



**AFDC13 (490) P3**

**ICS 67.160.10**

## **DRAFT TANZANIA STANDARD**

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**Craft beer – Specification**

FOR STAKEHOLDERS' COMMENTS ONLY

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**TANZANIA BUREAU OF STANDARDS**

## Craft beer — Specification

### 0 Foreword

Beer is a common commercial alcoholic beverage in the country. The beer industry is expanding very fast to cater for the increasing demand of the commodity. This Tanzania standard was prepared in order to ensure that beer consumers get safe and good quality product.

In the preparation of this Tanzania Standard, assistance was drawn from TZS 56:2015 published by Tanzania Bureau of Standards.

In reporting the result of a test or analysis made in accordance with this Tanzania standard, if the final value observed of the calculated is to be rounded off it shall be done in accordance with TZS 4 (see clause 2)

### 1 Scope

This Tanzania standard specifies requirements, methods of sampling and testing for craft beer.

### 2 Normative references

The following referenced documents are indispensable for the application of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

*TZS 4, Rounding of numbers*

*CODEX STAN 192, Codex general standard for food additives*

*TZS 789/EAS 12, Drinking (potable) water — Specification*

*TZS 538/EAS 38, Labelling of pre-packaged foods — General requirements*

*TZS 109: 1981 (1st Ed) - Food processing units – Code of hygiene*

*TZS 268/EAS 100, Food stuffs — Methods of determination of Lead*

*TZS 1491/ISO 1842, Fruit and vegetable products — Determination of pH*

*TZS 729/ISO 4832, Microbiology of food and animal feeding stuffs — Horizontal method for the enumeration of coliforms — Colony-count technique*

*TZS 118-1/ISO 4833-1, Microbiology of the food chain — Horizontal method for the enumeration of micro-organisms — Part 1: Colony-count at 30 degrees C — Pour plate technique*

*TZS1502/ISO 6634, Fruits, vegetables and derived products — Determination of arsenic content — Silver diethyldithiocarbamate spectrophotometric method*

*TZS 1492/ISO 17240, Fruit and vegetable products — Determination of tin content — Method using flame Atomic Absorption Spectrometry*

*TZS 799/ISO 16050:2003 Foodstuffs -- Determination of aflatoxin B1, and the total content of aflatoxins B1, B2, G1 and G2 in cereals, nuts and derived products -- High-performance liquid chromatographic method*

*TZS 471/EAS 104 Alcoholic beverages — Methods of sampling and testing*

### **3 Terms and definitions**

For the purposes of this standard, the following terms and definitions shall apply.

#### **3.1**

##### **alcohol**

as ethyl alcohol (C<sub>2</sub>H<sub>5</sub>OH)

#### **3.2 beer**

beverage, containing ethyl alcohol prepared by fermentation of sugars derived mainly from malted and/or unmalted cereal grains and/or permitted adjuncts with added hops and hops-derived products

#### **3.3 craft beer**

a beer made in traditional or non-mechanised way by small brewery, typically unfiltered

#### **3.4 malt**

cereal grain (often barley), which has been steeped, allowed to germinate and then dried or kilned to halt further germination

#### **3.5 unmalted cereal grains**

cereal grains that have not undergone the processes in 3.4

#### **3.6 brewing**

process of converting malt and/or unmalted cereal grains and adjuncts into beer

#### **3.7 adjuncts**

any other source of fermentable sugar

NOTE These materials are usually but not exclusively added in the brew house during the process of brewing.

#### **3.8 potable water**

water complying with the requirements of TZS.789

#### **3.9 Hops**

flowers of the family *Humulus Lupulus* used in brewing to add bitterness.

### **4 Requirements**

#### **4.1 General requirements**

##### **4.1.1 Essential ingredients**

The following ingredients shall be used for the production of craft beer:

- a) malted and/ or unmalted grains;
- b) hops and/or products derived from hops; and

- c) potable water complying with TZS 789
- d) Brewers' yeast
- e) Food grade CO<sub>2</sub>

#### 4.1.2 Optional ingredients

These may include adjuncts (see 3.7).

#### 4.1.3 craft beer shall be free from:

- a) any substances injurious to health;
- b) any extraneous matter;
- c) non-nutritive sweetening agents; and
- d) any added alcohol.

#### 4.1.4 classes

Craft beer shall be classified based on ethyl alcohol content given in Table 1

Non-alcoholic craft beer

Low alcohol/light beer

Mild beer any artificial colorants except for those colouring agents prepared from sugar, barley, malt or any cereal grains

Medium beer

Strong beer

## 4.2 Specific quality requirements

Craft beer shall comply with the specific quality requirements specified in Table 1.

**Table 1 — Quality requirements for craft beer**

S/No.	Characteristic	Requirement	Test method
(i)	Ethyl alcohol content, %, v/v: <ul style="list-style-type: none"> <li>Non-alcoholic craft beer</li> <li>Low alcohol/light craft beer</li> <li>Mild craft beer</li> <li>Medium craft beer</li> <li>Strong craft beer</li> </ul>	Less than 0.5 0.5 - 2.4 2.5 - 4.0 4.1 - 5.5 More than 5.50	TZS 471
(ii)	pH	3.0 - 4.8	TZS 1491
(iii)	Carbon dioxide, %, v/v <ul style="list-style-type: none"> <li>Bottled/ canned craft beer.</li> <li>Kegged craft beer</li> </ul>	2.4 - 7.0 1.0 - 3	TZS 471

## 5 Food additives

Food additives may be used in the preparation of craft beer in accordance with CODEX STAN 192.

## 7 Hygiene

### 7.1 Microbiological limits for craft beer

Craft beer shall be manufactured and handled in a hygienic manner in accordance with TZS 109 and shall conform to the microbiological limits stipulated in Table 2.

**Table 2 — Microbiological limits for craft beer**

S/N	Organism		
		Limits	Method of test
1.	Total plate count, cfu/ml, max	10 <sup>4</sup>	TZS 118-1
2	Coliforms cfu/ml	Absent	TZS 729
3	<i>E.coli</i> cfu/ml	Absent	TZS 731

## 8 Contaminants

### 8.1 Pesticide residues

All the raw materials used in the production of craft beer shall comply with the maximum residue limits for pesticides as established by the Codex Alimentarius Commission.

### 8.2 Heavy metal

Craft beer shall not contain heavy metals at levels exceeding the limits indicated in Table 3.

**Table 3 — Limits for heavy metal contaminants in craft beer**

S/N o.	Type of heavy metal	Limit	Test method
(i)	Arsenic (as As), mg/L, max.	0.01	TZS 1502
(ii)	Lead (as Pb), mg/L, max.	0.01	TZS 268
(iii)	Tin, (as Sn), mg/L, max.	150	TZS 1492

### **8.3 Mycotoxins limits**

8.3.1 Total aflatoxin in craft beer shall not exceed 10 ppb and 5 ppb for aflatoxin B1 when tested in accordance with TZS 779

8.3.2 Total fumonisins in craft beer shall not exceed total of 2 ppm when tested in accordance with Annex A

## **9 Sampling and testing**

Sampling and testing of craft beer shall be done in accordance with TZS 471

## **10 Packing, marking and labelling**

**10.1 Craft beer** shall be packed in suitable food grade containers.

### **10.2 Marking and Labelling**

In addition to the requirements of TZS 538 the following specific labelling requirements shall apply and shall be legibly and indelibly marked:

- a) name of the product shall be "craft beer",
- b) name, physical location and address of manufacturer;
- c) ethyl alcohol content, % by volume;
- d) list of ingredients in descending order of proportion by mass;
- e) net content
- f) a declaration by common name of any additives used;
- g) date of manufacture, batch identification number/code;
- h) best before date
- i) storage conditions
- j) country of origin; and
- k) statutory warnings.



## ANNEX A:

### Determination of fumonisins B1 and B2 in corn/sample by liquid chromatography with immune-affinity column cleanup

**Applicability:** It applicable to o determination of fumonisins B<sub>1</sub> in corn/sample at total levels from 0.5 to 2µg/g

#### Caution

Fumonisins are nephrotoxic, hepatotoxic, and carcinogenic to rats and mice, however effects on humans are not fully known. Wear protective gloves to reduce skin contact with extracts. Laboratory spills should be cleaned up by washing with a 5% dilution of commercial bleach (sodium hypochlorite) followed by water.

#### 1. Principle

Fumonisins are extracted from corn with methanol–acetonitrile–water (25 + 25 + 50, v/v/v), the filtered extract is cleaned up by an immunoaffinity column, and the fumonisins are eluted with methanol. The eluate is evaporated just to dryness, and the residue is dissolved in acetonitrile–water (50 + 50, v/v). O-Phthaldialdehyde and 2-mercaptoethanol is added to form fluorescent fumonisin derivatives, which are separated by reversed phase liquid chromatography (LC) with fluorescence detection.

#### 2. Reagents

- a) Methanol. —LC grade.
- b) Acetonitrile. —LC grade.
- c) O-Phthaldialdehyde (OPA). —CAS 643-79-8.
- d) 2-Mercaptoethanol (MCE) —CAS 60-24-2.
- e) Sodium dihydrogen phosphate solution. —0.1M. Dissolve 15.6 g NaH<sub>2</sub>PO<sub>4</sub>·2H<sub>2</sub>O in water and dilute to 1 L.
- f) Sodium tetraborate solution.—0.1M. Dissolve 3.8 g Na<sub>2</sub>B<sub>4</sub>O<sub>7</sub>·10H<sub>2</sub>O in water and dilute to 100 mL
- g) Hydrochloric acid.—2M. Dilute HCl (12M) 1+5 with water.
- h) Extraction solvent.—Acetonitrile–methanol–water (25 + 25+ 50, v/v/v).
- i) Acetonitrile–water.—50 + 50, v/v.
- j) Phosphate-buffered saline (PBS).—Dissolve 8.0g NaCl, 1.2g anhydrous Na<sub>2</sub>HPO<sub>4</sub>, 0.2g KH<sub>2</sub>PO<sub>4</sub>, and 0.2g KCl in approximately 990 mL water. Adjust pH to 7.0 with 2M HCl, and dilute to 1 L. Phosphate-buffered saline tablets can also be used.
- k) Immunoaffinity columns.—Specific for fumonisin cleanup with 100% cross reactivity for both FB1 and FB2. The column must have a total capacity of \$ 10 µg fumonisins B1 and B2 and should give a recovery of \$90% when



a calibrant solution of fumonisins B1 and B2 in methanol–PBS containing 5µg fumonisins is applied. Follow the manufacturer's instructions for the type of column used.

- l) LC mobile phase.—Methanol–0.1M NaH<sub>2</sub>PO<sub>4</sub> (77 + 23, v/v), adjusted to pH 3.35 with H<sub>3</sub>PO<sub>4</sub>. Filter mobile phase through 0.45µm membrane, and pump at 1 mL/min flow rate. Adjust com- position to conform with individual LC column characteristics.
- m) OPA reagent.—Dissolve 40 mg OPA in 1 mL methanol, and dilute with 5 mL 0.1M Na<sub>2</sub>B<sub>4</sub>O<sub>7</sub> solution. Add 50µL MCE and mix. Store in the dark for up to 1 week at room temperature in a capped amber vial.
- n) Fumonisins B1 and B2.—Crystalline form, purity of >95%
- o) Fumoinisin stock solution for LC. —Prepare calibrant solution containing fumonisins B1 and B2 in acetonitrile–water (50 + 50, v/v) at concentration of 100µg/mL for FB1 and 50µg/mL for FB2. Fumoinisin calibrant solution is stable up to 6 months when stored at 4°C. Pipet 500µL fumoinisin calibrant solution into 5 mL calibrated volumetric flask. Dilute to volume with acetonitrile–water (50 + 50,v/v), and shake well to obtain stock solution containing FB1 at 10 ng/µL and FB2 at 5 ng/µL
- p) Fumoinisin working calibrant solutions for LC.—Prepare 4 LC calibrant solutions in separate 5mL volumetric flasks according to Table 2001.04C. Dilute contents of each flask to volume (5 mL) with acetonitrile–water (50 + 50, v/v).

### 3. Apparatus

- a) Centrifuge bottle.—Plastic, 250 mL, with screw cap.
- b) Centrifuge—Operating up to 2500 ×g
- c) Filter papers.—Whatman No. 4, 12 cm.
- d) Glass microfiber filters. —Whatman GF/A, 9 cm.
- e) Reservoir.—25 mL with Luer tip connector for immunoaffinity column.
- f) Calibrated microliter syringe or microliter pipet–25–1000 µL
- g) Vacuum manifold.—To accommodate immunoaffinity columns.
- h) Liquid chromatograph.—LC pump delivering 1 mL/min constant flow rate and with injection system calibrated to deliver 20µL; and data system.
- i) LC column. —Stainless steel (150×4.6 mm id), packed with 5µm C18 deactivated reversed-phase material, preceded by corresponding reversed-phase guard column or guard filter (0.5µm porosity.
- j) Fluorescence detector.—Fitted with flow cell and set at 335 nm (excitation) and 440 nm (emission).

### 4. Extraction

Permit materials to reach room temperature before removing test portion. Weigh, to nearest 0.1g, 20g test portion of corn into 250 mL centrifuge bottle, and add 50 mL extraction sol- vent, 2(h). Cover centrifuge bottle, and shake bottle for 20 min with orbital shaker. Centrifuge for 10 min at 2500×g, and filter supernatant through filter paper,3 (c), avoiding transfer of solid ma-terial on filter. Again

extract remaining solid material by adding 50 mL extraction solvent, 2 (h), to centrifuge bottle and shaking bottle for 20 min. Centrifuge for 10 min at 2500  $\times$ g, and filter extract through the same filter paper. Collect and combine the 2 filtrates, and pipet 10 mL filtrate into 100 mL flask. Add 40 mL PBS, 2 (j), and mixwell. Filter diluted extract through microfiber filter, C(d), and collect 10 mL filtrate (equivalent to 0.4 g test portion) for cleanup through immune-affinity column.

## **5. Immunoaffinity Column Cleanup**

Follow manufacturer's instructions for the type of column used. Remove top cap from column, and connect column with reservoir. Remove end cap from column and attach column to vacuum manifold. Pipet 10 mL filtrate into reservoir. Let filtrate flow through column at ca 1–2 drops/s and discard eluate. Wash column with 10 mL PBS, 2(j), at rate of 1–2 drops/s until air comes through column. Place 4 mL vial under column. Elute fumonisins with 1.5 mL LC grade methanol at 1 drop/s, and collect fumonisins in vial. Evaporate eluate just to dryness under stream of N at ca 60°C. Retain dried residue at ca 4°C for derivatization and LC analysis

## **6. Calibration Curve**

Prepare calibration curves, using working calibrant solutions 2 (p). These solutions cover the range of 0.025–2.000  $\mu$ g/g for FB1 and the range of 0.0125–1.000  $\mu$ g/g for FB2. Prepare calibration curves, before LC analysis, according to Table 2001.04C, and check plots for linearity. If curve is not linear, repeat derivatization following instructions carefully and/or reduce the range of the calibrants.

## **7. Derivatization and LC Analysis**

Redissolve purified residue in 200  $\mu$ L acetonitrile–water (50 + 50, v/v), B(i). Transfer 50  $\mu$ L aliquots of extract or standards to bottom of 1 mL test tube, and add 50  $\mu$ L OPA reagent, B(m). Mix solution for 30 s with vortex mixer, and inject 20  $\mu$ L derivatized solution (equivalent to 20 mg matrix) into LC system exactly 3 min after add-ing OPA reagent. With the described LC mobile phase, B(l), and column, C(i), satisfactory (baseline) resolution of FB1–OPA and FB2–OPA must be obtained, with expected retention times at ca 6 and 15 min, respectively. If fumonisin content of derivatized extract is higher than calibration range, dilute purified extract with acetonitrile–water (50 + 50, v/v), B(i), derivatize with OPA reagent, and repeat LC analysis.

## 8. Quantification of Fumonisin B1 and B2

Quantify FB1 and FB2 by measuring peak area (or peak height) at retention time of each fumonisin and comparing measured value with corresponding calibration curve.

From calibration curves determine amounts of FB1 and FB2 (in  $\mu\text{g}$ ) in aliquot of test solution injected into LC column.

Separately calculate concentrations (CFB) of FB1 and FB2 in micrograms per gram ( $\mu\text{g/g}$ ), as follows:

$$\text{CFB} = 10^3 \frac{M_A}{10^3 M_B} \times \frac{1}{20} \frac{M_A}{20}$$

where

$M_A$  is mass of FB1 or FB2 (in  $\mu\text{g}$ ) in aliquot of test solution injected on column, as determined from calibration curve,

$M_B$  is mass of matrix (in mg) injected on column (20 mg), obtained as follows:

$$\frac{20 \text{ g (Extraction step)}}{100 \text{ mL}} \times \frac{10 \text{ mL (dilution)}}{50 \text{ mL}} \times \frac{10 \text{ mL (purified aliquot)}}{0.2 \text{ mL}} \times \frac{0.05 \text{ mL (derivatization)}}{0.1 \text{ mL}}$$

0.02 mL (injected aliquot)

And  $10^{-3}$  is a factor to convert  $M_A$  and  $M_B$  from ng to  $\mu\text{g}$  and mg to g, respectively.