SAMPLING AND TRANSPORT OF AQUACULTURED MARINE FISH

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Contents

1. SCOPE

This document covers the procedures for the sampling, control, handling and transport of shellfish and finfish that is aquacultured at officially monitored and controlled production areas that are approved and classified, where relevant, by Department Forestry, Fisheries and the Environment (DFFE). Sampling of shellfish and finfish takes place at various production areas that are located over a wide area in South Africa, covering but not limited to the East Coast, South-West Coast and the West Coast.

2. BACKGROUND

The risks to food safety of cultured fish include environmental and veterinary drug residues, the accumulation of biotoxins and pathogenic microbiological organisms in shellfish indicated by the presence of E. coli. The aquaculture industry is monitored and controlled in terms of the Marine Living Resources Act, 1998 (Act No. 18 of 1998). Shellfish species farmed in South Africa include *Haliotis midae* (abalone), *Crassostrea gigas* (oyster), *Mytilus galloprovincialis* (Mediterranean Mussel) and *Choromytilus meridionalis* (Black Mussel). The finfish farmed include *Argyrosomus japonicus* (Kob) and *Seriola lalandi* (Cape Yellowtail).

The South African aquaculture industry has established international markets, especially in the East for the export of cultured shellfish. The Industry with Government furthermore identified the development and promotion of the aquaculture industry as a key component of the Ocean Economy to be tapped into to promote this industry.

The NRCS and the DFFE has jointly established the South African Molluscan Shellfish Monitoring and Control Program and the South African Aquacultured Marine Fish Monitoring and Control Programme. The National Regulator for Compulsory Specifications (NRCS) and DFFE entered a Memorandum of Understanding (MoU) in March 2014 (Annexure A) in order to effectively implement the food safety programmes. The aim of the MoU is to provide the basis on which the DFFE and NRCS undertake to partner with each other in the implementation of each other's mandates. The mandates include the monitoring, control, and sampling of aquaculture products for sale for human consumption and the certification thereof for export and the implementation of the applicable requirements of the various Compulsory Specifications by the NRCS for affected products. The MoU also generally provides for all matters governing the relationship between the parties arising from the agreement. The NRCS is responsible to administer and implement the sampling programme for marine aquaculture farms in terms of the above-specified sanitation programs.

The sampling of shellfish and finfish is aimed at ensuring food safety of aquaculture products through the implementation of the food safety programmes, namely, the South African Shellfish Monitoring and Control Programme (SASM&CP), South African Aquacultured Marine Fish Monitoring and Control Programme (SAMFM&CP) and the National Residue Control Programme (NRCP). The objectives of the food safety programmes include providing guarantees to domestic, international markets and consumers that South African aquaculture products are safe for human consumption. Shellfish and finfish are deemed by the international trading countries as specialized products that command a high price in the market.

3. DEFINITIONS

a) **Aggregate sample:** means an aggregate of incremental samples taken from the same sampled portion.

b) **Aqua-cultured Marine Finfish:** means Finfish that has been cultured for human consumption.

c) **Aqua-cultured Shellfish:** means Marine Molluscan shellfish and crustaceans that have been cultured for human consumption.

d) **Filter feeders:** means species commonly referred to as mussels, oysters and clams.

e) **Final sample:** means a part of the reduced sample that is submitted for analysis.

f) **Fish:** means cultured marine living resources intended human consumption, including filter feeders shellfish, non-filters and finfish

g) **Incremental sample:** means a quantity taken from one point in the sampled portion that make up the aggregate sample must be collected at random from different representative places in the sampled portion and must all be more or less equal in size.

h) **Laboratory sample**: means a final sample intended for the laboratory (as received by the laboratory)

i) **Lot:** means an identified quantity of fish/feed determined to have common characteristics, such as origin, variety, type of packaging, packer, consignor and/or labelling, and in case of a production process, a unit of production from a single plant using uniform production parameters or a number of such units, when produced in continuous order and stored together.

j) **Non-filter feeders:** means species commonly referred to as gastropods, Pectinidae, urchins and crustaceans

k) **Reduced sample:** means the aggregate sample after it has been thoroughly mixed into one homogenous sample

l) **Representative Sample**: means a sample that is drawn on a random basis, that is unaltered from sampling until testing and that has been taken at the correct sampling point in the production area and representative of the production lot.

m) **Sampled portion:** means a lot or an identified part of the lot that will be sampled and that is of homogenous nature.

4. GENERAL PRINCIPLES OF SAMPLING

Sampling shall be strictly executed according to this procedure:

4.1. Sampling shall be executed at the harvesting area and not at the pack-house or at shore, to ensure traceability of the samples at all times

4.2. Sampling shall strictly be undertaken by the sampling officer

4.3. Samples shall under no circumstances leave the control of the sampling officer unless couriered by a third party to the laboratory.

4.4. Veterinary medicine residue samples that are not personally delivered to the laboratory by the sampler must be placed into a tamper-proof bag and sealed before the sample is dispatched

4.5. Samples shall only to be taken at the officially demarcated sampling points, each with unique coordinates noted in a sampling plan.

4.6. No samples shall be taken in the absence of such official sampling stations, when applicable

4.7. In the event where the sampling cannot be executed at the required frequency, due to unforeseen circumstances (bad weather, breakdown of transport vessel), then this to be

immediately reported to the manager responsible for the production area. Sampling to be executed the very next day if unforeseen circumstances do not persist.

4.8. The temperature of samples with a transport time of less than 4 hours to be less than the temperature at the point of sampling, when reaching the laboratory
4.9. Samples should be cleaned of dirt, sediment and other organisms
4.10. Samples shall be cleaned in the sea water at the sampling site
4.11. Samples shall be drained before being placed in the sampling bag
4.12. The labels to be affixed to the sampling bags directly after sampling
4.13. Damaged and diseased specimens shall not be sampled
4.14. Sampling shall be done under optimum hygienic conditions
4.15. Bio-security procedures at production sites to be respected and followed
4.16. Full traceability shall be ensured for all samples at the sampling point
4.17. Sampling officers to report unusual observations, such as boating activity, dredging activities, animals in the water, plankton blooms, etc.)
4.18. Identify, package and store samples without delay after the sample has been taken
4.19. On becoming aware that an unsuitable sample has been taken, notify the laboratory and FSO within 24 hours by phone, followed up within 3 working days in writing
4.20. Individually pack each sample in packaging so that the sample does not contaminate any other sample or packaging material, and to prevent any error in identification of the sample
4.21. All necessary equipment shall at all times be available and coordinated by the sampler
4.22. All samples to be secured and sealed by the sampler to ensure that it is tamper proof
4.23. For veterinary residues and contaminants testing the residue sampling report to be completed.
4.24. For micro testing the BF 97 to be completed
4.25. For bio-toxin testing the BF 42A to be completed
4.26. All samples to be accompanied by a sample submission form obtainable at the respective laboratories
4.27. The sampling form shall be clearly and neatly completed for all samples
4.28. Samples shall be identified with the following information on the labels:

- collectors name
- the time and date of sampling
- the facility name and facility number or sample station from which the sample was taken
- official code number of the sample for veterinary medicine and environmental residues samples
- the sample type e.g. mussel, abalone, finfish etc.
- intended analysis

5. SAMPLING AND TESTING OF VETERINARY MEDICINE AND ENVIRONMENTAL RESIDUES

5.1. The production facility, veterinary medicine and environmental residues to be tested for in finfish and shellfish are listed in the National Residue Plan (NRP).
5.2. The sample size and sampling frequency for the testing of fish is outlined in the National Residue Control Programme.
5.3. The samples shall be submitted to the laboratory indicated in the National Residue Control Programme.
5.4. The procedure for the sampling required during the implementation of contingency measures is outlined in in Figure 1.
5.5. Samples collected for analysis for veterinary medicinal products and environmental contaminants (excluding dyes) shall comprise of stock close to the market size.

5.6. Samples to be tested for unauthorized substances (including dyes) shall be taken at farm level at all stages of farming including fish ready to be placed on the market for consumption.

5.7. The samples shall be placed into a waterproof bag that is tied off and then placed into a tamperproof bag that is to be securely sealed to prevent tampering of the sample.

5.8. The samples bag shall be clearly labeled as outlined below.

5.9. The Residue Sampling Report (Appendix 2) shall be completed and submitted to the Food Safety Office of the Directorate: Sustainable Aquaculture Management (SAMSanitation@dffe.gov.za) within 24 hours of sampling.

5.10. Where a tamperproof bag is used, the seal number shall be accurately recorded on the Residue Sampling Report (Appendix 2) and the Laboratory Sample Submission Form (Appendix 3).

5.11. The samples shall be chilled for same day delivery, otherwise frozen.

5.12. The laboratory shall log whether the sample arrived in a tamper proof bag and if it was tampered with.

5.13. Should the sample arrive in tamper proof bag, the laboratory shall cut the bag open along the dotted line at the bottom of the tamper proof bag as evidence that the seal at the top of the bag was not tampered with.

5.14. A sub-sample and, the tamper proof bag, shall be kept by the laboratory until requested to be disposed of by the Food Safety of office of the Directorate: Sustainable Aquaculture Management. This is in the event of a retest being required for legal processes.

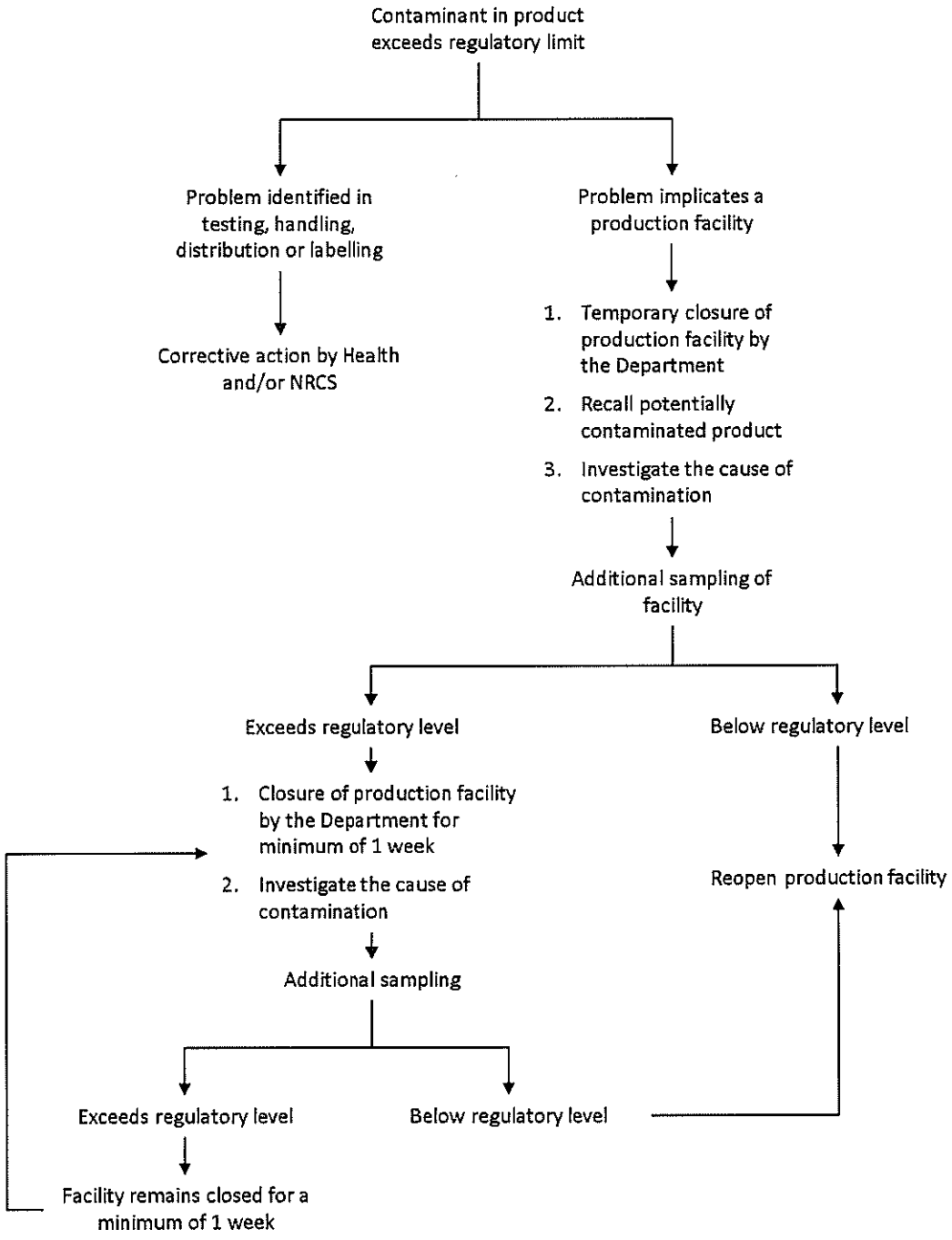

Figure 1: Veterinary medicine, environmental residue and radionuclide contingency plan

6. SAMPLING AND TESTING OF MICROBIOLOGICAL CONTAMINANTS

6.1. The filter feeder samples shall be taken from the production areas.
6.2. The non-filter feeder samples shall be taken from the end-of-line lots.
6.3. The sample size and sampling frequency for filter feeders is outlined in the Microbiological and Biotoxin Sampling Schedule.
6.4. Samples collected for analysis for microbiological contaminants shall comprise of stock close to the market size.
6.5. The stated Representative Monitoring Point (RMP) location should be used to identify the position of sampling for bivalves.
6.6. The samples shall be placed into a sterile waterproof bag that is tied off and then placed into a second clear bag.
6.7. The samples shall be clearly labelled as outlined below.
6.8. The label shall be placed into a waterproof sleeve and placed into the second bag referred to above.
6.9. Samples should be transported in cooler boxes between 2˚C and 10˚C.
6.10. In the event of overnight storage, micro samples to be stored at 3±2˚C.
6.11. The samples shall arrive at the laboratory within 24 hours of sampling.

Table1: Number of samples for microbiological contaminants per commodity

Species	**Min Sample size (Count) (Min 100 g flesh)**
Abalone	10
Lobster	5
Urchin	5
Pectinidae	5
Finfish	5
Mussels	20
Oysters	20
Clams	20

7. SAMPLING AND TESTING OF MARINE BIOTOXINS

7.1. The following biotoxins shall be tested for in filter feeders and non-filter feeders that utilize natural seawater for production:
- Paralytic Shellfish Toxin (PST)
- Lipophilic Shellfish Toxins (LST)
- Amnesic Shellfish Toxin (AST)

7.2. The filter feeder samples shall be taken from the production areas from a depth of at least 1 meter below the surface.
7.3. The non-filter feeder samples shall be taken from the end-of-line lots.
7.4. The sample size and sampling frequency for the shellfish is outlined in the Microbiological and Biotoxin Sampling Schedule.
7.5. Sampling should be done by the method used for commercial harvesting
7.6. The temperature of the water to be measured and recorded on the BF 97 and BF 42A as applicable

7.7. The stated Representative Monitoring Point (RMP) location should be used to identify the position of sampling for bivalves.
7.8. Samples collected for analysis for biotoxins shall comprise of stock close to the market size.
7.9. The samples shall be placed into a waterproof bag that is tied off and then placed into a second clear bag.
7.10. The samples shall be clearly labelled as outlined below.
7.11. The label shall be placed into a waterproof sleeve and placed into the second bag referred to above.
7.12. The samples shall be chilled for same day delivery, otherwise frozen.

Table 2: Number of samples for biotoxin testing per commodity

Species	**Sample size (Count) (Min 100 g flesh)**
Abalone	5
Lobster	5
Urchin	5
Pectinidae	5
Finfish	5
Mussels	30
Oysters	20
Clams	20

8. SAMPLING AND TESTING OF RADIONUCLIDES

8.1. Samples shall be taken for the testing Radionuclides (Cesium 134 and 137).
8.2. The procedure for the sampling required during the implementation of contingency measures is outlined in in Figure 1.
8.3. The samples shall be submitted to the laboratory indicated in the National Residue Control Programme.
8.4. The sample size and sampling frequency for the testing of fish is outlined in the National Residue Control Programme.
8.5. The samples shall be placed into a waterproof bag that is tied off and then placed into a second clear bag.
8.6. The samples shall be clearly labelled as outlined below.
8.7. The label shall be placed into a waterproof sleeve and placed into the second bag referred to above.
8.8. The samples shall be chilled for same day delivery, otherwise frozen.

9. SAMPLING METHODOLGY OF VETERINARY MEDICINE AND ENVIRONMENTAL RESIDUES

9.1. SAMPLING OF FILTER FEEDER AND NON-FILTER FEEDER LOTS

9.1.1. Randomly select the minimum number of incremental samples to be taken from the lot or sublot as given in Table 3.
9.1.2. The aggregate sample combining all incremental samples shall be at least **1 kg**.
9.1.3. The incremental samples shall be of similar weight.
9.1.4. The weight of an incremental sample shall be at least **100 grams**.

Table 3: Minimum number of incremental samples to be taken from the lot or sublot

Weight of lot/sublot (kg)	Minimum number of incremental samples to be taken
<50	3
50-500	5
>500	10

9.2. SAMPLING OF FINFISH LOTS

9.2.1. Fish are considered to be of comparable size and weight where the difference in size and weight does not exceed about 50 %.

9.2.2. The number of incremental samples to be taken from the lot are defined in **Table 3 and it shall comprise of meat and skin in its natural proportions, excluding the guts.**

9.2.3. The aggregate sample uniting all incremental samples shall be at least **1 kg**.

9.2.4. Where the lot to be sampled contains small fish (individual fish weighing < about 1 kg), the whole fish is taken as incremental sample to form the aggregate sample.

9.2.5. Where the resulting aggregate sample weighs more than 3 kg, the incremental samples may consist of the middle part, weighing each at least 100 grams of the fish forming the aggregate sample.

9.2.6. The whole part to which the maximum level is applicable is used for homogenisation of the sample.

9.2.7. The middle part of the fish is where the center of gravity is. This is located in most cases at the dorsal fin (in case the fish has a dorsal fin) or halfway between the gill opening and the anus.

9.2.8. Where the lot to be sampled contains larger fish (individual fish weighing **more than about 1 kg)**, the incremental sample consists of the middle part of the fish. Each incremental sample weighs at least 100 grams.

9.2.9. **For fish of intermediate size (about 1-6 kg)** the incremental sample is taken as a slice of the fish from backbone to belly in the middle part of the fish.

9.2.10. For very large fish (e.g. > about 6 kg), the incremental part is taken from the right side (frontal view) dorso- lateral muscle meat in the middle part of the fish. Where the taking of such a piece of the middle part of the fish would result in significant economic damage, the taking of three incremental samples of at least 350 grams each may be considered as being sufficient independent of the size of the lot or alternatively an equal part of the muscled meat close to the tail part and the muscle meat close to the head part of one fish may be taken to form the incremental sample being representative for the level of dioxins in the whole fish.

9.2.11. For the sampling of lots of fish containing whole fish of different size and/or weight, the samples shall be constituted as for the predominant size or weight category.

9.2.12. Where a size or weight class/category is predominant (about 80 % or more of the lot), the sample is taken from fish with the predominant size or weight. This sample is to be considered as being representative for the whole lot.

9.2.13. Where no particular size or weight class/category predominates, then it must be ensured that the fish selected for the sample are representative for the lot.

9.3. SAMPLING OF FEEDLOTS

9.3.1. Randomly select the required number of units as indicated in **Table 4** from the whole sampled portion for sampling.

9.3.2. Take the incremental sample from each of the selected units using a shovel and place into a container to make up the aggregate sample of **4 kg.** The incremental samples must be of approximately equal size.

9.3.3. Thoroughly mix the incremental samples in the container to make up the reduced sample (aggregate sample).

9.3.4. Subsample the final sample (500g) designated as the laboratory sample and place into a container (plastic bag) that is securely sealed and include the letter "L" on the container label.

9.3.5. Subsample the final sample (500g) designated as the retention sample and place into a container (plastic bag) that is securely sealed and include the letter "R" on the container label.

9.3.6. If the sample portion (Lot) consists of 20 bags or less, subsample the final samples (500g) directly from the selected feed bag.

9.3.7. The laboratory sample and the retention sample shall then be placed into a clearly labelled tamperproof bag that is sealed.

9.3.8. Should there be multiple lots of feed on the premises each lot shall be sampled separately as indicated above

Table 4: Number of units (bags) sampled from each sample portion(production lot)

Size of sampled portion (units)#	**Minimum number of units from which incremental sample is taken (units) #**
1 to 20	1
21 to 150	3
151 to 400	5
> 400	¼ of the √ number of units making up the sampled portion, up to 40 units*

Where the number obtained is a fraction, it shall be rounded up to the next whole number.

Feed that is packaged in bags is referred to in the table as units.

10. EQUIPMENT REQUIREMENTS FOR SAMPLING

The availability and the use of proper equipment for the protection and preservation of the samples are essential during the sampling process. As is highlighted in section 4 above, the sampler is responsible for ensuring that equipment is always available to sample the required number of samples during a sample run per day and that the equipment is arranged from the laboratory or the regional office timeously, in order to prevent any deviation from the sampling procedure.

The following equipment to be available as is applicable:

- Sterile micro bags for microbiological samples
- Chemical bags for residue and biotoxin samples
- Tamper proof security bags for residue samples
- Cleaning buckets and brush

- Container for homogenising samples for residue samples
- Cooler box
- Ice packs
- Insulating foam when necessary
- Self- adhesive labels
- Permanent markers for labelling
- Thermometer
- Disinfectant
- Shovel for feed (Small handheld with sides)

11. PREPARATION AND PACKAGING OF SAMPLES

11.1. Samples, especially shellfish to be rinsed with fresh potable water or clean sea water from the immediate sampling area

11.2. Shellfish not to be totally re-immerse in water to prevent the possible introduction of contamination at the sampling point

11.3. Ice packs and foam packaging to be used when the transport time is more than 4 hours to the laboratory as per the following configuration:

TOP 2 LAYERS OF FOAM
TOP LAYER OF 3 ICE PACKS
TOP LAYER OF FOAM
SAMPLES
BOTTOM LAYER OF FOAM
BOTTOM LAYER OF 3 ICE PACKS

11.4. When the transport time is less than 4 hours, then other suitable insulating material may be used such as bubble wrap, newspaper, etc.)

11.5. Samples intended for micro testing must not be frozen

11.6. The time and temperature to be recorded on the sample submission form

12. TRANSPORT OF SAMPLES

12.1. The above stipulated storage and transport temperature to be validated at the testing laboratory on arrival

12.2. Cooler boxes with effective insulating capabilities are to be used for the transport of samples.

12.3. When samples are couriered to the laboratory, the sampling officer should liaise with the laboratory regarding the sample delivery and the requirement to measure the sample surface temperature and to record this and the condition of the samples

12.4. The sampling officer to ensure that the surface temperature of the sample is measured and that this and the condition of the samples are recorded at the laboratory

13. HEALTH AND SAFETY AND BIOSECURITY MEASURES

13.1. Sampling officers shall:

13.2. Comply with the Health and Safety policies of the NRCS.

13.3. At all times respect, familiarize and implement the bio-security measures that are prevalent at the sampling points/ farms.
13.4. Treat all disposable items as clinical waste.
13.5. Ensure that hands are disinfected between each sampling station to prevent the spread of disease or contaminate the next sample in the series.
13.6. Ensure that sampling equipment is clean before re-use to prevent cross-contamination of samples.
13.7. Ensure that sampling equipment is suitable for the samples to be taken.

14. REFERENCES

14.1. South African Shellfish Monitoring and Control Programme
14.2. South African Aquacultured Marine Fish Monitoring and Control Programme

15. ATTACHMENTS

APPENDIX 1: BF 42 A
APPENDIX 2: RESIDUE SAMPLING REPORT
APPENDIX 3: LABORATORY SAMPLE SUBMISSION FORM

APPENDIX 1: BF 42 A

RETURN OF SAMPLES

Product:	From:	Date sampled:	Carton No:	
Live Cultivated Abalone: ☐	Hermanus: ☐			Sampled by:
Live Cultivated Oysters: ☐	West Coast: ☐			Signature:
Live Cultivated Mussels: ☐	Northern Cape: ☐			Date:
	Eastern Cape: ☐			

Distribution

Name of Laboratory: Assurecloud ☐ Other ☐

	Sample mass (g)	Biotoxin			Time Sampled	Remarks
		PST	LST	AST		
						Please test for: PST ☐ LST ☐ AST ☐
Farm Number/Name:						NOTE: A Report of the results must be forwarded to:
Number of Animals:						COMPANY NAME:
Representative samples of Cultivated Live Abalone from different tanks ☐						DFFE ☐
Representative samples of Cultivated Live Oysters from different tanks ☐						NRCS ☐
Representative samples of Cultivated Live Mussels from different tanks ☐						
			Batches sampled:	Size class	Batch code	
Harvesting date:						
Consignment date:						
Test date:						
Witnessed by:			Received by:			
Date:			Date:		Time:	
Time:			Water Temperature:			

Comments:

BF 42A: 01-02-2023

APPENDIX 2: RESIDUE SAMPLING REPORT

RESIDUE SAMPLING REPORT
(email completed copy to SAMSanitation@daff.gov.za)

DAFF Sampling Schedule ID Number: (1) 2017/FF/01 & (2) 2017/FF/02
(copy from sampling schedule)

Date: 2017/01/20 Time: 13h00

NRCS Sampler's Name: CS Hingle & B. Govender

Farm name: Mtunzini Fish Farm Code: 3867

Address 2 Hailey Street
Mtunzini

Farm Manager's name or representative: Mr Niall Stallard

Sample Identification

Sample Type: (Fin fish) / abalone / oysters / mussels (delete as applicable)

Age Category: 6 months

Pond identification: Panda Pool 13

Origin if not from farm: —

Treatment in 4 weeks before: Hydrogen peroxide & Formalin

Number of individuals: ± 40 units [760g]

Test Method Names required (1) Stilbenes (A1)
(copy from Sampling schedule) (2) Hormones (A3)

Test Method IDs required (1) & (2) 0337
(copy from sampling schedule)

Evidence Bag seal number: AS 037996

Sample type: (Routine) / Suspect (delete as applicable)

Owner notes:

Number of reports 1st copy

Signature owner

Signature sampler

APPENDIX 3: LABORATORY SAMPLE SUBMISSION FORM

SF.OUT.001 / REVISION NO: 2

REQUISITION FORM: OTHER MÉRIEUX NUTRISCIENCES SERVICES

Report to:

Account to:

Swift Silliker (Pty) Ltd t/a Mérieux NutriSciences
7 Warrington Road / Claremont
Cape Town / South Africa / 7708
Tel: +27 (21) 683 8436 / 08613 SWIFT
Fax: +27 (21) 683 8422 / Email: za-info@mxns.com
www.merieuxnutrisciences.com

Date: _____
Order No.: _____
Time Rec: _____
Tel. No.: _____
Fax No.: _____

PRODUCT TYPE	DESCRIPTION	TEST CODE	TEST METHOD	LABORATORY	ADDITIONAL COMMENTS

Customer: _____ Processed by: _____

Signature: _____

16. AMENDMENT REGISTER

Amendment History		
Amendment No.	**Date Approved**	**Nature of Amendment**
00	2018-03-01	First issue
01	2022-11-01	Second Issue: Entire document reviewed. Feed sampling, sampling methods for environmental and veterinary medicines residues and more definitions were included.
02	2023-05-19	Third Issue: The number of animals for microbiological testing for mussels, oysters and abalone have been amended to be in line with ISO