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Effect of Roselle (*Hibiscus sabdariffa*) and ginger (*Zingiber officinale*) as feed additives, on growth and haematology of *Clarias gariepinus* Juvenile

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The rebirth and use of medicinal plants in aquaculture has become necessary as the use of synthetic drugs and chemicals is been discouraged due to their aftermath effects on cultured organisms and aquatic environment. A 56-day study was conducted to assess the effects of Roselle and ginger as dietary additives, on growth and hematology of *Clarias gariepinus* juvenile. Total of 150 *C. gariepinus* juveniles (35.41±1.45 g) were assigned to five iso-nitrogenous diets as treatments having ginger and roselle added as additives at varying inclusion levels of 0.0, 2.0, 4.0, 2.0 and 4.0 g/100 g. Best growth performance was observed in 4.0 g ginger treated fish followed by 4.0 g roselle fed fish group, while 2.0 g roselle fed fish had the lowest growth performance. Significant changes ($p<0.05$) were observed in the haematology of *C. gariepinus* fed varying inclusion levels of ginger and roselle. Highest values for red blood cells (4.07±0.08), haemoglobin (11.61±0.57) and pack cell volume (34.33±0.88) were seen in 4.0 g ginger treatment group followed by the control (3.63±0.22, 9.93±0.92 and 30.33±2.73), respectively. No significant changes were observed in red blood cells indices (mean corpular volume, mean corpular hemoglobin and mean corpular hemaglobin concentration). The current study revealed that fish fed 4.0 g ginger diet had better growth and haematological profile.

Key words: Phytobiotics, growth, hematology, *Clarias gariepinus*.

INTRODUCTION

Aquaculture expansion and development in developing countries has been hampered by various factors which includes environmental degradation and reduced water quality, underdeveloped credit markets, conflicts over the

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use of land and water resources, lack of governance and regulation in production, poorly developed infrastructure, limited access to fish seed, feed, increased fish feed extraction from the world's ocean and diseases (World Bank, 2006). Disease which is one of the critical factors hampering aquaculture development ensued as a result of negative interactions between fish and pathogenic bacteria (Romero et al., 2012; Iheanacho et al., 2017a).

Antibiotics are chemical compounds used in treating and preventing diseases in aquaculture. However, the use of antibiotics in fish feeds has a good impact on growth (Rico et al., 2013). Antibiotics growth promoters (AGP) were supposed to increase growth rate as a result of improved gut health, resulting in better nutrients utilization and improved feed conversion (Visek, 1978). But now, antibiotics growth promoters are considered as human health risk factor for their possible role in the emergence of microbial resistance, breakage of the animal intestinal micro-ecological balance and the presence of antibiotics residues in resultant fish products (Okeke and Ososa, 2003). Feed supplements are costly to the extent that most fish farmers depend on it to supplement some essential minerals and vitamins found wanting in fish feeds but currently, can no longer afford to use them.

Natural materials such as medicinal plants could be widely accepted as feed additives to enhance feed utilization and aquaculture productive performance and sustainability (Levic et al., 2008). Phytogenic feed additives, also known as phytobiotics products are plant derived products, used in animal feeding to improve performance through amelioration of feed properties, promotion of production performance and improving the quality of animal origin food/feed (Alector and Osho, 2009). World Health Organization encourages using medicinal herbs and plants to substitute or minimize the use of chemicals through the global trend of go back to nature (Adewole, 2014).

Roselle (*Hibiscus sabdariffa*) is an annual dicotyledonous, erect, herbaceous tropical plant. The plant is cultivated majorly in the northern part of Nigeria as edible vegetable and considered to be medicinal (Ijeomah et al., 2012). The chemical constituents of the flower include the flavonoids, gossypetine and sabdaretine (Pietta, 2000). Roselle is reported to be diuretic, digestive, antiseptic, sedative, purgative, emollient, demulcent and astringent (Adewole, 2014). The calyces have many medicinal applications in curing kidney stone, pyrexia, liver damage, hypertension and leukemia (Abu-Tarboush et al., 1997; Estrella et al., 2000).

Ginger (*Zingiber officinale*) is also a herbaceous perennial plant which grows annual stems about a meter tall, bearing narrow green leaves and yellow flowers (Kim et al., 2008). Ginger has been reported to be rich in mineral elements (magnesium, potassium, phosphorus, calcium and zinc), vitamins (retinol, cholecalciferol, ascorbic acid, thiamine, riboflavin, niacin, folic acid, pantothenic acid and pyridoxine) and phytochemicals (flavonoid and alkaloid) (USDA, 2014; Iheanacho et al., 2017a). Shubha (2015) reported that ginger rhizomes also contain gingerol, shogoal and a potent proteolytic enzyme called zingibain. The author also reported that ginger is anti-platelet, antibacterial, antifungal, antiviral, anti-worm, anti-inflammatory and has anti-oxidative activity.

The use of medicinal plants in aquaculture has not being widely practised. The current study seeks to evaluate the effects of roselle and ginger supplemented diets as feed additives, on growth and haematology of African catfish (*Clarias gariepinus*).

MATERIALS AND METHODS

Study area

The study was carried out at the Animal House of the Department of Microbiology/Biotechnology, Federal University, Ndufu Alike Ikwo, Ebonyi State, Nigeria.

Study fish

One hundred and fifty (150) *C. gariepinus* juveniles were procured from a private fish farm in Abakaliki and transported in a 50 L gallon filled with water to the Animal House of the Department of Microbiology/Biotechnology, Federal University, Ndufu Aliko Ikwo within 30 and 40 min. The fish were acclimated in a tarpaulin tank (4 m x 2 m x 1 m) for two weeks and were fed commercial fish feed (coppens) throughout the 2 weeks acclimation period.

Collection, preparation and processing of Roselle flower (*Hibiscus sabdariffa*) and ginger (*Zingiber officinale*)

The dried flower (calyx) of roselle and rhizomes of ginger were purchased from Abakpa Market located at Abakalik town, Ebonyi State, Nigeria. Both materials were ground into powder, using electric blender (Philips, model HRI701/BC, made in China), thus sieved, and stored separately in an air tight container until use.

Experimental diets

Five iso-nitrogenous diets were formulated to yield 37% crude protein (Table 1). Roselle and ginger were included in the diets as feed additives at varying inclusion levels and coded as HS1 and HS2 for Roselle, while G1 and G2 were for ginger. The different inclusions were HS1 (2.00 g), HS2 (4.00 g), G1 (2.00 g) and G2 (4.00 g). The control diet had 0% inclusion of both roselle and ginger. Other feed ingredients used in the diet formulation includes fish meal (FM) soy bean meal (SBM), wheat offal, yellow maize (YM), vitamin/mineral premix, cassava starch, palm oil, bone meal and salt (Table 1). Pearson square method was used in feed formulation. Samples of the experimental diets were sent to the laboratory for proximate analysis. Samples were analysed following the procedure of A.O.A.C (2000).

Experimental design

A total of one hundred and fifty fish (150) (initial weight 35.41±1.53 g) were randomly distributed to 15 aquarium plastic tanks (1 m x 1

Table 1. Percentage composition (g/100 g) of experimental diet.

Ingredient	Control	HS1	HS2	G1	G2
Fish meal	26.09	26.09	26.09	26.09	26.09
Soybean meal	21.74	21.74	21.74	21.74	21.74
Yellow maize	21.74	19.74	17.74	19.74	17.74
Wheat offal	21.74	21.74	21.74	21.74	21.74
Palm oil	1.96	1.96	1.96	1.96	1.96
Vit/min premix	1.52	1.52	1.52	1.52	1.52
Cassava starch	1.74	1.74	1.74	1.74	1.74
Salt	1.52	1.52	1.52	1.52	1.52
Bone meal	1.96	1.96	1.96	1.96	1.96
Roselle	0.00	2.00	4.00	0.00	0.00
Ginger	0.00	0.00	0.00	2.00	4.00
Total	100	100	100	100	100
Calculated crude protein (%)	37.00	37.00	37.00	37.00	37.00

m x 1 m) at ten fish per treatment. Each tank contained 30 L of water. Experimental diets were randomly assigned using completely randomized design to triplicate tanks. The fish were fed 5% body weight per day in two portions (by 9.00 am and 5.00 pm) for 8 weeks. Quantity of feed was adjusted forth nightly after batch-weighing of experimental fish. Water in the tanks was partly removed by siphoning and replaced with fresh water every three days to avoid fouling resulting from faeces and uneaten food. Water quality parameters such as temperature, dissolved oxygen and pH of the experimental water tanks were monitored and measured daily using water testing kits (PRO-LAB^{TM} Flourida). Temperature, pH and dissolved oxygen were maintained at 27.12±0.04°C, 6.98±0.01 and 6.05±0.21 mg L^{-1}, respectively.

Growth parameters

Weight measurement of the fish was obtained at the end of the experiment and the following growth parameters were determined according to the formula of Iheanacho et al. (2018): Mean weight gain (SWG); Specific growth rate (SGR); Food conversion ratio (FCR);

Mean weight gain (SWG) = final weight - initial weight

Specific growth rate (SGR)

$$SGR = \frac{(\text{Ln mean final weight} - \text{Ln mean initial weight}) \times 100}{\text{Time (days)}}$$

Ln = Natural logarithm

Food conversion ratio (FCR)

$$FCR = \frac{\text{Weight of food fed (Dry gram weight)}}{\text{Weight gain of fish (Wet gram weight)}}$$

Condition factor = K = $100W/Lb^3$ (Gomiero and Braga, 2005)

Where, K = condition factor; W = the weight of the fish in gram (g); L = the total length of the fish in centimetre (cm); b = the value obtained from the length-weight equation.

Haematological analysis

Three fish per tank were sampled for blood collection at the end of the experiment. Blood was collected from the caudal vein into an EDTA lithium tubes. Blood samples were immediately transported to the haematology laboratory unit of the Federal University, Ndufu Alike Ikwo Medical Centre for Haematological Analysis. The blood was analyzed to determine the packed cell value (PCV) with micro haematocrit using heparinised capillary tube (25 mm), while red blood cell (RBC), white blood cell (WBC) counts, haemoglobin (Hb) concentration and red cell indices (mean corpular volume (MCV), mean corpular hemoglobin (MCH) and mean corpular haemoglobin concentration MCHC) were determined as described by Dacie and Lewis (2011).

Statistical analysis

Data obtained from the experiment were subjected to one-way analysis of variance (ANOVA), using Statistical Package for Social Science (SPSS 2006, version 22). Duncan multiple range test (DMRT) was used to compare the differences between means at $p<0.05$. Data were presented as mean ± SE.

RESULTS

Results on proximate composition of the experimental diets are presented in Table 2. Highest percentages of crude protein (30.38±0.06), ash (8.21±0.02), dry matter (93.28±0.02) and fat (4.24±0.01) were seen in ginger (G2) based diet. Growth performance of *C. gariepinus* fed roselle and ginger supplemented diets are presented in Table 3. Significant difference ($p<0.05$) was observed among treated groups AS compared to the control.

Data on haematological responses of *C. gariepinus* fed roselle and ginger supplemented diets are presented in Table 4. Significant differences ($p<0.05$) were seen among treated groups as compared to the control.

Table 2. Proximate composition (%) of experimental diets.

Parameter	Control (0.00)	HS1(2.0 g)	HS2(4.0 g)	G1(2.0 g)	G2(4.0 g)
Dry matter	92.66±0.03^{e}	92.80±0.02^{d}	92.97±0.03^{c}	93.14±0.02^{b}	93.28±0.02^{a}
Crude protein	25.74±0.03^{e}	28.83±0.02^{d}	29.33±0.03^{c}	29.59±0.06^{b}	30.38±0.06^{a}
Ash	7.82±0.02^{e}	7.88±0.02^{d}	7.96±0.01^{c}	8.09±0.02^{b}	8.21±0.02^{a}
Crude fibre	2.35±0.01^{d}	2.56±0.01^{b}	2.52±0.01c	2.63±0.01^{a}	2.54±0.01^{bc}
Moisture	7.34±0.03^{a}	7.20±0.02^{b}	7.03±0.03^{c}	6.86±0.02^{d}	6.72±0.02^{e}
Fat	3.88±0.01^{e}	3.97±0.01^{d}	4.05±0.01^{c}	4.17±0.01^{b}	4.24±0.01^{a}
*NFE	52.88±0.01^{a}	49.58±0.01^{b}	49.12±0.01^{c}	48.67±0.04^{d}	47.92±0.05^{e}

* Nitrogen free extract (NFE) = (100 - (crude protein + ash+ crude fibre + moisture + fat)). Data presented represents mean values of parameters and values in the same rows with the same alphabet superscript are not significantly different (p>0.05).

Table 3. Growth response of *C. gariepinus* juvenile fed Roselle and ginger supplemented diets.

Parameter	Control (0.0 g)	HS1 (2.0 g)	HS29 (4.0 g)	G1 (2.0 g)	G2 (4.0 g)
Initial weight (g)	35.46±0.50^{a}	35.22±0.42^{a}	35.08±0.52^{a}	35.28±0.22^{a}	35.68±0.19^{a}
Final weight (g)	62.50±1.53^{ab}	53.03±1.24^{b}	63.95±2.54^{ab}	56.82±2.41^{ab}	67.51±0.44^{a}
Weight gain (g)	27.04±1.45^{ab}	17.82±1.58^{b}	28.88±6.07^{ab}	21.55±2.20^{ab}	31.82±0.63^{a}
SGR (%g/d)	2.74±0.01^{b}	2.73±0.01^{b}	2.73±0.01^{b}	2.73±0.01^{b}	3.13±0.21^{a}
FCR	1.02±0.20^{b}	1.24±0.15^{a}	1.18±1.25^{a}	1.12±0.34^{ab}	0.99±0.51^{b}
Condition factor (k)	0.61±0.08^{a}	0.39±0.05^{a}	0.45±0.02^{a}	0.43±0.00^{a}	0.60±0.11^{a}

Data represents mean ± (SE) values of parameters estimated. Values in the same rows with the same alphabet superscript are not significantly different (p>0.05).

Table 4. Hematological data of *C. gariepinus* fed Roselle and ginger supplemented diets.

Parameter	Control (0.0 g)	HS1 (2.0 g)	HS2(4.0g)	G1 (2.0 g)	G2 (4.0 g)
PCV (%)	30.33±2.73^{ab}	26.00±3.22^{ab}	25.67±0.88^{b}	28.00±3.46^{ab}	34.33±0.88^{a}
Hb (g.dL^{-1})	9.93±0.92^{a}	8.57±1.23^{a}	8.31±0.99^{a}	9.15±1.07^{a}	11.61±0.57^{a}
RBC(10^{12} L)	3.63±0.22^{ab}	3.08±0.43^{b}	3.27±0.09^{ab}	3.29±0.35^{ab}	4.07±0.08^{a}
WBC(10^{9} L)	4.30±0.27^{b}	6.50±0.38^{a}	5.99±0.26^{a}	6.11±0.22^{a}	6.47±0.35^{a}
MCV (fL)	83.17±2.67^{a}	85.03±2.26^{a}	78.55±0.57^{a}	84.69±1.57^{a}	83.29±2.52^{a}
MCH (pg)	27.23±1.04^{a}	27.82±0.09^{a}	25.31±2.38^{a}	27.72±0.31^{a}	27.34±1.29^{a}
MCHC (%)	32.74±0.52^{a}	32.77±0.92^{a}	32.18±2.83^{a}	32.74±0.24^{a}	32.79±0.55^{a}

PCV, Packed cell value; Hb, haemoglobin; RBC, red blood cell; WBC, while blood cell; MCV, mean corpular volume; MCH, mean corpular hemoglobin; MCHC, mean corpular haemoglobin concentration.*Data represents mean ± (SE) values of parameters estimated. Values in the same rows with the same alphabet superscript are not significantly different (p>0.05).

DISCUSSION

Proximate examination of roselle and ginger based diets revealed significant difference (p<0.05) in proximate composition (Table 2). Higher values for crude protein, ash, dry matter and fat were found in ginger based diets followed by roselle based diets and the control. Kumar et al. (2014) reported 41.14 to 42.32 4% crude protein for ginger based diets.

Medicinal plants have been reported to be growth promoters and immune boosters in livestock and fish nutrition (Levic et al., 2008; Kumar et al., 2014; Reverter et al., 2014; Iheanacho et al., 2017a). The findings of the present study on growth performance of *C. gariepinus* juvenile fed roselle and ginger supplemented diets revealed that there were increases in treated groups as compared to the control, although, insignificant (p>0.05) (Table 3). Highest values in terms of final weight (67.51±0.44) and weight gain (31.82±0.63) were observed in fish group fed 4.0 g ginger supplemented diet followed by 4.0 g roselle treated groups. Significant increase in terms of SGR was seen in 4.0 g ginger treated group as compared to the control. Food conversion ratio (FCR) was observed to be highest in fish

group fed 2.0 g roselle supplemented diet and lowest in 4.0 g ginger treated groups. Kumar et al. (2014) reported enhanced growth in Indian catfish fingerling (*Mystus montanus*) fed ginger supplemented diets when compared with the control diet. Iheanacho et al. (2017a) reported significant increases in weight gain, specific growth rate and final weight when *C. gariepinus* juvenile were exposed to varying concentrations (0.25, 0.50, 0.75 and 1.0 g/35 mgL) of ginger as compared to the control. Adewole (2014) reported significant increases ($p<0.05$) in growth parameters (final weight, weight gain, specific rate and relative growth rate) in *C. gariepinus* fed roselle supplemented diets when compared with the control. The positive response to growth in treated fish especially at 4.0 g inclusion level of ginger could be attributed to the high proximate content of ginger (Table 2). Ginger is a good source of protein, mineral elements, vitamins and contains good number of phytochemical constituent that enhance growth and health of animals.

Hematological assays might give a useful guide on the physiological condition of fish (Haghighi and Rohani, 2013). The present study revealed significant changes in hematological parameters in the study fish. Highest values for PCV, Hb and RBC were seen in fish fed 4.0 g ginger supplemented diet as compared to other treatments and the control. There were no significant changes ($p>0.05$) in MCV, MCH and MCHC among treated groups as compared to the control. The present study revealed that ginger and roselle enhanced non specific immune response in *C. gariepinus*. The result of the present study collaborates the findings of Haghighi and Rohani (2013) who reported significant increases in RBC, PCV, Hb and WBC values in rainbow trout (*Oncorhynchus mykiss*) fed ginger supplemented diets when compared to the control. Kumar et al. (2014) also reported significant increases ($p<0.05$) in RBC, Hb, PCV and WBC values in Indian catfish (*Mystus montanus*) fed ginger based diets as compared to the control. Iheanacho et al. (2017a) reported similar findings when *C. gariepinus* were exposed to varying concentrations of ginger bath for 12 weeks.

Yahaya et al. (2012) opined that medicinal plants (roselle, ginger, ugwu and moringa) significantly increased haematological parameters in albino rat as compared to the control.

Conclusion

The present study revealed that both roselle and ginger supported the growth and hematology of *C. gariepinus* juvenile especially, 4.0 g ginger supplemented diets. The use of costly feed supplements should be discouraged. Since ginger and roselle are readily available all year round, there is need to encourage the local fish farmers to imbibe this feed biotechnology in order to minimize cost of fish production.

CONFLICT OF INTERESTS

The authors declare that there is no conflict of interest.

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REFERENCES

Abu-Tarboush HM, Ahmed SAB, Alkahtani HA (1997). Some nutritional properties of karkade (*Hibiscus sabdariffa*) seed products. Cereal Chem. 74:352-355

Adewole AM (2014). Effects of roselle as dietary additive on growth and production economy of *Clarias gariepinus*. J. Emerg. Trends Eng. Appl. Sci. 5(7):1-8.jeteas.scholarlinkresearch.com

Alector VA, Osho IB (2009). Formulation of medicinal plants for animal production. invited paper II. Proceedings of the Humboldt Kellog/5th Annual Agric. Conference on formulation of medicinal plants for crop and animal production in Nigeria: Realities and Challenges. 21st – 23rd April, 2009. Federal University of Technology Akure. pp. 7-18.

Dacie JV, Lewis SM (2011). Practical Hematology. 11th edition, New York: ChurchillLivingstone,

Estrella MCP, Mantaring JBV, David GZ (2000). A double blind randomized, controlled trial on the use of malunggay (Moringa oleifera) for augmentation of the volume of the breast milk among nonnursing mothers of preterm infants. Philippine J. Ped. 49:306.

Gomiero LM, Braga FMS (2005). The condition factor of fishes from two river basins in Sao Paulo state, Southeast of Brazil. Acta Sci. 27:73-78.

Haghighi M, Mostafa SR (2013). The effects of powdered ginger (*Zingiber officinale)* on the haematological and immunological parameters of rainbow trout *Oncorhynchus mykiss.* J. Med. Plant Herb. Ther. Res. 1:8-12.

Iheanacho S, Ogunji JO, Ogueji EO, Nwuba LA, Nnatuanya IO, Ochang SN, Mbah CE, Ibrahim BU, Haruna M (2017a). Comparative Assessment of Ampicillin Antibiotic and Ginger (*Zingiber officinale)* effects on Growth, Haematology and Biochemical Enzymes Of *Clarias gariepinus* Juvenile. J. Pharmacog. Phytochem. 6(3):761-767.

Iheanacho SC, Nworu SA, Ogueji EO, Nnatuanya I, Mbah CE, Anosike F, Okoye C, Ibrahim UB, Kogi E, Haruna M (2017b). Comparative assessment of proximate content and Organoleptic quality of African catfish (*Clarias gariepinus*) processed by smoking and solar drying methods. Afr. J. Agric. Res. 12(3):2824-2829.

Iheanacho S, Ikwo T, Igweze N, Chukwuidha C, Ogueji EO, Onyeneke R (2018). Effect of different dietary inclusion levels of melon seed (*Citrullus lanatus*) peel on growth, Haematology and Histology of *Oroechromis niloticus* Juvenile. Turkish J. Fish. Aquat. Sci. 18 (3):377-384.

Ijeomah AU, Ugwuona FU, Abdullah H (2012). Phytochemical composition and antioxidant properties of *Hibiscus sabdariffa* and *Moringa oleifera*, Nigerian J. Agric. Environ. 8(1):10-16.

Kim JS, Park HW, Yang JH, Shin TY, KimYC, Baek N, Kim SH (2008). "Cytotoxic components from the dried rhizomes of *Zingiber officinale* Roscoe". Arch. Pharmacol. Res. 31(4):415-418.

Kumar IV, Chettadurai G, Veri T, Peeran SH, Mohanraj J, (2014). Medicinal plants AS immunostimulants for health management in Indian Catfish. J. Coast. Life Med. 2(6):426-430, Doi: 10.12908IJCCM.2.20141432

Levic JG, Sinisia MG, Djuragic O, Slavica S (2008). Herbs, organic acids as alternative to antibioticgrowth promoter. Arch. Zootechn. 11:15-1

Okeke IN, Sosa A (2003). Antibotic resistance in Africa – discerning the

enemy as plotting a defence. Features Afr. Health 25(3):10-15.
Pietta PG (2000). Flavonoids as antioxidants. J. Nat. Prod. 63(7):1035-1042.
Reverter M, Bontemps N, Lecchini D, Banaigs B, Sasal P (2014). Use of plant extracts in fish aquaculture as an alternative to chemotherapy: Current statue and future perspectives. Aquaculture 433:50-61.
Rico A, Phu TM, Satapornvanit K, Min J, Shahabuddin AM, Henrik PJG, Murray FJ, Little DC, Dalsgaard A, Van den Brink PJ (2013). Use of veterinary medicines, feed additives and probiotics in four major internationally traded aquaculture species farmed in Asia. Aquaculture 412:231-243.
Romero J, Carmen GF, Paola N (2012). Antibiotics in Aquaculture – Use, Abuse and Alternatives. In Health and environment in aquaculture. InTech. In: Tech, Available from: http://www.intechopen.com/books/health-and-environment-in-aquaculture/antibioticsin-aquaculture-use-abuse-and-alternatives.
Shubha RS (2015). Medicinal uses of ginger (*Zingiber officinale* Roscoe) improves growth and enhances immunity in aquaculture. Int. J. Chem. Stud. 3(2):83-87.
US Food and Drug Administration (USDA) (2014). Code of Federal Regulations,; Essential oilsoleoresins (solvent-free), and natural extractives (including distillates): ''Substance generally Recognised As Safe'' Title 21, part 182, sec. 182.20.
Visek WJ (1978). The mode of growth promotion by antibiotics. J. Anim. Sci. 46:1447-1469.
World Bank (2006). Aquaculture: Changing the Face of the Waters. Meeting the Promies and Challenges of Sustainable Aquaculture. The World Bank, Wsahington.
Yahaya T, Okpuzor J, Ajayi T (2012). The prophylactic efficacy of Roselle (*H. Sabdariffa),* Moringa (*Moringa oleifera),* Ginger (*Zingiber officinale)* and Ugwu (*T. Occidentalis)* on the hematology and serum protein of albino rats (*Rattus norvegicus)* exposed to cement dust. J. Med. Plant 6:189-196.