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भारतीय मानक मसौदा

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(*आइ एस 5343 का दूसरा पुनरीक्षण*)

Draft Indian Standard

BUTYLATED HYDROXYANISOLE, FOOD GRADE - SPECIFICATION

(*Second Revision of IS 5343*)

ICS No. 67.220.20

Food Additives Committee, FAD 08 **Last Date of Comments: 20 June 2024**

FOREWORD

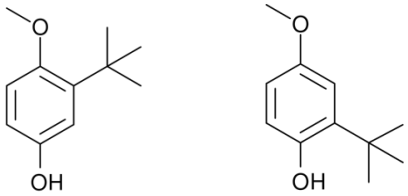
(*Formal clauses would be added later*)

Food additives are added to improve the appearance, flavour, texture or storage properties, etc of the processed foods. As certain impurities in these substances have been found to be harmful, it is necessary to have a strict quality control of these food additives. A series of standards have, therefore, been prepared to cover purity and-identification of these substances. These standards would help in checking purity, which requires to be checked at the stage of manufacture, for it is extremely difficult to detect the impurity once these substances have been added to the processed foods. Besides, these standards are intended to guide the indigenous manufacturers in making their product conform to specifications that are accepted by scientists, health authorities and national/ international bodies.

Butylated hydroxyanisole (BHA), food grade used as an antioxidant. BHA is permitted for use in foods under *Food Safety and Standards* (*Food Products Standards and Food Additives*) *Regulations*, 2011.

Chemical Names and Formulae:

A mixture of 3-and 2-tertiary butyl-4-hydroxyamsole; a mixture of 3- and 2-tertiary butyl-4-methoxyphenol. Its empirical formula is $C_{11}H_{16}O_2$ and molecular weight is 180.24. Structural formula of BHA is:



2-isomer 3-isomer

This standard was first published in 1969. In the preparation of this standard, assistance was derived from Food Chemical Codex (FCC), Third Edition, National Academy of Sciences, National Research Council, Washington DC and specification of BHA by FAO & WHO, 1962.

It was first revised in 1996 to provide additional requirements for 3-tertiary-butyl 4-hydroxyanisole, phenolic impurities and specific absorption and their test methods and to provide directions for storage and expiry date under the marking clause.

In this revision, the following major changes have been made:

a) The requirement for heavy metals has been removed as the limit of lead (contaminant in food colours) is already covered through the standard.
b) The marking requirements have been updated.

For the purpose of deciding whether a particular requirement of this standard is complied with, the final value, observed or calculated, expressing the result of a test or analysis, shall be rounded off in accordance with IS 2: 2022 ‘Rules for rounding off numerical values (*second revision*)’. The number of significant places retained in the rounded off value should be the same as that of the specified value in this standard.

1 SCOPE

This standard prescribes the requirements and the methods of test for butylated hydroxyanisole (BHA), food grade.

2 REFERENCES

The following Indian Standards contain provisions which through reference in this text, constitute provision of this standard. At the time of publication, the editions indicated were valid. All standards are subject to revision and parties to agreements based on this standard are encouraged to investigate the possibility of applying the most recent editions of the standards indicated below:

IS No.	*Title*
IS 1070 : 2023	Reagent grade water − Specification (*fourth revision*)
IS 1699 : 202X	Methods of sampling and test for food colours (*third revision*) [*Under preparation Doc: FAD 08 (23204)F*]

3 DESCRIPTION

BHA is a mixture of 3 and 2-isomer. It is a white or slightly yellow waxy crystalline solid with an aromatic odour. The material is insoluble in water, freely soluble in ethanol and propylene glycol.

NOTE - The solubility is intended only as information regarding approximate solubility and is not to be considered as a quality requirement and is of minor significance as a means of identification or determination of purity.

4 REQUIREMENTS

4.1 Identification

4.1.1 When 2 ml of 2.0 percent aqueous borax solution and a few small crystals of 2,6-dichloroquinonechlorimide are added to an ethanolic solution (1 percent by volume) of butylated hydroxyanisole, a blue colour shall appear.

4.1.2 When 2 ml of ferric chloride (0.2 percent $FeC1_3.6H_2O$ in absolute ethanol) and 2 ml of 0.2 percent 2,2'-bipyridine in absolute ethanol are added to 5 ml of 0.5 percent butylated hydroxyanisole in 50 percent ethanol, a red colour shall appear.

4.2 The material shall also conform to the requirements given in Table 1.

5 PACKING AND STORAGE

5.1 Packing

The material shall be filled in amber colour glass containers or any other containers with as little air space as possible. The container shall be such as to preclude contamination of the contents with metals or other impurities.

5.2 Storage

The material shall be stored in a cool and dry place so as to avoid excessive exposure to heat.

Table 1 Requirements for BHA, Food Grade

(*Clause* 4.2)

Sl No.	Characteristic	Requirements	Method of Test, Ref to
(1)	(2)	(3)	(4)
i)	a) Purity as $C_{11}H_{16}O_2$ after drying at 120°C for 4 hours, percent by mass, *Min* b) 3 tertiary butyl 4-hydroxyanisole, percent by mass, *Min*	98.5 85	Annex A Annex A
ii)	Melting point °C	48 – 63	Annex B
iii)	Sulphated ash, percent by mass, *Max*	0.05	Annex C
iv)	Arsenic (as As), mg/kg, *Max*	3	IS 1699
v)	Phenolic impurities, percent by mass, *Max*	0.5	Annex D
vi)	Specific absorption E 1 percent (1 cm cell) in ethanol at a) 290 nm b) 228 nm	 190 *Min* 210 *Max* 326 *Min* 345 *Max*	Annex E
vii)	Lead (as Pb), mg/kg, *Max*	2	IS 1699

6 MARKING

6.1 Each container shall be legibly and indelibly marked with the following information:

a) Name of the material, including the words 'Food Grade';
b) Name of the manufacturer or registered trade-mark, if any;
c) Net quantity when packed;
d) Lot/batch No.;
e) Month and year of manufacture;
f) Expiry date; and
g) Any other requirements as specified under the *Legal Metrology* (*Packaged Commodities*) *Rules*, 2011 and *Food Safety and Standards* (*Labelling and Display*) *Regulations*, 2020.

6.2 BIS Certification Marking

The product(s) conforming to the requirements of this standard may be certified as per the conformity assessment schemes under the provisions of the *Bureau of Indian Standards Act*, 2016 and the Rules and Regulations framed thereunder, and the products may be marked with the Standard Mark.

7 SAMPLING

The representative samples of the material shall be drawn according to the method prescribed in IS 1699.

8 QUALITY OF REAGENTS

Unless specified otherwise, pure chemicals and distilled water (*see* IS 1070) shall be employed in tests.

NOTE- ‘Pure chemicals’ shall mean chemicals that do not contain impurities which affect the results of analysis.

ANNEX A

[*Table* 1, *Sl No.* (i)]

DETERMINATION OF PURITY

A-1 GENERAL

Two methods, that is, infrared and calorimetric methods have been specified. Either method could be used.

A-2 INFRARED METEOD

A-2.1 Procedure

Weigh 1 000 g of butylated hydroxyanisole into a 10 ml volumetric flask, dissolve it in carbon disulphide dilute to the mark with this solvent and mix thoroughly. Fill a 0.15 mm liquid cell with the solution, insert in an infrared spectrometer and measure the spectrum from 10.5 to 12.5 µ using the 1.3 cm rock salt plate in the reference beam, 2x slits and normal scanning speed. Draw a background line on the spectrogram from 11.0 to 12.0 µ. Determine the net absorption of the sample at 11.42 µ by subtracting the background absorption at this wave length from the total absorption of the sample. Refer the net absorption value at a previously prepared standard reference curve to obtain the apparent butylated hydroxyanisole assay.

A-2.1.1 *Preparation of Standard Reference Curve*

Weigh 0.900, 0.950 and 1.000 g of 3-tertiarybutylhydroxyanisole reference standard into three 10 ml volumetric flasks. Dissolve the samples in carbon disulphide, dilute to the mark with this solvent and mix thoroughly. Measure the spectra of these three samples using the same conditions described under **A-2.1**, obtain the net absorption of the three samples at 11.42 µ and ‘plot these values against the percentage of butylated hydroxyanisole. The 0.900, 0.950 and 1.000 g samples represent 90, 95 and 100 percent butylated hydroxyanisole, respectively.

A-2.1.2 *Isomer Ratio Test*

Melt the sample in a water-bath and stir thoroughly. Weigh 1.000 g of the molten sample into a 10 ml volumetric flask. Dilute to the mark with carbon disulphide and shake until the sample is completely dissolved. Measure the infrared spectrum of the solution from 10 to 12 µ using a 0.4 mm cell with a 1.3 cm rock-salt plate in the reference beam. From the percentage transmittance readings at 10.75 and 10.95 µ, calculate the optical density values. Divide the optical density at 10.75 µ by that at 10.95 µ to obtain the optical density ratio. (Exact position of these absorption bands may vary, depending upon the instrument. If a recording instrument is used, the position of minimum transmission on the chart should be taken with a non-recording instrument, the exact length and slit setting should be determined.) Using the optical density ratio value, determine the percentage of 3-tertiarybutylhydroxyanisole in the sample by means of a calibration curve.

A-2.1.2.1 *Preparation of calibration curve*

Weigh (a) 1000 mg of 3-tertiarybutylhydroxyanisole; (b) 900 mg of 3-tertiarybutylhydroxyanisole and 100 mg of 2-tertiarybutylhydroxyanisole; (c) 800 mg of 3-tertiarybutylhydroxyanisole and 200 mg of 2-tertiarybutylhydroxyanisole to an accuracy of ±1

mg into three 10 ml volumetric flasks, dilute to the mark with carbon disulphide and shake until the sample is completely dissolved. Measure the infrared spectra for each of these three mixtures following the same procedure used for the sample. Plot the calculated optical density ratios obtained against the corresponding concentrations of 3-tertiarybutyl hydroxyanisole.

A-2.2 Calculation

Calculate the true butylated hydroxyanisole assay using the following equation:

Butylated hydroxyanisole, percent by mass = Apparent butylated hydroxyanisole assay (**A-2.1** and **A-2.1.1**) + 0.16 [100 - percent of 3-tertiarybutylhydroxyanisole (**A-2.1.2** and **A-2.1.2.1**)].

A-3 COLORIMETRIC METHOD

A-3.1 Reagents

A-3.1.1 *Ethanol* – 80 percent.

A-3.1.2 *Borax* **–** 2.0 percent, aqueous.

A-3.1.3 *2,6-Dichloroquinonechlorimide* **–** 0.0 1 percent.

A-3.2 Procedure

Prepare a solution of pure butylated hydroxyanisole in 80 percent ethanol containing 5.0 µg per millilitre. Place suitable aliquots (1 to 12 ml) of the butylated hydroxyanisole solution into small glass-stoppered bottles to give a range of 5-60 µg per aliquot. Add enough 80 percent ethanol to each bottle to give a total of 12 ml. Then add 2 ml of aqueous borax and 2 ml of 2,6-dichloroquinonechlorimide. Age the samples and the blank for 15 minutes. Using the blank as a reference standard, determine the optical density at 610 nm on a colorimeter or spectrophotometer. Plot the standard curve on regular coordinate paper using optical density versus concentration of butylated hydroxyanisole per aliquot. The points should fall on or near a straight line.

Proceed as above using a sample solution in 80 percent ethanol. From the optical density, find out the purity of butylated hydroxyanisole using the standard curve.

ANNEX B

[*Table* 1, *Sl No.* (ii)]

DETERMINATION OF MELTING POINT

B-1 APPARATUS

B-1.1 Oven or Oil-Bath

Maintained at about 75°C.

B-1.2 Sample Tube

25 mm × 150 mm test-tube closed with a cork stopper having two holes – one at the centre to take thermometer and one at the side to take an agitator.

B-1.3 Agitator

With a paddle formed by bending a piece of stainless steel wire to form a loop surrounding the thermometer.

B-1.4 Thermometer

B-1.5 Air Bath Tube

B-l.6 Water-Bath

Maintained between 55°C and 60°C.

B-2 PROCEDURE

Melt a representative sample by means of an oven or oil bath at about 75°C. Take a sample tube fill it to a depth of about 90 mm. Insert the stopper carrying the thermometer and stirrer, adjusting the thermometer so that the thermometer immersion mark is at the surface. The tip of the bulb should be about 1 cm from the bottom of the tube. Place the sample tube in an air-bath tub, and then place the air-bath tube in water-bath maintained between 55°C and 60°C. Gently stir the molten sample at the rate of about 20 strokes per minute. Record temperature readings at 30 second intervals to 0.1°C. The temperature of the sample will fall gradually at first, rise slightly and become nearly constant for 3 to 5 minutes. If the lowest descending temperature is more than 1.0°C below the average temperature of the plateau, the determination should be repeated using a slightly warmer water-bath. The temperature at which the thermometer reading is constant for 5 consecutive readings is taken as the melting point.

ANNEX C
[*Table* 1, *Sl No.* (iii)]
DETERMINATION OF SULPHATED ASH

C-1 REAGENT

Concentrated Sulphuric Acid

C-2 PROCEDURE

Weigh accurately about 2 g of the material in a tared crucible. Ignite, gently at first, until the material is thoroughly charred, cool, moisten the residue with 1 ml of sulphuric acid and ignite gently till the carbon is completely consumed. Cool the crucible in a desiccator and weigh.

NOTE - Carry out the ignition in a place protected from air currents and use as low a temperature as possible to effect the combustion of carbon.

C-3 CALCULATION

$$\text{Sulphated ash, percent by mass} = \frac{M_1}{M_2} \times 100$$

where,

M_1 = mass in g, of the residue; and

M_2 = mass in g, of the material taken for test.

ANNEX D

[*Table* 1, *Sl No.* (v)]

DETERMINATION OF PHENOLIC IMPURITIES

D-1 PROCEDURE

Phenolic impurities are determined by the method using silica gel G plates.

Solution 1- Dissolve 0.25 g of BHA in 10 ml of ether

Solution 2 - Dilute 1 ml of Solution 1 to 10 ml with ether and then dilute 1 ml of this solution to 20 ml with ether. Use the final dilution as Solution 2.

Spot 2 ml each of Solution 1 and Solution 2 on separate TLC plates and properly identify them. Place them in developing chamber containing chloroform as solvent and allow the solvent to ascend to a point of 15 cm above the sample spots. Develop the chromatograms by spraying a mixture containing 100 ml of 10.5 percent ferric ferrocyanide solution and 25 ml of 5 percent ferric chloride solution. Any blue violet spots appearing on chromatogram 1 (other than the major spot and the spot at *Rf* 0.35) are not more intense than the major spot appearing on chromatogram 2.

ANNEX E

[*Table* 1, *Sl No*. (vi)]

DETERMINATION OF SPECIFIC ABSORPTION

E-1 PROCEDURE

Prepare 1 percent solution of butylated hydroxyanisole in ethanol and find out its specific absorption in a suitable spectrophotometer using 1 cm cell at wavelengths 290 and 228 nm.