Full Length Research Paper

Characterization of diploid and triploid *Heterobranchus bidorsalis* using morphometric, meristic and haematological parameters

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The study investigates comparative changes in morphometric, meristic and haematological values of triploid and diploid strain of *Heterobranchus bidorsalis* with a view to establishing differences and comparative adaptability between the two strains. The experiment was carried out inside net hapas submerged inside $1 \times 1 \times 1.2 \text{ m}^2$ concrete tank where diploid (2n) and triploid (3n) fish were reared. Each hapa net contain 45 post fingerlings of the same genetic makeup. 10 post fingerlings of diploid and triploid strains of *H. bidorsalis* with average total length between 11.2 and 12.8 cm and 12.2 and 14 cm respectively were collected for morphometric and meristic parameters. Blood samples were also collected and analyzed packed cell volume (PCV), haemoglobin (Hb), white blood cell count (WBC), red blood cell count (RBC), mean cell hemoglobin concentration (MCHC), platelet, and mean cell volume (MCV) values of triploid and diploid fish were analyzed. This study shows the superiority of the triploid *H. bidorsalis* over the diploid strain. It also indicated that morphometric and meristic indices are the best parameters to characterize post juvenile diploid and triploid *H. bidorsalis*.

Key words: Heterobranchus bidorsalis, diploid, triploid, morphometric, meristic, heamatology,

INTRODUCTION

Recent trends all over the world, point to a decline in landing from capture fisheries, which is an indicator that fish stocks have approached or even exceeded the point of maximum sustainable yield; aquaculture therefore remains the only viable alternative for increasing fish production in order to meet the protein need of the people (Adewumi and Olaleye, 2011). Bakir et al. (1993) reported that *Heterobranchus* sp., which belong to the family Clariidae, is one of the species of freshwater fish that are mostly utilized in aquaculture, especially in the developing world like Nigeria. Tawari and Abowei (2011) reported that *Heterobranchus* sp. is one of the fish species that its fingerlings are available both in the wild and cultured medium in Nigeria. The use of sexually

sterile fish in fish production is advantageous for several applications, such as: controlling reproduction of exotic species; preventing potential backcross of hybrids with either parent species resulting in intermingling of genetic material; improving growth of aquaculture species because less energy is diverted for reproduction (Rottmann et al., 1991). Gain and loss of whole chromosomes leads to aneuploidy in which the chromosome number differs from the normal haploid (n) or diploid (2n) chromosome number, it changes to an exact multiple of the haploid number (e.g. 3n and 4n) which is termed triploidy tetraploidy, vbiolgylog and respectively (Strunjak-Perovic et al., 2003). Although there are many similarities between triploid and diploid fish, basic differences exist and conflicting results in terms of performance have been obtained in salmonids and other species (O'Flynn et al., 1997). The use of haematological values as indices of the state of animal health is receiving a lot of research effort (Awe et al., 2011). Blood

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examination is a good way of assessing the health status of an animal as it plays a vital role in the physiological, nutritional and pathological status of the animal (George et al., 1994). Hematological assessment in reared and wild fish is an important tool to evaluate fish health. They can be induced by the presence of pollutants and factors such as temperature, salinity, pH, dissolved oxygen concentration, carbon dioxide and inadequate management (Ranzani-Paiva and Silva-Souza, 2004). Haematological status of diploid, triploid and tetraploid fish could provide a better understanding of the comparative adaptability of these fish (Zexia et al., 2007). The aim of this work was to characterize diploid and triploid H. bidorsalis using morphometric, meristic and heamatological parameters so as to determine the changes that occurred in the normal diploid H. bidorsalis from the triploid H. bidorsalis.

MATERIALS AND METHODS

Experimental design

The design of the experiment was a completely randomized design (CRD). Two different strains of *H. bidorsalis* {the diploid (2n) and the triploid (3n)} of equal age (nine weeks each) were reared in net hapa submerged inside $1 \times 1 \times 1.2 \text{ m}^2$ concrete tank. Each hapa net contained 45 post fingerlings of the same genetic makeup, 10 post fingerlings each were randomly selected from the net hapa containing the diploid strain, and the net hapa containing triploid strains for morphometric, meristic and heamatological parameters examination.

Determination of morphometric, meristic and heamatological parameters

In the laboratory, morphometric parameters including; head width, frontal frontanelle (long), frontal frontanelle (small) caudal peduncle length, head to dorsal fin origin, distance between the eye, gap between dorsal fin and adipose fin, sex, body depth at anus, adipose fin length and fish weight were measured while the meristic parameters observed include; number of dorsal fin rays, number of pectoral fin rays, number of pelvic fin rays, number of anal fin rays, vomerine tooth plate width, vomerine tooth plate depth, number of left gill rakers, number of vertebrae (atlas), number of vertebrae (urostyle), number of vertebrae column, premaxillary tooth width, premaxillary tooth depth, nasal barbell length, mandibular barbell length and number of right gill rakers. These parameters were recorded for both 2n and 3n H. bidorsalis; the total length of the diploid fish ranged from 11.2 to 12.8 cm while that of the triploid ranged from 12.2 to 14.0 cm. Thereafter, blood was collected from the caudal peduncle of each fish using separate heparinized syringes and oxalate anticoagulant was added to prevent the blood from coagulating. Standard haematological procedure described by Blaxhall and Daisley (1973) were employed in the assessment of the various blood parameters. Haemoglobin (Hb) was done by the cyanomethaemoglobin method, packed cell volume (PCV) by microhaematocrit method, white blood cell count (WBC) was determined with the improved neubauor counter, different count was done on blood film stained with May Grumwald Gemsa Stain, red blood cell count (RBC) was estimated using the relationship between Hb and PCV (Mirale, 1982). Mean cell hemoglobin concentration (MCHC), and mean cell volume (MCV) were

calculated using the formulae mentioned by Dacie and Lewis (2001):

MCHC (%) = Hb / Hct \times 100

MCV (μ^3) = Hct / RBC × 10

Statistical analyses

Heamatological data collected from the experiment were subjected to one way analysis of variance (ANOVA) test using the Statistical Package for the Social Sciences (SPSS 13.0) for window software. Where significant differences occurred, group means were further compared with Duncan's multiple range test (SPSS, IL, USA).

RESULTS AND DISCUSSION

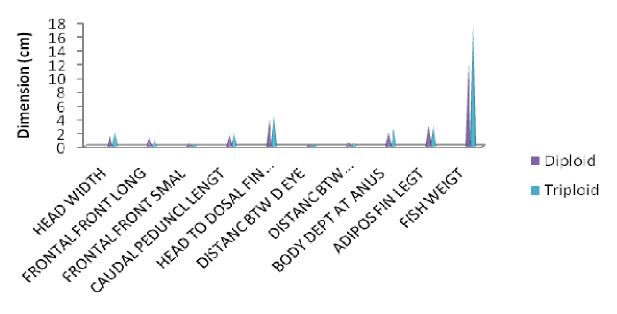
The mean value of the morphometric parameters (Figure 1) shows the superiority of the triploid fish over the diploid strain as the triploid have better weight, longer adipose fin length, better flesh at body depth towards the anus; triploid fish also have longer length between the head and dorsal fin origin and longer caudal peduncle length, at the head region; triploid fish have wider head width than the diploid strain while the diploid fish have longer frontal frontanelle for both small and long frontanelle. This agrees with the report of Tiwary et al. (1997) that triploid *Heteropneustes fossilis* showed better growth rates than normal diploid individuals under controlled laboratory conditions.

The meristic parameters (Figure 2) indicate that alteration in genetic makeup does not alter the vomerine tooth plate depth (cm) of triploid *H. bidorsalis*. Both diploid and triploid *H. bidorsalis* strain also have the same number of left gill raker; the same number of vertebrae (both atlas and urostyle) and the same number of right gill. However, the two strains have different numbers of anal fin rays, vomerine tooth plate width (cm), number of vertebrae column, premaxillary tooth width (cm), premaxillary tooth dept (cm), nasal barbell length (cm) and mandibular barbell length (cm) which can be used to differentiate them.

Study on the haemtological indices indicated that there is no significant difference in all the indices of diploid and triploid *H. bidorsalis* except MCHC (Table 1). This result disagrees with other researchers like Zexia et al. (2007) who used six month old fish for the haematological characterization of loach *Misgurnus anguillicaudatus* diploid, triploid and tetraploid specimens suggesting that haematological indices of diploid and triploid *H. bidorsalis* strain will not be significantly different at the early stage of life (at post fingerlings stage) except for MCHC.

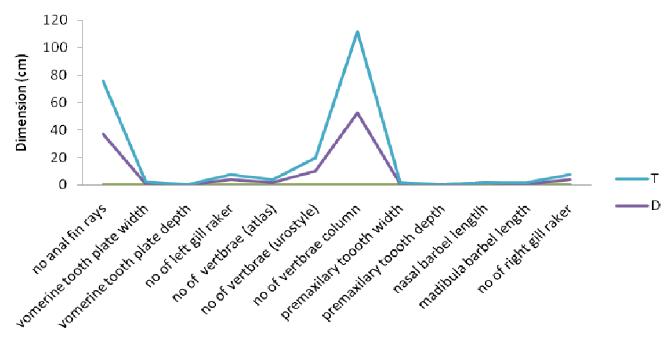
Conclusion

This study shows the superiority of the triploid *H. bidorsalis* over the diploid strain; it also indicates that a



Morphometric Parameters

Figure 1. Morphormetric parameters for diploid and triploid H. bidorsalis



Meristic Parameters



Table 1. Haematological parameters of triploid and diploid H. bidorsali.

Treatment	PCV	Hb (g/L)	RBC (×106 µL ⁻¹)	WBC (×103 µL ⁻¹)	Platelet (g/dL)	MCV (fL)	MCHC(g/dL)
D	27.000 ^a	8.900 ^a	2.4880 ^a	19160.0 ^a	100000 ^a	111.200 ^a	33.2000 ^a
Т	33.600 ^a	10.620 ^a	3.1680 ^ª	19020.0 ^a	94600 ^a	106.800 ^a	31.6000 ^b

Mean with different superscripts along the same row indicate significance difference at 95% confidence value. D, Diploid; T, triploid H. bidorsalis.

wide variation existed between morphometric and meristic indices of post juvenile diploid and triploid strains of *H. bidorsalis*. The haematological indices of the two strains did not show a significant difference. Since growth performance is a critical factor in aquaculture, it is therefore suggested that triploid *H. bidorsalis* should be cultured by farmer rather than culturing the diploid strain.

REFERENCES

- Adewumi AA, Olaleye VF (2011). Catfish culture in Nigeria: Progress, prospects and problems. Afr. J. Agric. Res. 6(6): 1281-1285.
- Awe S, Sani A, Tunde OE (2011). Haematological studies of rats fed with some selected locally produced fruit wines. Bioresearch Bull. 4: 217-222.
- Bakir HM, Melton SL, Wilson JL (1993). Fatty acid composition, lipids and sensory characteristics of white amur (*Ctenopharyngodon idella*) fed different diets. J. Food Sci. 58(1): 90-95.
- Blaxhall PC, Daisley KW (1973). Routine haematological methods for use with fish blood. J. Fish Biol. 5: 771-781.
- Dacie JV, Lewis SM (2001). Practical Haematology, 9th edition. Churchill Livingstone, London. p. 633.
- George HB, Donald ES, Colin RP (1994). Physiol. Biochem. 16: 156-258.
- Mirale JB (1982). Laboratory medicine haematology. 6th edition. The CV Mosby Pub. London. p. 883.

- O'Flynn FM, McGeachy SA, Friars GW, Benfey TJ, Bailey JK (1997). Comparisons of cultured triploid and diploid Atlantic salmon (*Salmo salar* L.). ICES J. Mar. Sci. 54: 1160-1165.
- Ranzani-Paiva MJT, Silva-Souza AT (2004). Hematology of Brazilian fish. In: Ranzani-Paiva MJT, Takemoto RM, Lizama M, de los AP (Ed.). Sanity of the aquatic organisms. São Paulo: Varela. pp. 89-120.
- Rottmann RV, Shireman JV, Chapman FA (1991). Induction and verification of triploidy in fish. SRAC Publication. 427: 2.
- Strunjak-Perovic I, Coz-Rakovac R, Topic PN (2003). Micronucleus occurrence in diploid and triploid rainbow trout (Oncorhynchus mykiss Walbaum). Vet. Med. Czech. 48(8): 215-219.
- Tawari CC, Abowei JFN (2011). An exposition of the potentials and utilization of sustainable culture fisheries in Africa. J. Appl. Sci. Eng. Tech. 3(4): 304-317.
- Tiwary BK, Kirubagaran R, Ray AK (1997). Induction of tripliody by cold shock in Indian catfish, Heteropneustes fossilis (Bloch). Asian Fish Sci. 10: 123-129.
- Zexia G, Weimin W, Khalid A, Xiaoyun Z, Yi Yang, James S, Diana HW, Huanling W, Yang L, Yuhua S (2007). Haematological characterization of loach *Misgurnus anguillicaudatus*: Comparison among diploid, triploid and tetraploid specimens. Comparative Biochem. Physiol. 147: 1001-1008.