



## Suspected Case of Rhabdomyosarcoma in a Cultured *Clarias gariepinus*

Okorie-Kanu, C. O.<sup>1</sup>; Agbakwuru, I. O.<sup>2</sup>. and Nwagbara, N. D<sup>1</sup>

<sup>1</sup> Department of Veterinary Pathology, Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria.

<sup>2</sup> Department of Veterinary Anatomy, Michael Okpara University of Agriculture, Umudike, Abia state, Nigeria.

\*Corresponding autho rukdrcokoriekanu@yahoo.co.uk; Tel No:+2348038993506

### INTRODUCTION

The over 30,000 species of fish so far reported in literature (Helfman *et al.*, 2009) presumes an immense opportunity for discovery of numerous disorders, especially spontaneous neoplasia. However, information on neoplastic diseases is still poor because of the large size of the environment (rivers, ocean etc) the animals live, difficulty in tracking the individual fishes and the effects of autolysis and predation on vast majority of samples which limit observation of aquatic disorders to only zoo and aquarium populations, tropical and marine aquaria and cultured fish (Lombardini *et al.*, 2014).

Because of the economic value of fishes, neoplastic diseases of aquatic animals are reported in fishes more than amphibians and reptiles as many of the tumors are observed during marketing and processing for consumption (Schlumberger and Lucke, 1948). Also, majority of such cases still unaccounted for as such cases are easily separated from the ones meant for the market and thrown away. Furthermore, the apparent low occurrence of rhabdomyosarcoma may not be unconnected to the tumor's low degree of differentiation which results in misdiagnosis in most cases (Vas *et al.*, 1993).

Reports on neoplasms of fish include those of the urinary track (Nakatsuru *et al.*, 2000; Lombardini *et al.*, 2010, 2014), hepatic (Mikaelian *et al.*, 1998), lymphosarcoma (Bowser *et al.*, 1985; Earnest-Koons *et al.*, 1997) and rhabdomyosarcoma (Haddow and Blake, 1933; Mousavi *et al.*, 2016).

Rhabdomyosarcoma originates from striated muscle cells and sometimes from undifferentiated pluripotent cells (Kusewitt, 2012). Gross descriptions usually reveal an irregularly elevated, lobulated and relatively firm mass (Hendrick *et al.*, 1998; Cooper and Valentine, 2002; Kusewitt, 2012). Microscopically, rhabdomyosarcoma can present as disorganized tissue composed of pleomorphic cells, usually fusiform to elongated, with marked anisokaryosis and sometimes multinucleated giant cells (Kusewitt, 2012). Cross striations may be present in histologic sections and normally are hard to detect but may be demonstrated by special stains such as phosphotungstic acid and haematoxylin (Hendrick *et al.*, 1998; Cooper and Valentine, 2002). Poorly differentiated tumor cells can be demonstrated using specific antibodies against striated muscle desmin, vimentin and actin antigens in tissues (Skalli *et al.*, 1988).

## CASE REPORT

A 6-month-old cultured male *Clarias gariepinus* weighing 950g with a mass on the tail region was scooped with a fishing-net upon observation of sluggish movement in a concrete pond having about one thousand fish. The weight of the fish was relatively lower than majority of them in the pond though they were hatched at the same time. The mass was located intramuscularly towards the tail region of the fish (Figure 1) and was composed of two nodules measuring about 1 and 1.5 cm respectively with areas of necrosis at the centre (Figure 2).

Blood was collected from the caudal vein and haematological parameters determined following standard procedures. The fish was subsequently euthanized and the mass carefully extirpated and fixed in 10% buffered formal saline. The tissue sample was processed by conventional methods and sections of about 5 $\mu$ m were stained with Haematoxylin and eosin.

## RESULTS AND DISCUSSION

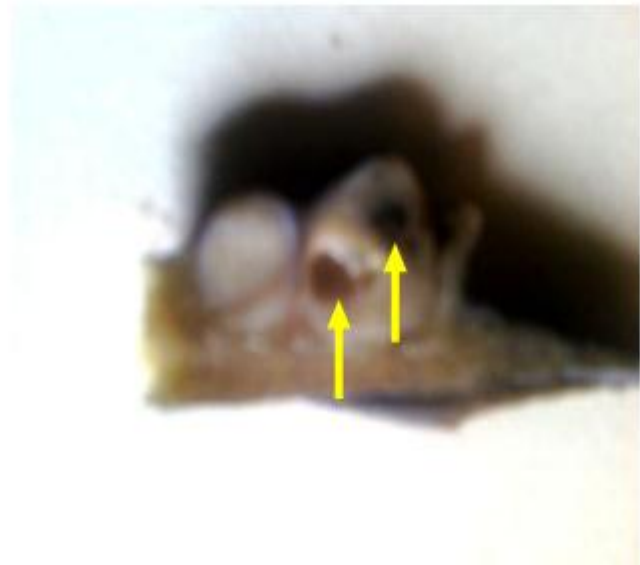


**Figure 1:** Tumor tissue at the caudal region of 6-month-old male *Clarias gariepinus*

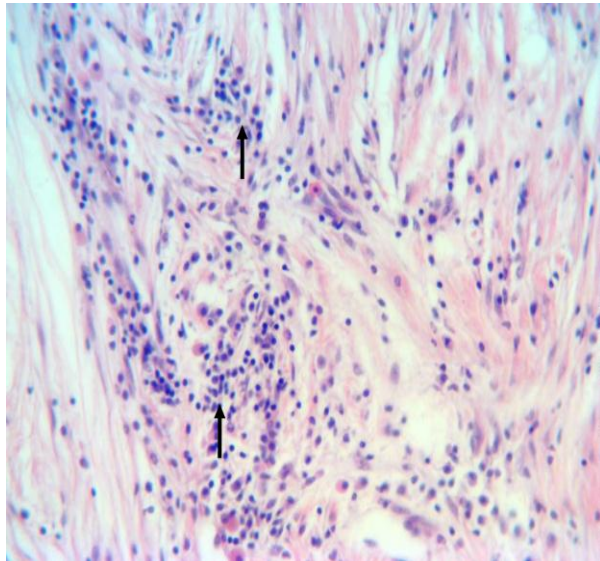
Packed cell volume – 24% (31.00 - 35.00), Haemoglobin concentration - 7.90 g/dl (10.86 - 11.90), Red blood cell count –  $1.80 \times 10^6/\mu\text{l}$  (2.37 - 2.69), Mean corpuscular volume – 133.33 fl (136.02 - 141.95), Mean corpuscular haemoglobin – 43.89pg (40.37 - 45.82), Mean corpuscular haemoglobin concentration – 32.92 g/dl (31.03 - 35.03), White blood cell count –  $18.45 \times 10^3/\mu\text{l}$  (24.50 - 32.15).

The blood picture shows mild microcytic, normochromic anaemia considering the reduced values when compared with the reference ranges (Okorie-Kanu and Unakalamba, 2015) of packed cell volume and RBC count and MCV and suggest prolonged maturation process due to red blood cells pathology occasioned by the tumor cells. The significantly reduced WBC count could have resulted from exhaustion following prolonged elaboration of the cells to the tumor tissue as was observed in figure 3.

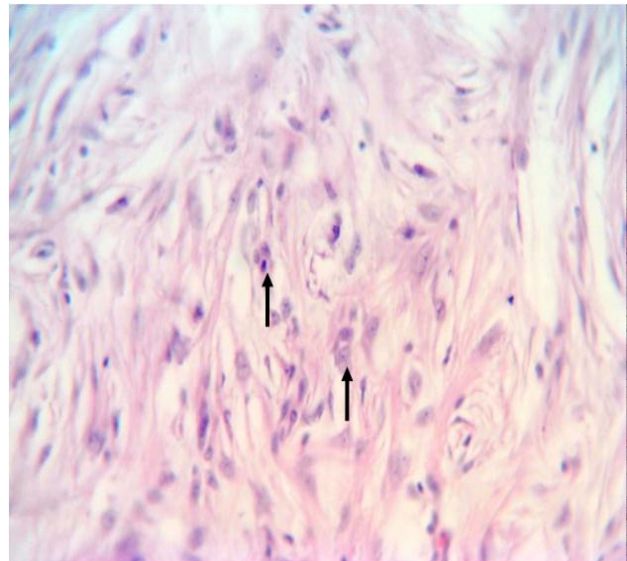
The cell morphology within the mass showed multinucleated and pleomorphic cells varying in appearance from elongated to round and strap-like cells haphazardly



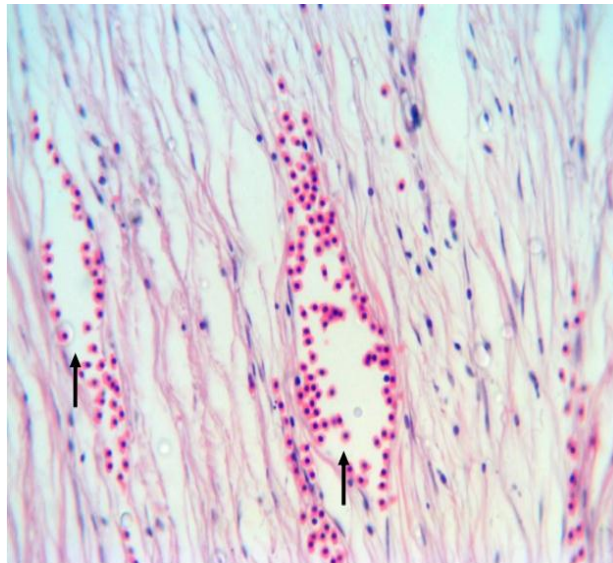
**Figure 2:** Necrotic areas on the centre of cut tumor tissue - arrows



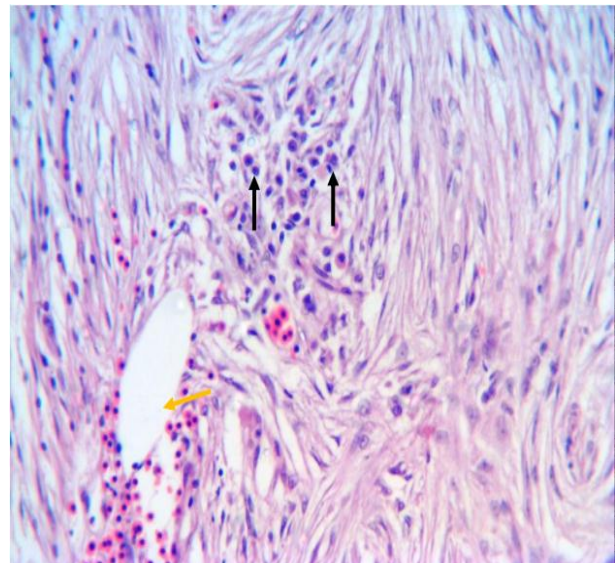
**Figure 3:** Massive infiltration of mononuclear cells and presence of mitotic cells in some segments of the tissue with striated tissue cells H&E x 400



**Figure 4:** Multinucleated-arrows and pleomorphic tumor cells haphazardly arranged H&E x 400



**Figure 5:** Well vascularized (arrows) tumor tissue with larger than normal capillaries H&E x 400



**Figure 6:** Well vascularized tumor tissue with wider than normal capillaries – yellow arrow, mitotic cells- black arrows and haphazardly arranged tissues H&E x 400

arranged (Figure 4). The tumor tissue was also well vascularized with larger than normal capillaries (Figures 5 and 6), a feature very important for growth and sustenance of tumor tissues and metastasis (Zhou *et al.*, 2011; Kusewitt, 2012; Chen *et al.*, 2015). There was also massive infiltration of mononuclear cells in some

segments of the mass (Figure 3 and 6). This was a chronic inflammatory activity as tumor cells are perceived as foreign by the host's immunological apparatus.

This case was observed in February, 2014 and since that time, all the concrete ponds in the farm had been under close monitoring for cases but none was observed before this

report. This may suggest that the tumor could be congenital and not as a result of virus or poison. This is (to the knowledge of the authors) the first case of rhabdomyosarcoma in fish in Nigeria.

## REFERENCES

- BOWSER P. R., MCCOY, C. P. and MACMILLAN, J. R. (1985). A lymphoproliferative disorder in a channel catfish, *Ictalurus punctatus* (Rafinesue). *Journal of Fish Diseases*, 8: 465 - 469.
- CHEN, L., LIN, Z. X., LIN, G. X., ZHOU, C. F., CHEN, Y. P., WANG, X. F. and ZHENG, Z. Q. (2015). Classification of microvascular patterns via cluster analysis reveals their prognostic significance in glioblastoma. *Human Pathology*, 46: 120 – 128.
- COOPER, B. J., and VALENTINE, B. A. (2002). Tumors of muscle. In: MEUTEN, D. J. (ed.) *Tumors in domestic animals*, 4th ed. Iowa State University Press, Ames, IA. pp. 319–363.
- EARNEST-KOONS, K. A., SCHACHETE, J. H. J. R. and BOWSER, P. R. (1997). Lymphosarcoma in a brook trout. *Journal of Wildlife Diseases*, 33: 666 – 669.
- HADDOW, A. and BLAKE, I. (1933). Neoplasms in fish: a report of six cases with a summary of the literature. *Journal of Pathology and Bacteriology*, 36: 41 - 47.
- HELFMAN, G., COLLETTE, B. B. and FACEY, D. H. (2009). *The Diversity of fishes: Biology, Evolution and Ecology* (2<sup>nd</sup> edition) Oxford, Wiley-Blackwell, UK pp. 3 – 16.
- HENDRICK, M. J., MAHAFFEY, E. A., MOORE, F. M., VOS, J. H. and Walte, E. J. (1998). Histological classification of mesenchymal tumors of skin and soft tissues of domestic animals, 2nd series. Armed Forces Institute of Pathology, Washington, DC. pp. 21–22.
- KUSEWITT, D. F. (2012). Neoplasia and Tumor Biology In: ZACHARY, J. F and MCGAVIN, M. D. (eds.) *Pathologic Basis of Veterinary Disease* (Fifth Edition) Mosby Inc., St Louis Missouri, USA, pp. 289-320.
- LOMBARDINI E. D., HARD, G. C. and HARSHBARGER (2014). Neoplasms of the urinary tract in fish. *Veterinary Pathology*, 51: 1000 – 1012.
- LOMBARDINI E. D., LAW M. and LEWIS, B. S. (2010). Nephroblastoma in two Siamese fighting fish (*Betta splendens*), *Fish Pathology*, 45: 137 - 139.
- MIKAELIAN, I., DE LAFONTAINE, Y., MENARD, C. (1998). Neoplastic and nonneoplastic hepatic changes in lake whitefish (*Coregonus clupeaformis*) from the St. Lawrence River, Quebec, Canada. *Environmental Health Perspective*, 106: 179 – 183.
- NAKATSURU, Y., MINAMI, K., YOSHIKAWA, A., ZHU, J. J., ODA, H., MASAHITO, P., OKAMOTO, N., NAKAMURA, Y. and ISHIKAWA, T. (2000). Ell WT 1 sequence and expression in spontaneous nephroblastomas in Japanese ell. *Gene*, 245: 245 – 251.
- OKORIE-KANU, C. O. and UNAKALAMBA, N. J. (2015). Normal haematological and blood biochemistry values of cultured *Clarias gariepinus* in Southeast, Nigeria. *Comparative Clinical Pathology*, 24: 1445-1450.
- REZAIIE, A., MOUSAVI, S. M. and ANSARI, M. B. (2016). Rhabdomyosarcoma in Silver Carp. *Journal of Aquatic Animal Health*, 28: 118-121.

- SCHLUMBERGER, H. C. and LUCKE, B. (1948). Tumors of fishes, amphibians and reptiles. *Cancer Research*, 8: 657 - 754.
- SKALLI, O., GABBIANI, G., BABAI, F., SEEMAYER, T. A., PIZZLATO, G. and SCHURCH, W. (1988). Intermediate filament proteins and actin isomers as markers for soft tissue tumor differentiation and origin, II Rhabdomyosarcomas. *American Journal of Pathology*, 130: 515 – 531.
- VAS, J. H., BORST, G. H. A., MARTINS DE LAS MULAS, J., RAMAEKERS, F. C. S., VAN MIL, F. N., MOLENBEEK, R. F., IVANYI, D. and VAN DEN INGH, T. S. G. A. M. (1993). Rhabdomyosarcomas in young pigs in a swine breeding farm: A morphologic and immunohistochemical study. *Veterinary Pathology*, 130: 271 – 279.
- ZHOU, J., LI, N., YANG, G. and ZHU, Y. (2011). Vascular patterns of brain tumors. *International Journal of Surgical Pathology*, 19: 709 – 717.