reflect natural biological differences in energy storage, reproductive status, or individual metabolic responses rather than treatment effects.

Dietary inclusion of *Moringa oleifera* leaf meal up to 19.5% did not adversely affect liver condition or metabolic health in adult *Clarias gariepinus*. Both male and female fish maintained HSI values within the normal physiological range, 1.0 - 2.5 % (Bwala *et al.*, 2026) throughout the feeding trial. This suggests that hepatic energy metabolism and nutrient storage remained largely unaffected. The slight fluctuations observed among treatments most likely reflect normal metabolic adaptation to dietary composition and sex-related physiological differences rather than pathological changes. These findings indicate that moderate dietary inclusion of MLM supports normal liver function while providing the nutritional and phytogetic benefits associated with *Moringa oleifera*.

CONCLUSION

Moderate dietary inclusion of MLM supports efficient nutrient partitioning between hepatic metabolism and reproductive development while maintaining normal liver physiology. Consequently, the results provide evidence that **6.5% MLM represents an appropriate inclusion level for finisher diets**, balancing metabolic health with optimal reproductive performance in *C. gariepinus*. The findings also demonstrate that female reproductive tissues are considerably more sensitive to dietary composition than male reproductive tissues. The results support the use of MLM at **6.5% inclusion** in finisher diets when maintaining reproductive performance of *C. gariepinus* is an important production objective. However, dietary inclusion of *Moringa oleifera* leaf meal up to **19.5%** in finisher diets did not alter liver size or compromise hepatic function in adult *Clarias gariepinus*.

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