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Aspartame (food grade) — Specification

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Foreword

Uganda National Bureau of Standards (UNBS) is a parastatal under the Ministry of Trade, Industry and Cooperatives established under Cap 327, of the Laws of Uganda, as amended. UNBS is mandated to co-ordinate the elaboration of standards and is

(a) a member of International Organisation for Standardisation (ISO) and

(b) a contact point for the WHO/FAO Codex Alimentarius Commission on Food Standards, and

(c) the National Enquiry Point on TBT Agreement of the World Trade Organisation (WTO).

The work of preparing Uganda Standards is carried out through Technical Committees. A Technical Committee is established to deliberate on standards in a given field or area and consists of key stakeholders including government, academia, consumer groups, private sector and other interested parties.

Draft Uganda Standards adopted by the Technical Committee are widely circulated to stakeholders and the general public for comments. The committee reviews the comments before recommending the draft standards for approval and declaration as Uganda Standards by the National Standards Council.

The committee responsible for this document is UNBS/TC 2 [*Food and Agriculture standards*], Subcommittee SC 6, [*Food Additives and contaminants*].

Introduction

Aspartame (3-Amino-N-(α-carbomethoxy-phenethyl)-succinamic acid, N-L-α-aspartyl-L-phenylalanine-1-methyl ester, $C_{14}H_{18}N_2O_5$) is a low calorie artificial sweetener, sugar substitute and flavour enhancer. It is 100 – 200 times sweeter than sucrose. It is one of the most popular artificial sweeteners and it is widely used in the preparation of beverages, desserts, sweets, dairy products, chewing gums, energy-reduced and weight control products, as a table-top sweetener and in the preparation of food for diabetics.

Aspartame is made of two naturally occurring amino acids, phenylalanine and aspartic acid. The phenylalanine in aspartame is slightly modified by adding a methyl group giving the product its sweet taste. Just like proteins, aspartame is digested once it reaches the intestines. It is fully broken down to aspartic acid and phenylalanine, which are absorbed into the body. In addition, the methyl group from the modified phenylalanine is released in the gut to form methanol. The methanol is absorbed and most of it used to produce energy.

The use of aspartame as a sugar replacer remains a controversial topic with some scientific studies validating its safety while others suggest it could have effects on the consumer.

In the Codex Alimentarius Commission International Numbering System, aspartame is assigned as INS 951 and the FAO/WHO Joint Experts Committee on Food Additives (JECFA) established the Acceptable Daily Intake (ADI) for aspartame at 0 – 40 mg/kg body weight.

This standard has been developed to ensure that that the usage of aspartame in food products conforms to acceptable limits.

Aspartame (food grade) — Specification

1 Scope

This Draft Uganda Standard specifies requirements, sampling and test methods for food grade aspartame.

2 Normative references

The following referenced documents are referred to in the text in such a way that some or all their content constitutes requirements 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.

AOAC 999.11, *Determination of Lead, Cadmium, Copper, Iron, and Zinc in Foods, Atomic Absorption Spectrophotometry after Dry Ashing*

US 277, *General standard for the labelling of food additives when sold as such*

US CAC/GL 50, *General guidelines on sampling*

FDUS ISO 760, *Determination of water — Karl Fischer Method (General method)*

3 Terms and definitions

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

ISO and IEC maintain terminological databases for use in standardization at the following address:

— ISO Online browsing platform: available at http://www.iso.org/obp

food grade material
material, made of substances which are safe and suitable for their intended use and which will not impart any toxic substance or undesirable odour or flavour to the product

4 Requirements

4.1 General requirements

Food grade aspartame shall be:

- a white, odourless, crystalline powder with a strong sweet aroma; and
- slightly soluble in water and ethanol/methanol;

A saturated aqueous solution of the product shall be acidic.

4.2 Specific requirements

Food grade aspartame shall comply with the specific requirements given in Table 1 when tested in accordance with the test methods specified therein.

Table 1 — Specific requirements for food grade aspartame

S/N	Characteristic	Requirement	Test method
i)	Purity as $C_{14}H_{18}N_2O_5$, % m/m (dry basis)	98.0 – 102.0	Annex A
ii)	Moisture %m/m, max.	4.3	FDUS ISO 760
iii)	Sulphated ash, % m/m (dry basis), max.	0.2	Annex B
iv)	pH (0.8% solution)	4.5 – 6.0	Annex C
v)	5-Benzyl–3,6–dioxo–2–piperazineacetic acid, (diketopiperazine), %m/m, max.	1.5	Annex D
vi)	Test for amine group	To pass test	Annex E
Vii)	Test for ester group	To pass test	Annex F

5 Contaminants

Food grade aspartame shall comply with the Maximum Levels of contaminants given in Table 2 when tested in accordance with the test methods specified therein.

Table 2 — Maximum Limits for contaminants in food grade aspartame

S/N	Contaminant	Maximum level	Test method
i)	Heavy metals (as Pb), mg/kg, max.	1	AOAC 999.11
ii)	Arsenic (as As), mg/kg, max.	3	AOAC 952.13

6 Packaging

The product shall be securely packaged in containers made of food grade materials conforming to US 1659. The packages shall preserve the quality of the product, prevent entry of light and preclude contamination from the external environment.

7 Weights and Measures

The weight of the product when packaged shall comply with the Weights and Measures Regulations.

8 Labelling

In addition to the requirements of US 277, the product label shall be legibly and indelibly labelled with the following:

i) Name of the product as Aspartame with the words ‘Food Grade’;

ii) Name and physical address of the manufacturer/distributor;

iii) Net weight in metric units;

iv) Batch/lot number;

v) Directions for storage;

vi) Date of manufacture; and the

vii) Expiry date.

9 Sampling

Representative samples of the product shall be drawn in accordance with US CAC/GL 50.

Annex A
(normative)

Assay: Test for purity

A.1 Reagents

A.1.1 Dimethylformamide

A.1.2 Thymol Blue

A.1.3 Lithium Methoxide/Sodium Methoxide

A.2 Procedure

Accurately weigh 150 mg of the sample, previously dried at 105 °C for 4 hours. Dissolve in 35 ml of dimethylformamide, add 5 drops of thymol blue, and titrate with a micro burette to a dark blue end-point with 0.1N lithium methoxide or sodium methoxide. Perform a blank determination and make any necessary correction. Each ml of 0.1N lithium methoxide/sodium methoxide is equivalent to 29.43 mg of $C_{14}H_{18}N_2O_5$.

Caution: Protect the solution from absorption of carbon dioxide and moisture by covering the titration vessel with aluminium foil while dissolving the sample and during the titration.

Annex B
(normative)

Determination of sulphated ash

B.1 Reagents

B.1.1 Dilute Sulphric acid – 10 percent (m/v).

B.2 Procedure

Transfer about 2 g of the sample, accurately weighed, to a tared 50-ml to 100-ml platinum dish or other suitable container and add sufficient dilute sulphuric acid to moisten the entire sample. Heat gently, until the sample is dry and thoroughly charred, then continue heating until all the sample has been volatilized or nearly all of the carbon has been oxidized. Cool, moisten the residue with 0.1 ml of sulphuric acid, and heat in the same manner until the remainder of the sample and any excess sulphuric acid have been volatilized. Finally ignite in a muffle furnace at 800 °C ± 25 °C for 15 minutes. Cool in a desiccator and weigh.

B.3 Calculation

Sulphated ash, percent by mass = $\frac{M_1}{M} X100$

where

M_1 is the mass, in grams, of residue after igniting, and

M is the mass, in grams, of the sample tested.

Annex C
(normative)

Determination of pH

C.1 Procedure

Dissolve 1 g of sample in 125 ml of distilled water and measure the measure the pH of the solution using a calibrated pH meter.

Annex D
(normative)

Test for 5-Benzyl–3,6–dioxo–2–piperazineacetic acid

D.1 Apparatus

D.1.1 Gas Chromatograph of a suitable type, equipped with a hydrogen flame ionization detector and designed for handling glass columns with on-column injection (Micro-Tek 220 or equivalent), containing a 1.83 m x 4 mm (inside diameter) glass column packed with 3 percent OV-1 on 80/100-mesh supelcoport. Condition the column overnight at 250 °C before readjustment and equilibration to the operating condition. To preclude build-up of silicon oxide, clean the detector with acetone frequently.

D.1.1.1 Operating conditions: The operating parameters may vary depending on the particular instrument used, but a suitable chromatogram may be obtained using the following conditions:

a) Column temperature 200 °C

b) Inlet temperature 200 °C

c) Detector temperature 275 °C

d) Carrier gas – Nitrogen, flowing at a rate of 75 ml per minute

e) Hydrogen and air flow to burner – optimised to give maximum sensitivity

f) Recorder – 1 mV full scale

D.2 Reagents

D.2.1 Silation reagent: Just before use, dilute 3 parts, by volume, of N, O-bis-(trimethylsilyl) acetamide with 2 parts of dimethylformamide.

D.2.2 Standard preparation: Transfer about 25 mg of 5-Benzyl-3, 6-dioxo-2-piperazineacetic acid Reference Standard, accurately weighed, into a 50-ml volumetric flask, dissolve in methanol, dilute to volume with methanol, and mix. Pipet 10 ml of this solution into a second 100-ml volumetric flask, dilute to volume with methanol, and mix. Pipet 3 ml of the second solution into a 2-dram vial, with Teflon-lined cap, and evaporate to dryness on a steam bath. Add 1 ml of the Silylation reagent to the residue, cap the vial tightly, shake and heat in an oven at 80 °C for 30 minutes. Remove the vial from the oven, shake for 15 seconds, and cool to room temperature.

D.2.3 Sample preparation: Transfer about 10 mg of the sample, accurately weighed, into a 2-dram vial, with Teflon-lined cap, add 1 ml of the Silylation reagent, cap tightly, shake, and heat in an oven at 80 °C for 30 minutes. Remove the vial from the oven, shake for 15 seconds, and cool to room temperature.

D.3 Procedure

Inject a 3 µl portion of the standard preparation into the gas chromatograph and obtain the chromatogram. Measure the height of the peak produced by the 5-benzyl-3, 6-dioxo-2-piperazineacetic acid, and record it as *P*. Under the stated conditions, the elution time is about 7-9 min. Similarly, inject a 3 µl portion of the sample preparation, obtain the chromatogram and measure the height of the peak produced by any 5-benzyl-3,6-dioxo-2-piperazineacetic acid contained in the sample, and record it as *p*.

D.4 Calculation

The percentage of 5-Benzyl–3, 6–dioxo–2–piperazineacetic acid in the sample can be computed as

$$\frac{3 \times M \times p}{500 \times m \times P}$$

where,

M is the mass, in milligrams, of the reference standard taken

m is the mass in milligrams, of aspartame analysed

p is the height of peak produced by 5-benzyl-3, 6-dioxo-2-piperazineacetic acid contained in sample; and

P height of peak produced by 5-benzyl-3, 6-dioxo-2-piperazineacetic acid contained in standard.

Annex E
(normative)

Test for amine group

E.1 Procedure

Dissolve 2 g of ninhydrin in 75 ml of dimethylsulfoxide, add 62 mg of hydrindantin, dilute to 100 ml with 4M lithium acetate buffer solution (pH 9), and filter. Transfer about 10 mg of the sample to a test tube, add 2 ml of the reagent solution, and heat. A dark purple colour is formed.

Annex F
(normative)

Test for ester group

F.1 Reagents

F.1.1 Methanol

F.1.2 Methanol saturated with hydroxylamine hydrochloride

F.1.3 5N potassium hydroxide

F.1.4 Hydrochloric acid

F.1.5 Ferric chloride

F.2 Procedure

Dissolve about 20 mg of sample in 1 ml of methanol, add 0.5 ml of methanol saturated with hydroxylamine hydrochloride, mix, and then add 0.3 ml of 5N potassium hydroxide in methanol. Heat the mixture to boiling, then cool, adjust the pH to between 1 and 1.5 with hydrochloric acid, and add 0.1 ml of ferric chloride. A burgundy/maroon colour is produced.

Bibliography

[1] IS 13657: 1993 (Reaffirmed in 2003), Aspartame, food grade — Specification

[2] JECFA monograph, Aspartame

[3] Food chemicals codex (5th Edition)

[4] EFSA Journal (2013) – Scientific opinion on aspartame

Certification marking

Products that conform to Uganda standards may be marked with Uganda National Bureau of Standards (UNBS) Certification Mark shown in the figure below.

The use of the UNBS Certification Mark is governed by the Standards Act, and the Regulations made thereunder. This mark can be used only by those licensed under the certification mark scheme operated by the Uganda National Bureau of Standards and in conjunction with the relevant Uganda Standard. The presence of this mark on a product or in relation to a product is an assurance that the goods comply with the requirements of that standard under a system of supervision, control and testing in accordance with the certification mark scheme of the Uganda National Bureau of Standards. UNBS marked products are continually checked by UNBS for conformity to that standard.

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