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DRAFT MALAWI STANDARD (SADC HARMONIZED)



Fresh and frozen whole fin fish – Specification

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FOREWORD

This draft standard is a Southern African Development Community (SADC) Harmonized Text covering the requirements and methods of tests for fresh and frozen whole fin fish.

The harmonization of standards and technical regulations in the SADC region is an obligation under the SADC protocol on Trade which was established under the SADC Treaty to provide for elimination of tariffs and non-tarriff barriers to trade.

This draft standard is identical to SADC HT 81, Fresh and frozen whole fin fish – Specification.

Acknowledgement is made for the use of the above standard.

TECHNICAL COMMITTEE

This draft standard was prepared by the Technical Committee MBS/TC 39, *Fish and fishery products,* and the following companies, organizations and institutions were represented:

Malawi Bureau of Standards.

MALDECO Fisheries

Malawi College of Fisheries;

Ministry of Agriculture, Irrigation and Water Development - Department of Fisheries;

Lake Harvest;

Lilongwe University for Agriculture and Natural Resources;

NOTICE

This standard shall be reviewed every five years, or earlier when it is necessary, in order to keep abreast of progress. Comments are welcome and shall be considered when the standard is being reviewed.

DRAFT MALAWI STANDARD

Fresh and frozen whole fin fish - Specification

1 SCOPE

This draft standard specifies requirements and methods of sampling and test for fresh and frozen whole fin fish intended for human consumption.

2 NORMATIVE REFERENCES

The following standard contains provisions, which through reference in this text, constitute provisions of this draft standard. All standards are subject to revision and, since any reference to a standard is deemed to be a reference to the latest edition of that standard, parties to agreements based on this draft standard are encouraged to take steps to ensure the use of the most recent edition of the standard indicated below. Information on current valid national and international standards can be obtained from the Malawi Bureau of Standards.

MS 19: Labelling of prepacked foods – General standard;

MS 21: Food and food processing units – Code of hygienic conditions;

MS 188: Edible salt – Specification;

MS 214: Potable water – Specification;

MS 237: Food additives – General Standard;

MS 302: General standard for contaminants and toxins in foods and feed;

MS 790: Code of practice for fish and fishery products;

MS 1241: Guidelines for the sensory evaluation of fish and shellfish in laboratories;

CODEXSTAN 233: Sampling plans for prepackaged foods (AQL-6.5);

ISO 4833: Microbiology of food and animal feeding stuffs – Horizontal method for the enumeration of microorganisms – Colony-count technique at 30 degrees;

ISO 6579: Microbiology of food and animal feeding stuffs – Horizontal method for the detection of Salmonella spp.;

ISO 6888: Microbiology of food and animal feeding stuffs – Horizontal method for the enumeration of coagulase-positive staphylococci (Staphylococcus aureus and other species);

ISO 7251: Microbiology of food and animal feeding stuffs – Horizontal method for the detection and enumeration of presumptive Escherichia coli – Most probable number technique;

ISO 7937: Microbiology of food and animal feeding stuffs – Horizontal method for the enumeration of Clostridium perfringens – Colony-count technique;

ISO 11290: Microbiology of the food chain – Horizontal method for the detection and enumeration of Listeria monocytogenes and of Listeria spp.;

ISO 16050: Foodstuffs – Determination of aflatoxin B1, and the total content of aflatoxin B1, B2, G1 and G2 in cereals, nuts and derived products – High performance liquid chromatographic method;

ISO 21527-1: Microbiology of food and animal feeding stuffs – Horizontal method for the enumeration of yeasts and moulds – Part 1: Colony count technique in products with water activity greater than 0.9;

ISO/TS 21872-1: Microbiology of food and animal feeding stuffs – Horizontal method for the detection of potentially enteropathogenic Vibrio spp. – Part 1: Detection of Vibrio parahaemolyticus and Vibrio cholera; and

ISO/TS 21872-2: Microbiology of food and animal feeding stuffs – Horizontal method for the detection of potentially enteropathogenic Vibrio spp. – Part 2: Detection of species other than Vibrio parahaemolyticus and Vibrio cholera;

ISO 27085: Animal feeding stuffs – Determination of calcium, sodium, phosphorus, magnesium, potassium, iron, zinc, copper, manganese, cobalt, molybdenum, arsenic, lead and cadmium by ICP- AES

AOAC 990.04: Mercury (Methyl) in sea food by liquid chromatography-Atomic A b s o r p t i o n Spectroscopy (LC-AAS); and

AOAC 977.15 / 974.14: Mercury in fish by Flame Atomic Absorption Spectroscopy (FAA).

3 DEFINITIONS

For the purpose of this draft standard, the following definitions shall apply:

3.1

fresh fin fish

fish that has not been frozen, dried or otherwise preserved, except by chilling including fish in rigor

3.2

food grade

packages which safeguard the hygienic, nutritional, technological and organoleptic qualities of the products

3.3

gutted fish

fish from which the guts have been removed. Gutting consists of bleeding the fish and removal of the stomach and gut

3.3

rigor mortis

the stiffening of the muscles of fish which results from a series of complex changes that take place in the tissues shortly after death

3.4

fin fish

fresh water and marine vertebrate fish

3.5

whole fish

fish as captured, ungutted

3.6

chilling

The process of cooling fish to a temperature approaching that of melting ice (0 °C to 4 °C)

3.7

clean water

water which meets the same microbiological standards as potable water and is free from objectionable substance.

3.8

frozen fish

fish that has been subjected and maintained at temperatures of -18 °C or lower

4 ESSENTIAL COMPOSITION AND QUALITY FACTORS

4.1 General requirements

Fresh and frozen finfish may be presented as uneviscerated, eviscerated, and shall;

- **4.1.1** be in a sound, wholesome condition;
- **4.1.2** be bright in appearance;
- 4.1.3 have a fresh characteristic smell of its species;
- 4.1.4 have prominent, bright, clear and moist eyes
- 4.1.5 have bright red gills;
- **4.1.6** have bright abdominal blood;
- **4.1.7** be firm, and shall have elastic flesh;
- **4.1.8** have scales which adhere strongly to the skin where this is normal;
- 4.1.9 be free from diseases and parasites; and
- **4.1.10** Where present on fresh fish; slime shall be transparent.

4.2 Specific requirements

- **4.2.1** Fresh fish shall be stored at chilling temperature;
- 4.2.2 Frozen fish shall be maintained at a temperature of -18 °C or lower; and

4.2.3 When tested as per annex A the level of histamine in species with high levels of histidine such as scromboids shall not exceed 10 ppm.

4.3 Microbiological limits

The product shall comply with microbiological limits given in Table 1.

Table 1: Microbiological limits for fresh and frozen whole fin fish

S/No	Micro-organisms	Max. limits	Method of test
i)	Salmonella per 25 g	Absent	ISO 6579
ii)	<i>E. coli</i> per gram	Absent	ISO 7251
iii)	Listeria monocytogenes	Absent	ISO 11290 Part 1
iv)	Staphylococcus aureus cfu per gram	10 ²	ISO 6888
v)	Clostridium perfringens per gram	Absent	ISO 7937
vi)	Vibrio Spp per gram	Absent	ISO 21872
vii)	Total viable count per gram	10 ⁵	ISO 4833

6 FOOD ADDITIVES

If used, food additives shall comply with MS 237.

7 CONTAMINANTS

Fresh and frozen whole fin fish shall conform to those maximum levels for heavy metals and other contaminants as stipulated in MS 302.

8 HYGIENE

8.1 The products covered by the provisions of this draft standard shall be prepared and handled in accordance with the appropriate sections of the MS 21 and 790.

8.2 The final product shall be free from any foreign material that poses a threat to human health.

9 PACKAGING AND LABELLING

9.1 Packaging

Fresh and frozen whole fin fish shall be packaged in food grade containers.

9.2 Labelling

In addition to the requirements in MS 19, the following specific labelling requirements shall apply and shall be legibly and indelibly marked:

- 9.2.1 Name of the product shall be "Fresh or frozen fin fish";
- 9.2.2 Storage and transportation conditions declaring the temperature to be -18 °C or lower;
- 9.2.3 Name and physical address of processor;
- 9.2.4 Net weight in metric units;
- **9.2.5** Date of production;
- 9.2.6 Batch or code number;
- 9.2.7 Expiry date; and

9.2.8 Country of origin and/or water body.

9.3 Labelling of non-retail containers

Information on the above provisions shall be given either on the container or in accompanying documents, except that the name of the product, lot identification, and the name and address of the processor or packer as well as storage instructions, shall appear on the container.

However, lot identification, and the name and address of the processor or packer may be replaced by an identification mark provided that such a mark is clearly identifiable with the accompanying documents.

10 METHOD OF SAMPLING AND TEST

Sampling and tests shall be done as per test methods described in respective tables in this draft standard.

ANNEX A

(Normative)

DETERMINATION OF HISTAMINE

A.1 PRINCIPLE

Sample is extracted with 75 % (v/v) methanol. Extract is passed through ion exchange column. o– Phthaldialdehyde solution is added to eluate to form fluorescent histamine derivatives. Fluorescent intensity of derivatives is measured using fluorometer and histamine is quantified using external standards.

A.2 APPARATUS

Rinse all plastic and glass containers with HCI (1 + 3) and H₂O before use.

A.2.1 Chromatographic tube - 200 7 id mm polypropylene tube fitted with small plastic stopcocks and ca 45 cm Teflon tubing. Control flow rate at >3 ml/min by adjusting height of column relative to tubing outlet. Alternatively, use 2-way valve in place of tubing.

A.2.2 Photofluorometer - equipped with medium pressure Hg lamp with excitation at 350 nm and measuring emission at 444 nm.

A.2.3 Pipettes - 1 and 5 ml

A.3 REAGENTS

A.3.1 Ion-exchange resin, Bio-Rad AG 1- X 8, 50 – 100 mesh or Dowex 1- X 8, 50 – 100 mesh. Convert to -OH form by adding ca 15 m 2 M NaOH/g resin to beaker. Swirl mixture and let stand < 30 min. Decant liquid and repeat with additional base. Thoroughly wash resin with H₂O, slurry into fluted paper and wash again with H₂O. Prepare resin fresh weekly and store under H₂O. Place glass wool plug in base of tube, **A.2.1**, and slurry in enough resin to form 8 cm bed. Maintain H₂O level above top of resin bed at all times. Do not regenerate resin in packed column; rather, use batch regeneration in beaker when necessary. Wash column with about 10 ml H₂O before applying each extract.

A.3.2 Phosphoric acid, 3.57 N. Dilute 121.8 ml 85 % H_3PO_4 to 1 litre. For other concentration H_3PO_4 , volume required for 1 litre 1.19 M acid = 17493/ (density H_3PO_4 percent H_3PO_4). Standardize 5.00 ml by titration with 1.00 M NaOH to phenolphthalein end point, and adjust concentration if necessary.

A.3.3 O-Phthaldialdehyde (OPT) solution, 0.1 % (w/v). Dissolve 100 mg OPT in 100 ml distilled-in- glass methanol. Store in amber bottle in refrigerator. Prepare fresh weekly.

A.3.4 Histamine standard solutions. Store in refrigerator.

A.3.4.1 Stock solution, 1 mg/ml as free base. Accurately weigh ca 169.1 mg histamine 2 HCl (98 %) into 100 ml volumetric flask, and dissolve and dilute to volume with 0.1M HCl. Prepare fresh weekly.

A.3.4.2 Intermediate solution, 10 μ g/ml. Pipet 1 ml stock solution into 100 ml volumetric flask, and dilute to volume with 0.1M HCl. Prepare fresh weekly.

A.3.4.3 Working solutions, 0.5, 1.0, and 1.5 μ g/5 ml. Pipet 1, 2, and 3 ml intermediate solution into separate 100 ml volumetric flasks, and dilute each to volume with 0.1 M HCl. Prepare fresh daily.

A.3.5 Methanol, 75 % (v/v). Place 75 ml MeOH (distilled in glass) into 100 ml volumetric flask or stoppered graduated cylinder. Dilute to volume with H_2O . Swirl flask while adding H_2O .

A.4 PREPARATION OF STANDARD CURVE

Pipet duplicate 5 ml aliquots of each working standard solution into separate 50 ml glass or polypropylene Erlenmeyers. Pipet in 10 ml 0.1 M HCl to each flask and mix. Pipet in 3 ml 1 M NaOH and mix. Within 5 min,

pipet in 1 mI OPT solution and mix immediately. After exactly 4 min, pipet in 3 mI $3.57 \text{ NH}_3\text{PO}_4$ and mix immediately. It is important to mix thoroughly after each addition and at least once during OPT reaction. (Run 6 – 10 OPT reactions simultaneously by adding reagents to Erlenmeyers in set order.) Prepare blank by substituting 5 ml 0.1 M HCl for histamine solution. Within 1.5 h, record fluorescence intensity (I) of working standard solutions with H₂O in reference cell, using excitation wavelength of 350 nm and emission wavelength of 444 nm. Plot I (corrected for blank) against μ g histamine/5 ml aliquot.

A.5 DETERMINATION

A.5.1 Extract prepared sample with 75 % (v/v) methanol. Pass 4 - 5 ml H₂O through column, **A.2.1** and discard eluate. Pipet 1 ml extract onto column and add 4 - 5 ml H₂O. Immediately initiate column flow into 50 ml volumetric flask containing 5.00 ml 1.00M HCI. When liquid level is ca 2 mm above resin, add ca 5 ml H₂O and let elute. Follow with H₂O in larger portions until ca 35 ml has eluted. Stop column flow, dilute to volume with H₂O, stopper, and mix. Refrigerate eluate.

A.5.2 Pipet 5 ml eluate into 50 ml Erlenmeyer, and pipet in 10 ml 0.1 M HCl. Proceed as in **A.4**, beginning "Pipet in 3 ml 1 M NaOH...."

A.5.3 If test sample contains >15 mg histamine/100 g fish, pipet 1 ml sample–OPT mixture into 10 ml beaker containing exactly 2 ml blank–OPT mixture, and mix thoroughly. Read fluorescence of new solution. Dilute and mix aliquots with blank–OPT mixture as needed to obtain measurable reading. This approximation indicates proper dilution of eluate required prior to second OPT reaction needed for reliable quantitation of test sample. Alternatively, use sensitivity range control of fluorometer (if instrument has one) to estimate dilution. Use these approximations to prepare appropriate dilution of aliquot of eluate with 0.1NHCI, and proceed as in A.4, beginning "Pipet in 3 ml 1 M NaOH"

A.6 CALCULATIONS

A.6.1 Plot of I (measured by meter deflection or recorder response and corrected for blank) against μ g histamine/5 ml test solution should be straight line passing through origin with slope = m = [(la /1.5) + lb + 2lc]/3. mg Histamine/100 g fish = (10)(F)(1/m)(ls)

 μ g Histamine/g fish = 10(mg histamine/100 g fish)

where Is, Ia, Ib, and Ic = fluorescence from test sample, 1.5, 1.0, and 0.5 μ g histamine standards, respectively; and F = dilution factor = (ml eluate + ml 0.1M HCl)/ml eluate. F = 1 for undiluted eluate.

A.6.2 If calibration plot is not linear, use standard curve directly for quantitation. Each subdivision on abscissa should be $\leq 0.1 \ \mu g$ histamine/5 ml test solution. Read all values from curve to nearest 0.05 μg histamine/5 ml test solution.

mg Histamine/100 g fish = (10)(F)(W)

 μ g Histamine/g fish = 10× (mg histamine/100 g fish)

where $W = \mu g$ histamine/5 ml test solution as determined from standard curve.

THE MALAWI BUREAU OF STANDARDS

The Malawi Bureau of Standards is the standardizing body in Malawi under the aegis of the Ministry of Industry and Trade. Set up in 1972 by the Malawi Bureau of Standards Act (Cap: 51:02), the Bureau is a parastatal body whose activities aim at formulating and promoting the general adoption of standards relating to structures, commodities, materials, practices, operations and from time to time revise, alter and amend the same to incorporate advanced technology.

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