Uganda Journal of Agricultural Sciences, 2014, 15 (2): 113 - 125 ISSN 1026-0919

Common fish diseases and parasites affecting wild and farmed Tilapia and catfish in Central and Western Uganda

J. Walakira[1], P. Akoll[2], M. Engole[1], M. Sserwadda[1], M. Nkambo[1], V. Namulawa[1], G. Kityo[1], F. Musimbi[1], I. Abaho[4], H. Kasigwa[5], D. Mbabazi[1], D. Kahwa[3], I. Naigaga[3], D. Birungi[3] J. Rutaisire[6] and S. Majalija[3]

[1]National Fisheries Resources Research Institute, Uganda
[2]College of Natural Sciences, Department of Biological Science, Makerere University, Uganda
[3]College of Veterinary Medicine Animal Resources and Biosecurity, Makerere University, Uganda
[4]Bulindi Zonal Agricultural Research and Development Institute, Uganda
[5] Mbarara Zonal Agricultural Research and Development Institute, Uganda
[6]National Agricultural Research Organization, Uganda

Author for correspondence: johnwalakira2003@gmail.com

Abstract

Intensification of aquaculture production in Uganda is likely to result into disease out-breaks leading to economic losses to commercial fish farms and associated natural aquatic ecosystems. This survey assessed health profiles of selected commercial fish farms and adjacent natural aquatic ecosystemsto identify fish diseases and parasites affecting Nile tilapia (*Oreochroms niloticus*) and African catfish (*Clarias gariepinus*) in aquaculture systems in Uganda. Fish farms encounter disease out-breaks that cause low survival rates (0 - 30%), especially catfish hatcheries. Health management issues are not well understood by fish farmers, with some unable to detect diseased fish. Current control strategies to control aquatic pathogens include use of chemo-therapeutants and antibiotics. Bacterial pathogens isolated included *Flavobacterium columnare, Aeromonas* sp., *Edwardsiella* sp., *Psuedomonus* sp., *Steptococcus* sp., *Staphylococcus* sp., *Proteus* sp., and *Vibrio* sp. A high occurrence of *Flavobacterium columnare* exists in both asymptomatic and symptomatic fish was observed. Parasites included protozoans (*Ichthyopthirius multiphilis, Trichodina* sp. and *Icthyobodo* sp.) and trematodes (*Cleidodiscus* sp. and *Gyrodactylus* sp.). Diagnosis and control of diseases and parasites in aquaculture production systems requires adoption of a regional comprehensive biosecurity strategy: the East African (EAC) region unto which this study directly contributes.

Key words: African catfish, fish disease, Nile tilapia, parasite

Introduction

Fish disease out-breaks adversely affect aquaculture production (Subasinghe *et al.*, 2001; Bondad-Reantaso *et al.*, 2005), and losses are particularly high in the tropics where mitigative intervention are limited (Leung and Bates, 2013). Although aquaculture is increasing in the East African region (Rutaisire *et al.*, 2009), the risk of losing profits due to diseases and parasites is already manifesting (Akoll and Mwanja, 2012). Despite the minimal profit margins for fish farmers in Uganda (Hyuha *et al.*, 2011), aquaculture remains the great potential for reducing the national fish deficit (Dickson *et al*., 2012; Nunan, 2014).

Cases of aquatic diseases incidences leading to mortality rates of 60% have been reported in hatcheries and grow-out systems in Uganda (NaFIRRI unpublished). Infectious parasites and bacteria are reported to affect private and public fish farms with profound effects (Akoll *et al*., 2012a; Akoll *et al*., 2012b; Steigen *et al*., 2013). Consequently, concerns for risks of trans-boundary disease transmissions in the East African region cannot be ignored (Akoll and Mwanja, 2012).

Bacterial pathogens (*Flavibacterium* sp., *Pseudomonas* sp. and *Aeromonas* sp.) have been isolated from farmed fish under stressful environmental conditions (Tamale *et al*., 2010).Transmission of parasites to farmed tilapia and catfish from wild fish collected from Lake Nyabihoko in Ntungamo district was reported (NaFIRRI unpublished). Similarly, fungal infections, notably *Saprolegnia* and *Branchiomyces* remain a challenge to hatchery operators, causing significant economic losses.

Control measures practiced by fish farmers in Uganda are not very effective and well understood largely due to insufficient information that can guide researchers, policy makers and farmers to develop control or preventive strategies against potential aquatic diseases (Akoll and Mwanja 2012). The promotion of commercialisation of aquaculture in Uganda is now addressing the importance of diseases as a production risk factor that can significantly negate the marketability of aquaculture products. This paper therefore contributes to the knowledge base required in the development and adoption of regional comprehensive biosecurity strategy in the EAC.

Materials and methods

Study area and fish sample collection

Fish samples were collected from forty-four commercial fish farms located in Central and South-Western Uganda during 2012 and 2013 (Fig. 1). From each site, 20 live fish (asymptomatic and symptomatic) samples were randomly harvested from culture systems, and 5-10 live wild fish samples were collected from the natural water source (adjacent streams, rivers and lakes). Live fish were transported in 20-L buckets (with source water) to College of Veterinary Medicine Animal Resources and Biosecurity and College of Natural Sciences, Department of Biological Science laboratories at Makerere University, Uganda.

Farmer's perception

Semi-structured interviews were conducted to understand the history of disease outbreaks and management strategies applied by farmers. The content of the interview questioned included farmers' ability to detect or assess disease conditions in aquaculture farms. Farm records were examined to assess; frequency of outbreaks, number of mortalities and estimates of the number of diseased fish.

Parasitology

Fish were euthanised in clove oil (400 mg l^{-1}) following Borski and Hodson (2003) protocols, and using guidelines for Humane Euthanasia of Laboratory Animals. Parasitological examination was performed following Noga (2010) protocols. Small fish ($\leq$3 cm) were squashed between slides and examined under a light microscope for ecto-parasites, encysted and free endo-parasites. Gross pathology and

Figure 1. Study area where fish samples were collected and interviews conducted.

identification of parasites in visceral organs, including the gills of fish, was performed using a light microscope.

Bacteriology

Fish samples were aseptically dissected to collect internal tissues samples (i.e. kidney, liver and spleen) for bacteriology using the Blue Book (2010). Sterile bacteriological swabs were inoculated on brain heart infusion (BHI) media and incubated at 26 - 28 f C for 24 - 48 h. Pure cultures were obtained from colonies with identical morphology, then re-streaked on BHI media. All isolates were kept in BHI broth with 30% glycerol at “80 °C. Swabs were also done around necrotic areas of sick fish and inoculated on Hsu-shots media (with Tobramycin); which is a selective media for columnaris disease. Pure isolates were analysed morphologically, biochemically and physiologically following Blue Book (2010), and Plumb and Hanson (2011).

Histology

Samples with clinical signs of diseases were preserved in Bouin’s solutions for histo-pathological studies. Digital Imaging technique (Laurinavicius *et al*., 2012) was applied to visualise sections of liver, intestines and gills. Pathological changes in tissue sections were analysed using protocols described by Tacon (1992), Bernet *et al*. (1999), and Roberts (2012).

Water quality

From each site pH, Temperature (T), dissolved oxygen (DO), Unionised Ammonia (TAN) concentrations and Total alkalinity (TA) were measured *in situ,* using the Hach® (Loveland, CO, USA) water testing kit following Boyd and Tucker (1992) methods.

Results

Farmer's perception on diseases

In this study, 69% of fish farmers interviewed had never seen sick fish in their farms (Fig. 2a). However, 31% had observed diseased or dead fish in their production units. About 12% did not know how to identify sick fish on their culture systems (Fig. 2b). About 67% observed the health status of fish using feeding response techniques.

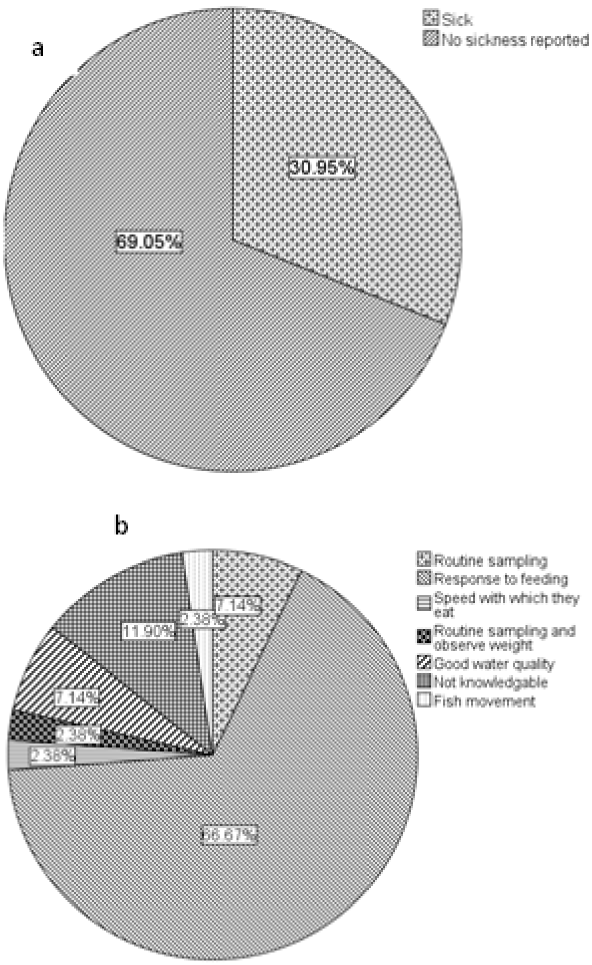

Figure 2 (a). Proportion of fish farmers who reported fish sickness on their fish farms. (b) Methods used by fish farmers to detect ill health of the fish on their fish farms.

Mortalities of up to 70% in catfish fingerling and juvenile were experienced in hatcheries. Occasional mortalities of farmed tilapia and catfish were observed in table-sized fish, when fed with poorly prepared or stored pelletised feeds. A few hatchery operators applied a combination of antibiotics (oxy-tetracycline), salt, formalin solution and potassium permanganate to reduce mortalities. In most cases farmers administered these drugs without advice from veterinary personnel or fish experts. Some farm managers accessed information related to disease treatment from internet. Estimated yields from available farm records were; (i) earthen ponds less than 2 kg/m^3, (ii) tanks less than 60 kg/m^3.

Parasitology

Digenetic trematodes

About 22% tilapia samples harvested from earthen ponds were infected with yellow cysts of digenetic trematodes (*Clinostomum* sp.) embedded beneath the scales of fish (Plate 1).

Histo-sections of infected fish revealed that metacercariae caused no pathological changes within the dermal layer.

Monogenetic trematodes

Microscope examination revealed 30% of fish samples had monogenetic trematodes and *Cleidodiscus* sp. Infections, especially on gills (Plate 2). Incidences were high in fish farms that applied organic

Table 1. Disease outbreaks observed on aquaculture farms in central and western Uganda

Period observed	N	Percent (%)
One week and still sick	2	4.8
A month ago	1	2.4
Over 4 months ago	10	23.9
Never sick	27	64.3
Not applicable (lack knowledge on fish diseases)	2	4.8

Plate 1. Arrows showing encysted *Clinostomum sp*. inside scales of table-size and fingerlings farmed tilapia.

fertilisation (e.g. animal manure) in ponds with water quality parameters ranging: pH =6.8 to 8.0; temperature = 24.3 to 25.7 °C and dissolved oxygen = 2.34 to 5.10 mg l^{-1}. Trematodes were prevalent in farmed and wild fish (tilapia and catfish) causing hyperplasia in gills (Fig. 5). Catfish were more susceptible to gyrodactylosis infections as the intensity of 1–50 parasites per fish were observed compared to tilapia with 3 parasites per fish. However, the intensity reduced with increase in size.

Protozoans

Over 90% of fish samples (Tilapia and catfish) examined had low incidences of ciliated protozoans, *Trichodina* sp. and *Icthyobodo* sp. mainly observed on fish gill filaments. Two cases of white spot disease (*Ichthyopthirius multifilis)* were found in catfish hatcheries located in the central Agricultural Ecological zone.

Bacteriology

Biochemical analysis revealed presence of gram-negative and gram-postive bacteria. The negative included *Flavobacterium columnare*, *Psuedomonus* sp., *Aeromonas* sp., *Klebseilla* sp., *Escherisia coli, Proteus* sp.; while gram positives included *Streptococcus* sp., *Staphylococcus* sp. and *Vibrio* sp. Over 70% of fish farms sampled had a high incidence of *Flavobacterium columnare, Pseudomonus* sp., *Vibrio* sp. and *Aeromonas* sp.

Samples with abnormal behavior or appearance included upright posture (Plate 4) of catfish fingerlings in tanks that

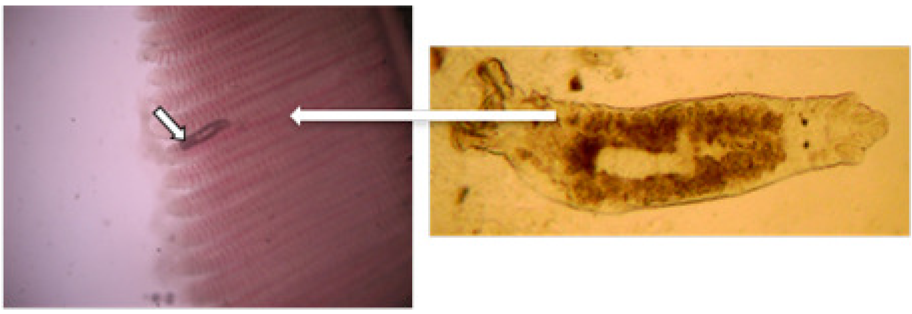

Plate 2. *Gyrodactylus* sp. attached to gill filament of *O. niloticus* gills and isolated specimen in Uganda.

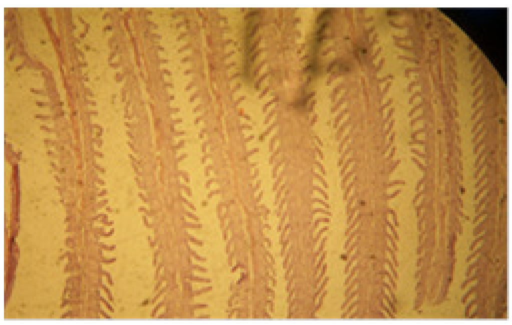

Plate 3. An intermediate of stage II and III gill hyperplasia in farmed *O. niloticus* infected with monogenetic trematodes in Uganda.

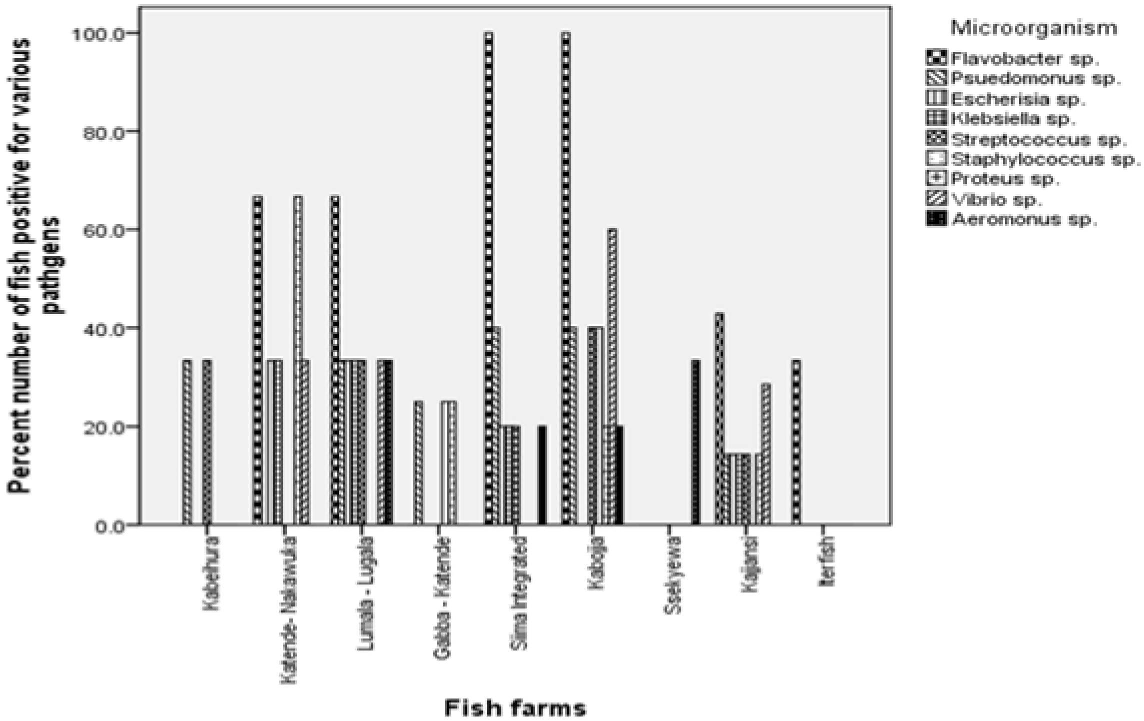

Figure 3. Isolated bacteria per given fish farm in Uganda.

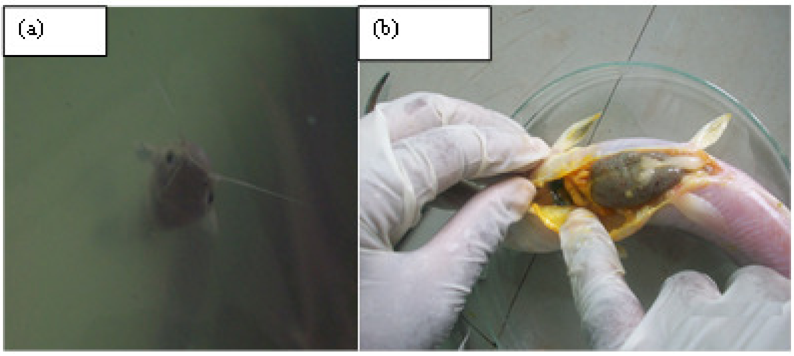

Plate 4. (a). Characteristic upright posture in catfish. (b) Laboratory examination of visceral organs.

tested positive for *Flavobacterium columnare* with enlarged internal organs. Afew catfish brood-stock from ponds had dermal petechial haemorrhages covering 70% of the body.

Histopathology

Fish samples that had systemic infections and tested with *Flavobacterium columnare* showed focal necrosis of the

Plate 5. Dermal petechial hemorrhages and inflated abdomen observed in farmed African catfish (*Clarias gariepinus*) from a brood-stock earthen pond in Uganda.

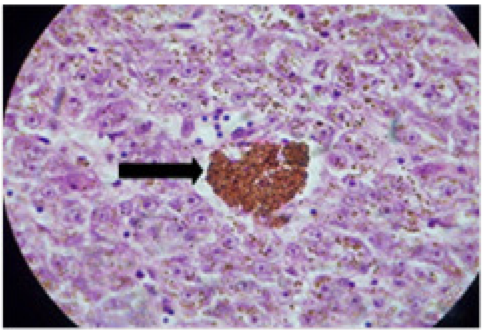

Plate 6. Cell infiltration in the liver and Cross section of the intestines of *O. niloticus*.

Table 2. Ranges of water quality parameters measured from earthen ponds and tanks

Parameter	Range (ponds)	Range (hatchery tanks)
pH	6.47 - 6.72	7.01 - 7.05
Dissolved oxygen (mg L^{-1})	2.7 - 3.8	6.5 - 8.4
Temperature (°C)	23.0 - 23.4	24.7 - 27.4
Unionised ammonia-nitrogen (mg L^{-1})	0.01- 2.00	0.01- 0.10
Total alkalinity (mg L^{-1})	61 -128	61 -128

liver with inflammatory responses (Plate 6).

Discussion

Production from fish farms in this survey is relatively low (ponds: < 2kg/m^3; tanks < 60 kg/m^3), and usually experience disease and parasite outbreaks. Majority fish farmers had no adequate knowledge on fish health management practices, and continue to incur losses. The aquaculture sector in Uganda aims at producing at least 200,000 mt annually, (MAAIF, 2010). To realise this production level there is need to build the capacity of fish farmers, extension officers and policy makers to handle disease and parasite outbreaks. Most fish farmers could not identify and manage fish diseases and parasites occurring in their farms or surrounding water environments. This study provides a basis to which an effective aquatic biosecurity program at local, national and regional levels can be developed.

Isolated fish pathogens and parasites include; gram-negative bacteria: *Flavobacterium columnare*, *Aeromonas* sp., *Edwardsiella* sp., *Psuedomonus* sp., *Vibrio* sp. and *Proteus* sp.; gram positive bacteria *Streptococcus* sp., and *Staphylococcus* sp.; ciliated protozoans (*Ichthyopthirius multifilis*, *Trichodina* sp. and *Icthyobodo* sp.) and fish flukes (*Gyrodactylus* sp. and *Clinostomum* sp.). Conversely, similar pathogens and parasites {i.e. trematodes (Digenes and monogenes), ciliated protozoans and bacteria (*Aeromonas* sp. and *Edwardsiella* sp.)} were reported to occur in farmed *O. niloticus* and *C. gariepinus* in Uganda and Kenya (Akoll *et al*., 2012; Akoll*et al*., 2012; Akoll and Mwanja, 2012). Epitheliocystis caused by novel bacteria *Cand. Actinochlamydia clariae' gen. nov.,* sp. *nov.* is reported to infect farmed catfish (*C. gariepinus*) in synergy with ciliated protozoans (*Trichodina* sp. *and Ichthyobodo* sp.)which is a potential problem to the aquaculture industry in Uganda (Steigen *et al*., 2013).

High mortalities in aquaculture are caused when parasites infect farmed fish and subsequently invaded by bacterial pathogens (Xu*et al*., 2012). Isolated bacteria *Streptococcus* sp. and *Staphylococcus* sp. are important in aquaculture research and development since they can cause human diseases (e.g. neonatal meningitis and mastitis), and their ability to resist antibiotics (Smith *et al*., 1994; Geng*et al*., 2012; Cabello *et al*., 2013). All isolated pathogens in this study have a potential to cause economic losses in aquaculture enterprises if not properly managed (Austin and Austin, 2007; Boylan, 2011; Fulde and Valentin-Weigand, 2013).

Africa has a declining per *capita* fish supply and aquaculture is reported to contribute to food security and income generation (Béné and Heck, 2005; Hishamunda and Ridler, 2006; Heck *etal*., 2007). In Africa and Uganda in particular, aquaculture has the potential to reduce the national fish deficit (Dickson *et al*., 2012; Nunan, 2014), but the occurrence of aquatic pathogens can impede its development. Commercialisation of aquaculture involves movement of aquatic materials (e.g. fish seed and brood-stocks) within and outside national boundaries but these increase the risks of introducing pathogens (Subasinghe *et al*., 1998; Subasinghe and Phillips, 2002).

It is important to note that fish pathogens were easily isolated from wild and farmed fish samples. However, the etiology was not investigatedwhich makes it difficult to infer the source of these pathogens in various aquaculture facilities. It has been documented that wild fish can transmit aquatic pathogens to farmed fish and vice versa e.g. wild fish are potential amplifiers of pathogenic *Streptococcus iniae* strains to farmed fish Zlotkin *et al*. (1998) while wild fish usually harbor pathogens that are transmitted to aquaculture establishments (Meyer, 1991). Escapees from fish farms to surrounding aquatic environments usually spread diseases, for example salmon cage and land-based farming is responsible for epizootics like furunculosis, sea lice and *Gyrodactylus salaris* that affect wild populations (Naylor *et al*., 2000; Naylor *et al*., 2005; Krkošek *et al*., 2006). Small-scale aquaculture farms growing Nile tilapia (*O. niloticus*) and/or ornamentals along the Zambezi River are reported to introduce Epizootics Ulcerative Syndrome disease to wild populations (Andrew *et al*., 2008; Huchzermeyer *et al*., 2012).

Fish farming in Uganda is largely small-scale oriented with many farms located in rural areas. Small-scale farmers are resource-poor and lack knowledge to manage fish diseases (Subasinghe and Phillips, 2002). Information generated in this study reveals how farmers lack technologies to manage aquatic diseases and parasites. For example, most farmers had relatively low dissolved oxygen levels in their ponds that stress farmed fish and make them susceptible to fish pathogens. Aquatic pathogens can spread rapidly in stressed fish when water quality conditions become poor (Boyd, 1979; Boyd, 2000). Participatory efforts to adopt biosecurity measures at all levels will improve fish production and ensure a safe aquatic environment. Georgiadis *et al.* (2001) suggested for increased cooperation among epidemiologists, fish scientists and farmers when evaluating causes and management of infectious diseases in aquaculture. Concerted efforts to control aquatic diseases at national and regional levels will effectively avert risks diseases in East Africa.

We recommend strategies such as developing a national fish health diagnostic plan with enforceable regulatory framework; build capacity of stakeholders to manage and access basic fish pathology management skills; develop participatory research that increases production and profitability; adopt simple biosecurity measures that safeguards farmed and wild fish populations; and build a communication platform that responds to disease break outs.

Acknowledgement

This work was supported by the Agricultural Technology and Agribusiness Advisory Services (ATAAS) and Lake Victoria Environmental Management Project II projects funded by Government of Uganda and World Bank. We thank our colleagues from NaFIRRI especially Mr. Martin Turyashemererwa, and the District Fisheries Department who provided additional information, insight and expertise that greatly assisted this research.

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