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**High fructose syrup — Specifications  
and test methods**

*Sirop à haute teneur en fructose — Spécifications et méthodes d'essai*

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Published in Switzerland

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## Foreword

ISO (the International Organization for Standardization) is a worldwide federation of national standards bodies (ISO member bodies). The work of preparing International Standards is normally carried out through ISO technical committees. Each member body interested in a subject for which a technical committee has been established has the right to be represented on that committee. International organizations, governmental and non-governmental, in liaison with ISO, also take part in the work. ISO collaborates closely with the International Electrotechnical Commission (IEC) on all matters of electrotechnical standardization.

The procedures used to develop this document and those intended for its further maintenance are described in the ISO/IEC Directives, Part 1. In particular, the different approval criteria needed for the different types of ISO documents should be noted. This document was drafted in accordance with the editorial rules of the ISO/IEC Directives, Part 2 (see [www.iso.org/directives](http://www.iso.org/directives)).

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights. Details of any patent rights identified during the development of the document will be in the Introduction and/or on the ISO list of patent declarations received (see [www.iso.org/patents](http://www.iso.org/patents)).

Any trade name used in this document is information given for the convenience of users and does not constitute an endorsement.

For an explanation of the voluntary nature of standards, the meaning of ISO specific terms and expressions related to conformity assessment, as well as information about ISO's adherence to the World Trade Organization (WTO) principles in the Technical Barriers to Trade (TBT), see [www.iso.org/iso/foreword.html](http://www.iso.org/iso/foreword.html).

This document was prepared by Technical Committee ISO/TC 93, *Starch (including derivatives and by-products)*.

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Any feedback or questions on this document should be directed to the user's national standards body. A complete listing of these bodies can be found at [www.iso.org/members.html](http://www.iso.org/members.html).

## Introduction

High fructose syrup (HFS) is a sweetener made from starch. The starch is first broken down into glucose by enzymes and is then further processed by glucose isomerase to convert some of its glucose into fructose. "HFS 42" refers to 42 % and "HFS 55" to 55 % fructose composition, respectively. HFS 42 is mainly used for processed foods and breakfast cereals, whereas HFS 55 is used mostly for the production of soft drinks.

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# High fructose syrup — Specifications and test methods

## 1 Scope

This document specifies the chemical and microbiological requirements for and test methods of high fructose syrup.

## 2 Normative references

The following documents are referred to in the text in such a way that some or all of 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.

ISO 1743, *Glucose syrup — Determination of dry matter content — Refractive index method*

ISO 4833-1, *Microbiology of the food chain — Horizontal method for the enumeration of microorganisms — Part 1: Colony count at 30 °C by the pour plate technique*

ISO 4833-2, *Microbiology of the food chain — Horizontal method for the enumeration of microorganisms — Part 2: Colony count at 30 °C by the surface plating technique*

ISO 5809, *Starches and derived products — Determination of sulphated ash*

ISO 11212-1, *Starch and derived products — Heavy metals content — Part 1: Determination of arsenic content by atomic absorption spectrometry*

ISO 11212-3, *Starch and derived products — Heavy metals content — Part 3: Determination of lead content by atomic absorption spectrometry with electrothermal atomization*

ISO 21527-2, *Microbiology of food and animal feeding stuffs — Horizontal method for the enumeration of yeasts and moulds — Part 2: Colony count technique in products with water activity less than or equal to 0,95*

OFFICIAL METHOD AOAC 999.11: 1999 *Determination of lead, cadmium, copper, iron, and zinc in foods, Atomic absorption spectrophotometry after dry ashing*

## 3 Terms and definitions

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

ISO and IEC maintain terminology databases for use in standardization at the following addresses:

- ISO Online browsing platform: available at <https://www.iso.org/obp>
- IEC Electropedia: available at <https://www.electropedia.org/>

### 3.1

#### **brix**

percentage of soluble solids per sucrose of an aqueous solution

### 3.2

#### **electrical conductivity**

passage of electrical current through an electrolyte solution by means of free ions present in that solution

**3.3 fructose percentage**

amount of fructose present in the solution, in terms of dry matter

**3.4 high fructose syrup**

sweet and clear condensed liquid obtained by starch hydrolysis, and isomerization partly of glucose to fructose

**3.5 high fructose syrup HFS 42**

liquid containing no less than 97 % of total saccharides, expressed as a percentage of total solids, of which 92 % consists of monosaccharides

**3.6 high fructose syrup 550 g/kg HFS 55**

liquid containing no less than 95 % of total saccharides, expressed as a percentage of total solids, of which 94 % consists of monosaccharides

**4 Requirements**

**4.1 Physical indices**

Physical indices shall comply with the requirements given in [Table 1](#).

**Table 1 — Physical indices of high fructose syrup**

Item	Limit
Appearance	Transparent
Colour	Max 20 International Unit
Odour and taste	No off odour and foreign taste
Foreign material	Free of any foreign matter

**4.2 Chemical indices**

Chemical indices shall comply with the requirements given in [Table 2](#).

NOTE [Subclauses 5.3, 5.4, 5.5](#) and [5.6](#) incorporate the ISBT Manual<sup>[2]</sup> with modifications.

**Table 2 — Chemical indices of high fructose syrup**

Item	Limit		Test method
	42	55	
Brix	Min 69	Min 74	ISO 1743
pH	3,3 to 4,5	3,3 to 4,5	<a href="#">Subclause 5.1</a>
Sulfate ash, weight percent dry matter	Max 0,1	Max 0,1	ISO 5809
Sulfur dioxide, mg/kg	Max 3	Max 3	ISBT method <sup>[2]</sup>
Fructose percentage, dry weight percentage	Min 42	Min 55	<a href="#">Subclause 5.3</a>
Glucose percentage, dry weight percentage	Max 52	Max 42	<a href="#">Subclause 5.3</a>
Hydroxymethylfurfural, mg/kg	Max 75	Max 75	<a href="#">Subclause 5.4</a>
Electrical conductivity (μS/cm)	Max 70	Max 70	<a href="#">Subclause 5.5</a>



### 4.3 Contaminant limits

Contaminant limits shall comply with the requirements given in [Table 3](#).

**Table 3 — Contaminant limits of high fructose syrup**

Item mg/ kg	Limit		Test method
	42	55	
Arsenic (As)	Max 1	Max 1	ISO 11212-1
Lead (Pb)	Max 0,1	Max 0,1	ISO 11212-3
Chloride (Cl)	Max 50	Max 50	<a href="#">Subclause 5.6</a>
Copper (Cu)	-	Max 1,5	AOAC method

### 4.4 Microbial limits

Microbial limits shall comply with the requirements given in [Table 4](#).

**Table 4 — Microbial limits of high fructose syrup**

Microbes CFU <sup>a</sup> /g	Max limit		Test method
	42	55	
Mould	10	10	ISO 21527-2
Yeast	10	10	ISO 21527-2
Total plate count	20	20	ISO 4833-1, ISO 4833-2
<sup>a</sup> Colony forming unit (CFU).			

## 5 Test methods

### 5.1 pH

The pH measurement of high fructose syrup should be done by immersion of standardized pH electrodes into the sample.

### 5.2 Sulfur dioxide

The measurement of sulfur dioxide in high fructose syrup should be determined in accordance with ISBT method (see Reference [\[2\]](#)).

### 5.3 Fructose and glucose content

#### 5.3.1 General

In order to determine the fructose content of high fructose syrups, the sample is passed through a metal ion-modified cation exchange column. The individual sugars are separated by molecular exclusion and ligand exchange. The eluted sugars are detected using a differential refractometer detector.

#### 5.3.2 Equipment

**5.3.2.1 Liquid chromatograph**, capable of accommodating a 22 cm to 31 cm temperature-controlled column and equipped with a constant flow pump and differential refractometer detector with attenuation capabilities.

**5.3.2.2 Chromatography columns**, pre-packed cation exchange columns (calcium or silver) are recommended. Examples of acceptable columns are: Hi-pex Ca, 300 \* 7,7 mm for separating DP1-DP7 saccharides.

**5.3.2.3 Guard column**, to protect the analytical column described in [5.3.2.2](#) by inserting a deionizing pre-column available from chromatographic column manufacturers.

**5.3.2.4 Chromatographic column heater**, a thermostatically controlled metal block heater accommodating two columns, capable of operating at temperatures up to 80 °C ( $\pm 0,5$  °C).

**5.3.2.5 Sample injector**, use a loop injector having a capacity of 10  $\mu$ m to 50  $\mu$ m.

### 5.3.3 Reagents

**5.3.3.1 Solvent**, degassed, purified water should be filtered through a 0,22  $\mu$ m membrane filter prior to use; it is maintained at approximately 80 °C.

**5.3.3.2 Carbohydrate standards**, possible suppliers of the sugar standards are as follows: fructose, dextrose, maltose and maltotriose.

### 5.3.4 Instrument parameters

- Flow rate 0,5 ml /min;
- Column temperature 80 °C;
- Detector temperature 45 °C.

### 5.3.5 Procedure

Prepare a 0,1 % to 0,5 % standard mixture of glucose, fructose, maltose and maltotriose with distilled water and find the retention time of these sugars in chromatograph. It is not necessary to draw a calibration curve. To determine the concentration of each sugar, consider the ratio of the corresponding area for each sugar to the total area.

## 5.4 Hydroxymethylfurfural (HMF)

### 5.4.1 Eluent preparation

- Solvent A: Into a 1 000 ml volumetric flask, dissolve 4,49 g of potassium phosphate monobasic with high performance liquid chromatography (HPLC) water. Add 25 ml acetic acid and fill to volume with HPLC water. Adjust the pH with 1 N sodium hydroxide (NaOH) solution to pH 3,5.
- Solvent B: HPLC grade methanol.
- Filter both eluent through 0,45  $\mu$ m filter and degas by sonication. Use a mixture of two solvents (A and B) in a ratio of 95 to 5.

### 5.4.2 Sample preparation

- Weigh accurately 2,5 g of sugar sample into beaker.
- Add 2,5 g of HPLC water to beaker. Mix well until dissolved.
- Filter sample through 0,22  $\mu$ m syringe filter into HPLC vial, and cap.