Tilapia Biology
On Farm Biosecurity

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Presentation outline

- **Part 1. Basics**
- Biogeography of Tilapia
- speciation
- Breeding & reproductive biology
- Advances in tilapia production technology
- **Part 2. Understanding Biosecurity concepts**
- Components of a farm biosecurity
- Farm compartmentalization
- Risk identification and matrix
- *Disease surveillance plan*
- Biocontainment and exclusion measures
- *Disinfectants and disinfections*
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- **. Documentation and record keeping**
- Biosecurity checklist

Gross microscopic examination

Primary data gathering

Biogeography of Tilapia

- There are approximately 70 species of tilapia that have been so far listed as native to Africa
- Only few tilapia species have been cultured in farms which include Nile tilapia (***Oreochromis niloticus***), Mozambique tilapia (***O. mossambicus***), three spotted tilapia (***O. andersonii***), longfin tilapia or largemouth tilapia (***O. macrochir***), Galilee tilapia (***Sarotherodon galilaeus***), Blue tilapia (***O. aureus***), blackchin tilapia (*S. melanotheron*) and redbelly tilapia (***Tilapia zillii***).
- Their distribution is historical – introductions, geophysical separations of continents
- The most popular species under aquaculture is Nile tilapia,
- Nile tilapia is a tropical species that prefers to live in shallow water.
- The lower and upper lethal temperatures for Nile tilapia are 11 °C and 42 °C respectively

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Oreochromis niloticus

Oreochromis mosambicus

Red Tilapia

Tilapia Zilli

	Substrate spawners Tilapia: T. zillii, T. rendalli T. sparmanii	Paternal /biparental mouth brooders. Sarotherodon: S. galilaeus S. melanotheron	Paternal mouth brooders Oreochromis: O. niloticus, O. aureus O. homorum, O. mossambicus O. macrochir
Broodfish morphology	Little or no dimorphism between sexes; both sexes exhibit breeding colors. Long period of pair-bonding: species monogamous at least for one brood	**Little dimorphism and color differences** between sexes. Monogamy at least for one brood	**Dimorphism between sexes. Males generally larger, with conspicuous breeding colors,** enlarged jaws, and modified papillae. Male grow 30% more than female
Spawning site	Shallow water about 50 cm deep. Substrate variable; pebbles and sand preferred. Nests solitary.	Shallow water Substrate variable, muddy sand and pebbles. Nests in common spawning grounds.	Shallow water, depth variable 0.15-8 m. Substrate variable, mud, sand and pebbles. Nests in common spawning sites.
Territorial behavior and nest building	Territory set up by both sexes and defended by both after pair bonding.	Territory established by both sexes of courting pair	Male solely sets up and defends territory and is visited by ripe females
Spawning	Long courtship lasting several days may precede spawning. Up **to 7000-8000** yolky, olive green, 1-1.5 x 1-2 mm adhesive eggs laid on pre- cleaned substrate. Male passes over eggs to fertilize them	Courtship lasting several hours to few days precedes spawning **Up to 1500** greenish-brown 1.5-2.0 x 2.0-3.5 mm non-adhesive eggs shed in batches in a shallow nest. Eggs show vestigial adhesive layer After all eggs are laid and fertilized, both parents or the males only pick up eggs for mouth brooding.	Courtship lasts several hours. Up to **2000 non-adhesive 1-2 mm x 1.5-3 mm** eggs shed in batches in shallow nest. After fertilization, each batch is picked up into the mouth by the female. Females may also snap up semen directly from genital papillae. This behavior prominent in species that have genital papillae modified into tassels to attract females.
Brood care	Both parents guard, protect, aerate the brood, and help move clutch to different nest sites. Fry at first feeding are **4-6 mm and show feeble swimming ability. Fry survival relatively low.**	Parents stay close to each other. Eggs and fry brooded in mouth until ready for release. Brood may not be collected once released. Fry are 7-9 mm at first feeding, and have well- developed fins. **Fry survival high**.	Female solely involved in brood care. After spawning, female leaves nest to rear her clutch In safety. Extended period of care during which fry seek shelter in mother's mouth. Fry brooded until free-swimming. **First feeders are already good swimmers. Fry survival high**.

General characteristic of Tilapia

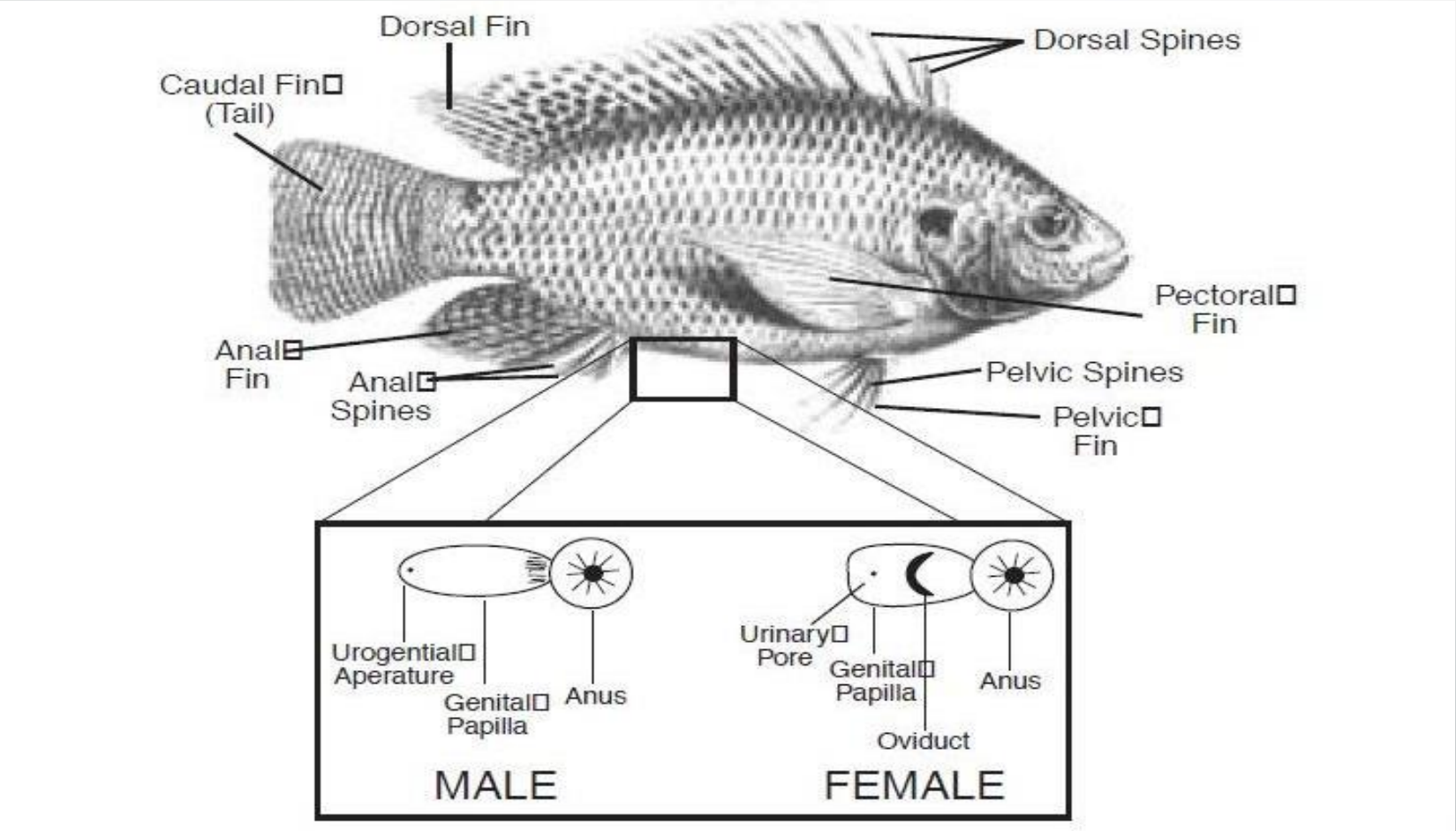
- • High yielding
- • Excellent breeder – the fish fecundity is very good. One female is capable of producing 2 fry per gram weight of female. 1800 eggs/ 500g female
- • Efficient converter of organic and agricultural wastes into high quality protein – Tilapia is an efficient filter feeder. The fish can utilize efficiently green water.
- • Resistant to disease – the fish is hard and resistant to some known disease such Epizootic Ulcerative Syndrome (EUS)
- • Very hardy – tolerates wide environmental conditions
- • Tolerant to overcrowding conditions
- • Able to grow in either fresh or brackish water

BREEDING AND REPRODUCTIVE BIOLOGY OF TILAPIA

- **Morphology of sexual organs and maturation**

The **male genital papilla is larger than that of the females** and has 2 openings the **urogenital** opening, where the milt and urine are excreted and the **anus**, for the discharge of fecal waste. The female has a flatter and shorter papilla with 3 openings; the **anus**, the **urethra** (for excretion of urine) and the **oviduct**, where eggs pass through.

Reproductive organs of tilapia



Tilapia Breeding

- These are nesting fish . Males make nest. A mark of territory.
- The nest is the breeding home for the fish
- Courtship – males court females into the nest

Depending with the species the eggs are released and fertilized. They are nurtured i.e brooded in the mouth by female or male

- The rate of egg development after fertilization is dependent on temperature of the water
- Hatchability also depends on several factors , water quality and fish handling

Egg development stages

Stage	After Fertilization
Morula stage	6-8 hrs
-Pigmentation stage	45-50 hrs
Hatching stage	70-90 hrs
Yolk sac resorption stage	6-10 days
Feeding stage	12-14 days

Breeding aspects

- Androgenisation - changing of females into males by feeding the fish with feed that has been mixed with 17 alpha methyl testestorone hormone for 21-30 days. The most common practice
- Manually selecting the males from the females. This has challenges of committing errors leading to higher mixed sex ratio
- YY male technology- this has not been adopted by many farms. Highly technical. The initial development of YY-males requires feminization of XY fry during their sexually undifferentiated stage and the identification of these newly created "sex-reversed females" (females XY) through a progeny test

Advances in tilapia production

- Artificial reproduction through collection of eggs and incubation. This works well in RAS. Care has to be taken on management of eggs, sanitation (disinfection of eggs) , management of DO, ammonia in the RAS system & Broodstock reconditioning.

  Hatchability must be above 65% and fry survival > 80%
- Selective breeding program- this is not genetic manipulation but selection for desirable traits in fish population
- RAS and bioflocs
- Cage farming – large and small cages

Critical Control Points for Tilapia reproduction

Critical control Points	Range optimal conditions	rationale
Nutrition	Maintain nutritionally balanced diet 36%CP, 7% fat, balanced vit/mineral premixes with >250 IU vit C	Higher protein and vitamins required for gonadal development.
Temperature	Maintain consistent optimum temperature in captivity 26-32°C	Fluctuation of temperature affects bio energetics of the fish and affects hormonal action in fish
Oxygen	Maintain optimum oxygen supply in ponds by aeration to avoid huge daily diurnal DO changes. <5mg/l	Oxygen is the most critical stressor and accounts for 70% fish physiological requirements.
Stocking density	Maintain not more than 4 fish /m2. Sex ratio 1: 2 or 1:3	Too many males encourages fighting. This affects breeding as territorial fights take toll in the pond
Broodstock size	200-300g. Broodstock replacement plan every year where possible.	Larger Broodstock are less productive than young. Fecundity becomes inversely related to size fo the fish. Larger fish produce larger eggs but less number of eggs.
Husbandry	Exercise less handling and disturbance at this period. Stress should be minimized. Avoid huge size differentiation between males and females. Too large males versus smaller females is not encouraged	Natural processes must be encouraged without disturbances. Maintain harmonious times of feeding and visiting the area.

KEY PERFROMANCE INDICATORS

Production Stage	Growth cycle	Feed	FCR	SGR	AGR (g/day)	Survival	Preferred culture
0.02 -2g	5 weeks	45-47% CP		13%	0.05	>85%	Ponds (hapa)
2g-15g	12 weeks	40% CP (crumbles, 2mm Pellets)	1.2-1.4	5.1%	0.14	>65%	Ponds
15g-80g	10 weeks	30-32% CP,(large crumbles, 3mm extruded pellets)	1.5-1.6	1.3%	0.95	>80%	Cages (15mm net HDP
80g-500g	20 weeks	30-32% CP,(5-6mm extruded pellets	1.6-1.8	1.2%	2.95	>85%	Cages (20mm - 32mm)
Overall	47 weeks		1.6	2.7%	1.52	>38%	

Part 2. Understanding Biosecurity concepts

- **Definition of Biosecurity**

Biosecurity can also be defined as "the compendium of measures and activities aimed to **prevent the inclusion, spread and escape of pathogens in a farming environment**".

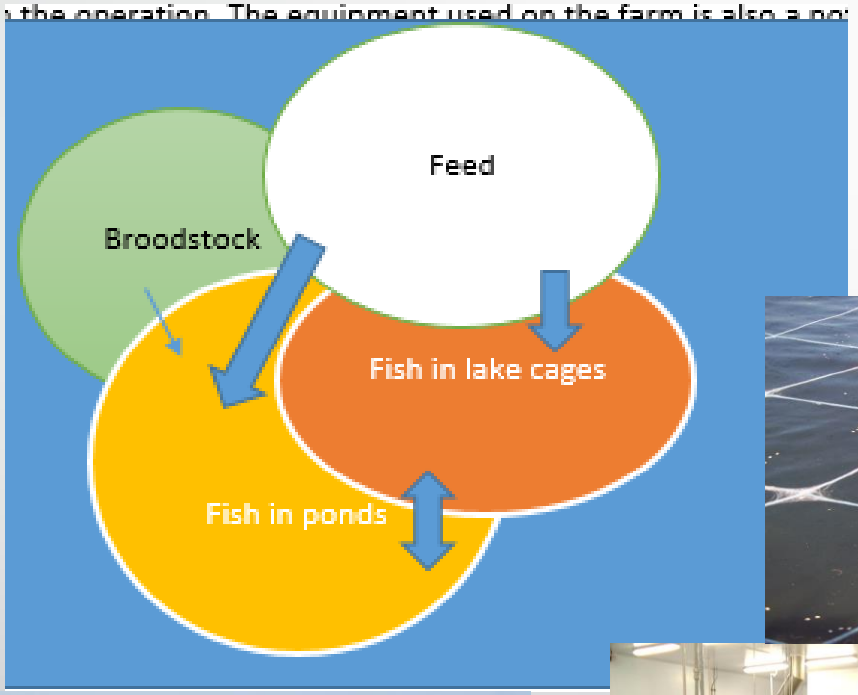
Biosecurity can be applied at trans-border level, national, regional, local level, down to a farm or even a group of farming units within a farm.

For the Purpose of this lecture, will focus on farm level biosecurity

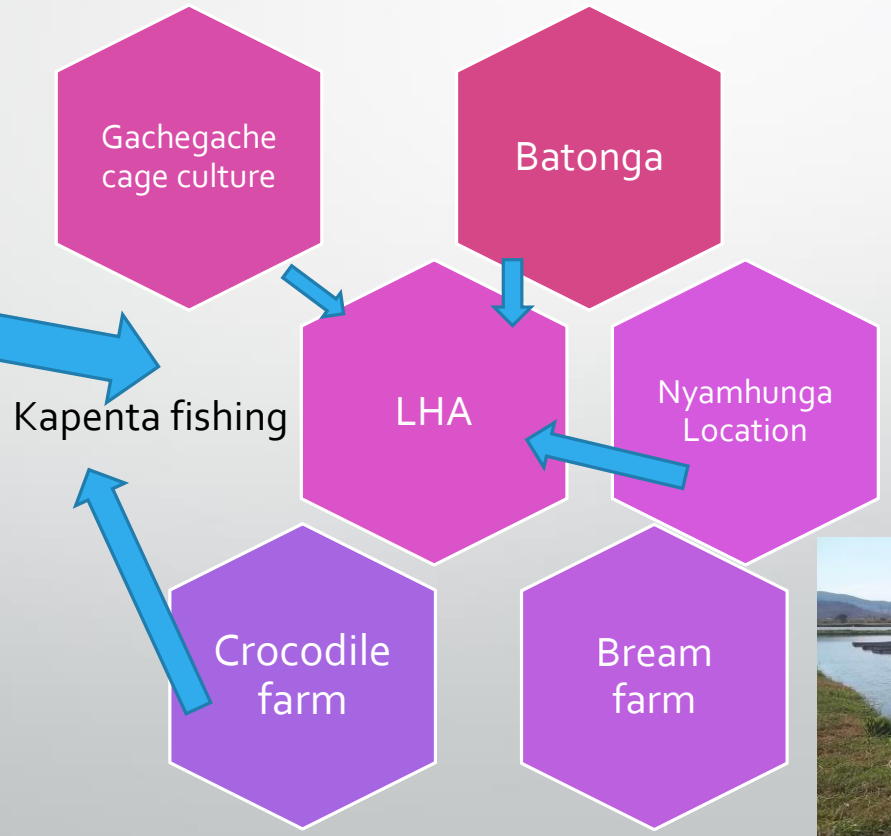
Typical Farm Lake Harvest

CAGE Sites on Lake (distance between sites 500m)

Farm compartments



Location of farm in relation to other farms and bioscenes



Importance of mapping the farm layout

- The layout of the farm (MAP) is important in several aspects.
- It reveals the distance of the farm to other farms and shows activities around the farm. Activities such as abotoirs, animal husbandry farms, major highways (transporting highways), councils sewers.
- It shows the connectivity within the business.
- . A haphazard plan is a recipe for disaster in disease management on both prevention and controlling point of view.
- The farm requires controlled access or areas (CAA)
- and Restricted Acess areas (RAA).

Mapping Biosecurity fences on A farm

- *Carry out risk assessment of each unit in the business*

  E.g hatchery , feed, cages on the lake
- *Understand the risks and their sources (RISK MAPPING)*
- *Prioritize the risk*
- *Develop critical control points for each risk*
- *Monitoring and surveillance*
- *Contingence planning*

FACTS ABOUT PATHOGEN

To carry out risk assessment the following understanding is needed

- the source of the pathogen, its preferred site and method for attaching to and penetrating the host, its adaptations for surviving the host immune system, and its strategies for obtaining nutrients required for proliferation and growth.
- Environment
- Husbandry practice
- Disease agent

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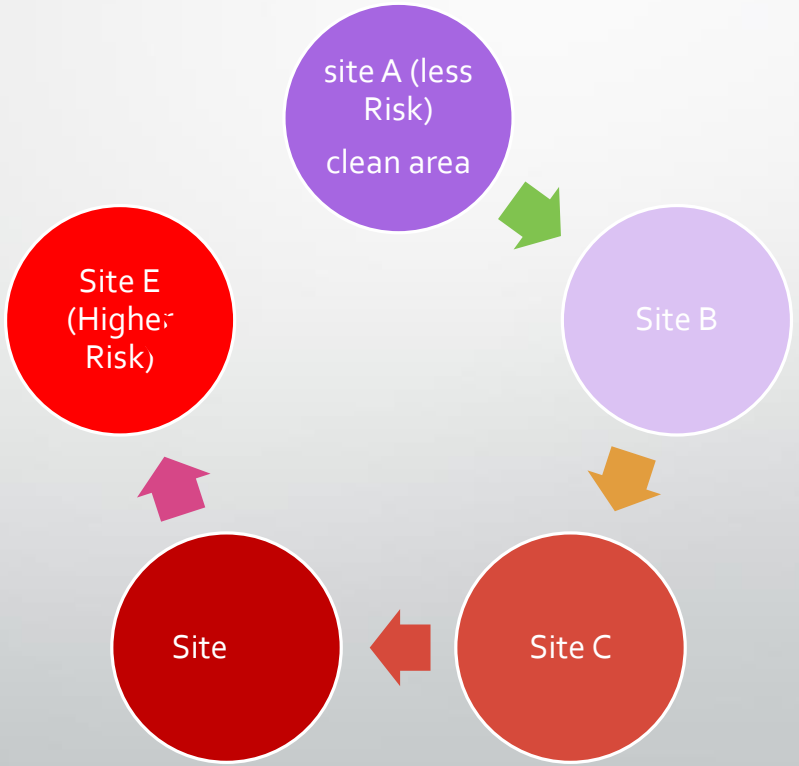
Movement	People and vehicle movement	
Water sources	Spring, dam	
Fish sources	External or internal	
Animal vectors	Dogs, birds	
Husbandry	Business practices	
Unit access	Gateways to the farm	
Unit Staff & equipment	employees	
Feed source	Internal or external	

Mapping vessel and people movement



Areas for Risk Analysis on a Farm

Risk analysis should be conducted on

- **water source,- potential sources of contaminants**
- **feed source, - raw material , transportation or processing contamination**
- **pathogen regimes, - understand potential pathogens**
- **fish sources – are fish coming from safe sources, history of infections**
- **Predators, ecological risks – birds, reptiles, people interaction**
- **Nutritional risk – inadequacy of nutritional balance of diet**

Scoring likelihood of occurrence

	Consequence			
Operational continuity	Mortality collection and treatment needed, but without interruptions to the farm	Some farm areas must be isolated for quarantining and/or treatment	Total culling of infected stocks is required. Large downtimes for disinfection expected	Long-term closure of the farm due to quarantining
Mortality rate	up to 20%	20% to 50%	50% to 70%	100%
Chronic infections	No chronic problems after the fact	Cumulative mortalities up to 50% at the end of cycle	Cumulative mortalities up to 70% at the end of cycle	Cumulative mortalities up to 100% at the end of cycle
Economics	Losses less than 10% of the production cost for the batch/year class	Partial losses amounting to 50% of the cost of production for the batch/year class	Total loss of a batch/year class	Total loss of cashflow to continue operations
Reputation	Mention at managerial level with some corrections on procedures	Internal scrutiny leading to internal auditing and revision of procedures	Persistent sector and public security	Sector concerns, government enquiries or sustained negative press
	Insignificant (1)	**Negligible (2)**	**Moderate (3)**	**Extensive (4)**

Example of scoring likelihood and impact of a pathogen

Pathogen	Type	Geographical distribution	Likelihood Score	Consequence Score	Risk level
Lactococcus garvieae (= Enterococcus seriolicida)	bacterium	Australia, Brazil, Europe, Israel, Japan, Saudi Arabia, Red Sea, South Africa, Taiwan, USA, Zimbabwe*, Zambie*	4	3	7
Flavobacterium columnare (= Flexibacter/Cytophaga columnaris)	bacterium	Worldwide	4	1	5
Saprolegnia parasitica	fungus		4	1	5
Trichodina	protozoan parasite	Cameroon, USA, Vietnam, Nigeria	4	1	5
Amyloodinium species			4	1	5

Disease prioritization

Fish Species	Pathogen	Priority Level
Tilapia (O.Niloticus)	S. agalactiae (= Str. Difficilis, S. agalactiae 1-Ia *, 1-III, 2-Ib**)	High
	Lactococcus garvieae (= Enterococcus seriolicida)	High
	Tilapia Lake Virus (TiLV)	High
	Francisella species	High
	Francisella noatunensis subspecies orientalis (asiatica)	High
	Iridiovirus	High
	Pseudomonas aeruginosa	Medium
	Pseudomonas fluorescens	Medium
	Pseudomonas mosselii	Medium
	Aeromonas hydrophila (=Aer. liquefaciens, Aer. punctata)	Medium
	Flavobacterium columnare (= Flexibacter/Cytophaga columnaris)	Medium
	Dactylogarus species	Medium
	Gyrodactylus species	Medium
	Hahella chejuensis	Medium
	S. iniae (Str. shiloi)	Low
	S. dysgalactiae	Low
	Aerococcus viridans	Low
	Aeromonas dhakensis	Low
	Edwardsiella tarda (Paracolobactrum anguillimortiferum, Edw. Anguillimortifera)	Low

Carrying out risk Analysis

Disease	Is the disease present in the area (Y/N)	Likelihood of introducing this disease	What would be the impact if introduced on production	Describe the impacts	For each disease state likelihood and impact
lactococus garviae	No	yes	significant	loss of production due to fish death fish size >300g	7

Critical control points for each sub unit e.g Hatchery

Critical Point	Yes	No		Ref. SOP
	Fish Movement			
The facility is prepared to stop all fish movements to prevent the spread of disease	yes			Fish transfer standard operating procedure,
Strict quarantine protocols are in effect for external incoming fish (In case of external fish importation)				Quarantine procedures
Fish health is monitored continuously	yes			Fish monitoring protocol, swabs protocol
Contact between farmed and wild fish is prevented	yes			Fish stocking protocol
Purchases of fishes are limited to suppliers with trusted fish health programs (In case of external fish importation)				
The health status of the fish brought into the unit is known	Yes			Fish monitoring protocol, swabs protocol
Only fishes previously inspected (visually, test) are accepted to the unit	Yes			Fish monitoring protocol, swabs protocol
Proof of treatments, vaccination, disease-free status are always recorded and requested when movement	Yes			Fish treatment protocol

CCP continued

	Equipment			
Each unit/tank has its own set of maintenance equipment		NO		Cleaning protocol, disinfection
All equipment used in the quarantine areas is exclusive to such areas				N/A
If any equipment is moved, it is carefully disinfected prior to moving	Yes			Disinfection protocol
Any non-disposable items in contact with fish are routinely disinfected between uses	Yes			
The staff knows the common disinfectants and has been trained to use them	Yes			Training manuals

CCP

	Unit access			
Access to the unit is generally restricted (e.g. physically, control points)	Yes			VHP
Only single access points to the unit are used to control and monitor visitors	Yes			VHP
Gates/doors are locked when not in use	Yes			
Correct signage and labelling is set up to inform visitors and staff about biosecurity measures in effect and the requirement for authorization access	Yes			Instructions at gate, access to premises form
Vehicle traffic into the unit is closely monitored	Yes			Security observation book
A visitor log is kept	Yes			Visitors log book
Visitors that have been in another fish farm in the previous 48h are not allowed to enter the farm	Yes			Access to Premises form
Visitors and suppliers are requested to follow unit's biosecurity measure	Yes			Access to premises

Disinfectants

- Disinfection is employed as a disease management tool in aquaculture establishments as part of biosecurity plan.
- Disinfection is used to prevent entry or exit of target pathogenic agents to or from an aquaculture establishment or compartment, as well as the spread of pathogenic agents within aquaculture establishments.
- Disinfection may be used during emergency disease response to support the maintenance of disease control zones and for disease eradication (stamping-out procedures) from affected aquaculture establishments. The specific objective of disinfection will determine the strategy used and how it is applied.

Examples of disinfectants

	Disinfectant	Indications	Comments
1	Quaternary ammonium compounds	• Virus, bacteria, hands Gill bacteria, plastic surfaces	IPN virus resistant
2	Calcium hypochlorite	• Bacteria and viruses on all Clean surfaces and in water.	Can be neutralized with sodium thiosulphate**
3	Iodine (Idophores)	• Controls bacteria, viruses, protozoa, fungi pathogens and parasites. • Eyed eggs, Gametes during fertilization. • Hands, smooth surfaces, Nets etc.	
4	Glutaraldehyde	• Controls Viral, bacterial pathogens	
5	Formaldehyde	• Controls Viral, bacterial pathogens	
6	Sodium hypochlorite	• Nets, boots and clothing	Dangerous – See precautions indicated in general recommendations

Bio containment

- **Biocontainment measures:**
  - All water and sludge exiting the facility must be disinfected
  - Personnel working in an area under quarantine should not work in other areas or should be allowed in the quarantine area only at the end of his/her working shift
  - Tanks/Ponds/Hatchery/Cages with presence of disease should be disconnected from the system
  - If handling is frequent, each tank/cage/pond should contain its own set of tools
  - Sanitary barriers (footbaths, changing areas) should be placed in entrance and exits. These areas should be kept clean and refreshed

Protocols

- The biosecurity is focused on managing, minimizing the impacts of disease and preventing disease spreading on the farm which is compartmentalized as Ponds (land operation), Cages (lake operations) and Administrative & maintenance area (offices, net mending sections on land).
- Biosecurity at LHA borders on managing the systems and processes such as Restrictions, Controls, disinfections, sanitations and hygiene aspects.

Management	Description	status
Restrictions	Some parts of the farm are restricted of entry, barricades in place. Lake ops –buoys and site markers	Available both on Lake and farm
Controls	Processes are managed through established protocols i.e. standard operating procedures, and Visitors registers	Available and being updated
Disinfections	Disinfections apply as propounded by Oie disinfection protocols. Quaternary ammonium compounds are being used on farm. e.g BAC 50	On- going, procuring alternate disinfectants
Sanitation	Highly sensitive points have sanitizers at entry points, equipment is also sanitised	On going
Hygiene	Roasters are used to manage hygiene at workplaces, swabs taken to check efficiency of the system.	On going

Protocols and disease management systems

Aspect	Description
Diagnostics services	A basic fish lab is on site fitted with microscope and other equipment for environment monitoring. Capacities to diagnose streptococosis are available,
Microscopic examination	Gross microscopic examination is done monthly on fish to check emerging disease symptoms
Gram staining programs	gram staining for strep done at company lab
Systematic sampling	A program of sampling for PCR analysis in place, regional samplings
Advanced analysis PCR	Working with Central vet lab, Singapore and UNZA for PCR analysis
Basic biosecurity facilities	Footbath, sanitisers, wheel bath facilities, PPE, in place, disposal sites available
Information Bank	Since 2007, LHA has made several isolations, information available but no markers kept for reference at site except of one sample kept at idexx lab in South Africa
Farm Risk assessments	Assessments being conducted to assess exposures i.e Feed, water quality, ecological risks, fish
Fish health audits	Audits are done each month
Surveillance system	Monitoring and surveillance program in place, streptocosis and viral disease are being monitored monthly. Viral through UNZA, bacteria through UNZA, CVL and Singapore MSD this include the emerging Tilapia Lake virus.
Disease reporting System	Updates shared on disease challenges
Antimicrobial Resistance action Plan	An updated AMR action plan available.
Vaccination	Fish vaccination taking place

Measures for Reducing Risk of introductions of pathogen

2. Preventing Pathogen Spreading

- Quarantine principles
- All in all out principle
- Disinfections
- Cleaning rosters (hygiene)
- Barriers (fence)
- Use of bird nets
- Fallowing of sites

Reduce conditions that INCREASE FISH SUSCEPTIBILITY to infections

- Stress reduction measures
- Managing fish holding units stocking densities
- Managing fish environment (DO, temperature, ammonia etc.)
- Fish health surveillance (Gross microscopic examinations)
- Good nutrition (correctly balanced nutrients)
- Good feeding practices (manage aggressive feeding frenzy)

Strategy on lake

1. There is therefore need for carrying out risk assessments
2. Monitor Dissolved oxygen levels
3. Good Management of feeds and feeding regimes
4. Fish handling protocols
5. Removal of mortalities
6. Manage safe distances between sites
7. Putting measures to reduce susceptibility

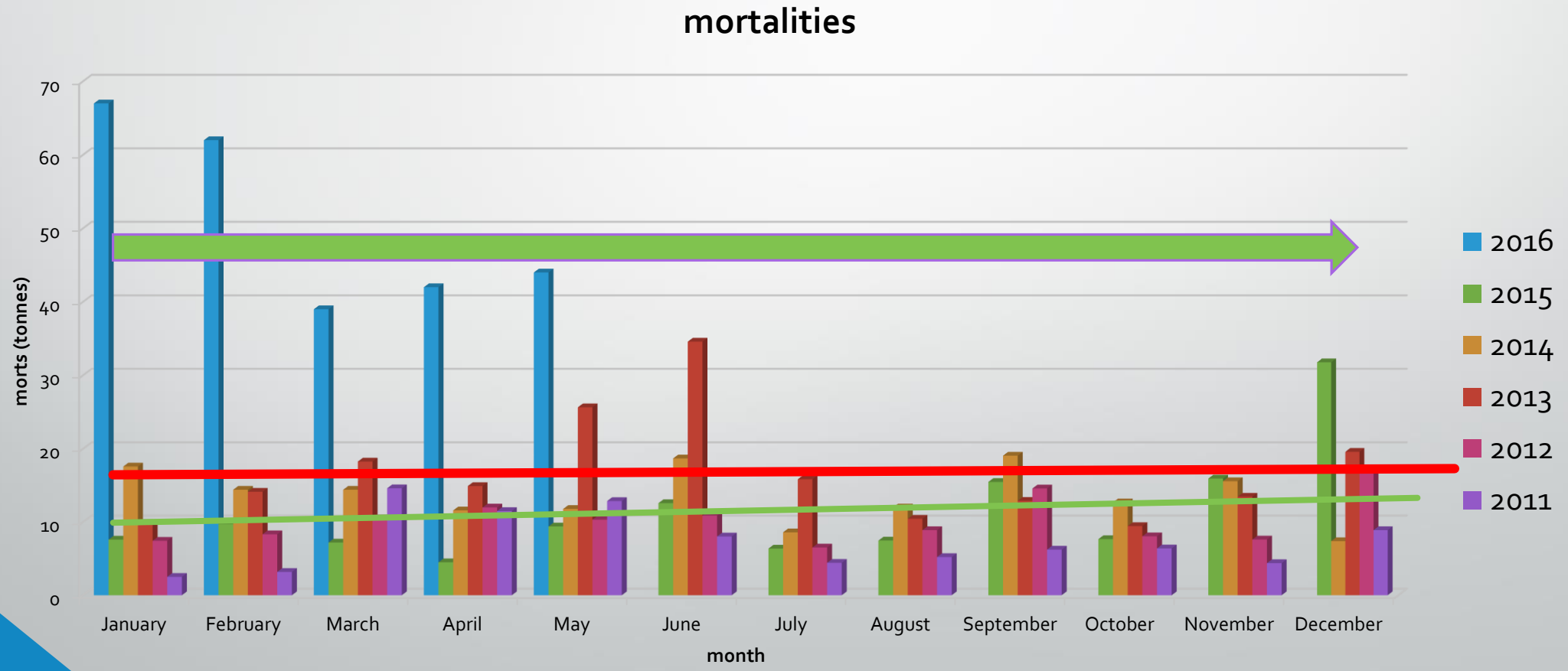
Lake Harvest

Reducing spreading of pathogen on the Lake

- Disinfection of equipment
- Nets are dried before reuse
- Bird nets – birds stress the fish as well as acting as vectors for diseases
- Regular removal of mortalities

Lake Harvest

Why talk of biosecurity monitoring?



Fish health Monitoring and Surveillance

- Fish health monitoring – checking of parasites in fingerlings , juveniles, production fish
- Documentation of fish health data
  - Fish condition
  - Recording of types of pathogen isolated
  - DO, temperature and general water quality parameters

Lake Harvest

Documentation

- Risk assessment is performed on ;

-water,

- feed,

-sites

-predators

-pathogens

- Intervention methods developed and
- Presented in a veterinary Health Plan (VHP) and this is supported by a host of protocols
- Regular audits are then performed based on HVP and protocols
- Recommendation from audits are used to review the system and update the VHP

Major issues threatening the Aquaculture Farms

- Disease threat – there is little information moving around on fish disease (Poor reporting system)
- Shared water bodies – absence of protocols or management agreements binding operators on each side of the lake
- Need for Carrying out carrying capacity studies of the Lake to avoid overloads and over intensification of production
- The threat of disease importation through fingerling imports (country preparedness on screening of fish for pathogens)
- Inadequate labs for fish pathogen examinations
- Shortage of fish specialists /veterinarians