# CYCLOTETRAGLUCOSE SYRUP
(TENTATIVE)

*New specifications prepared at the 68th JECFA (2007) and published in FAO JECFA Monographs 4 (2007). A temporary ADI "not specified" was established at the 68th JECFA (2007).*

*Information is required on (1) the level of total saccharides and the test method and (2) the unidentified saccharides. The specifications will be withdrawn if the requested information is not made available by the end of 2008.*

**SYNONYMS** Cyclotetraose syrup, Cyclic nigerosyl-(1→6)-nigerose syrup, cycloalternan syrup, cycloalternanotetraose syrup

**DEFINITION** A mixture consisting of mono-, di- and oligosaccharides, of which cyclotetraglucose is the major component. It is produced from hydrolyzed food-grade starch by the actions of a mixture of 6-α-glucosyltransferase α-isomaltosyltransferase derived from *Sporosarcina globispora*, and cyclodextrin glucosyltransferase derived from *Bacillus stearothermophilus*. The final product is a syrup or a spray-dried solid.

Assay Not less than --- % of total saccharides (information required) and 30 – 40% of cyclotetraglucose on the anhydrous basis

**DESCRIPTION** Slightly sweet tasting, colourless and odourless, clear viscous liquid or dry white crystalline mass.

**FUNCTIONAL USES** Carrier

**CHARACTERISTICS**

IDENTIFICATION

Chromatography The retention time for the major peak in a HPLC chromatogram of the sample corresponds to that for cyclotetraglucose in a chromatogram of cyclotetraglucose standard using the conditions described in the Method of Assay. The retention time of cyclotetraglucose is approx. 62 min.

PURITY

Water (Vol. 4) Not more than 30% for the syrup and not more than 10% for the syrup solids (Karl Fischer Method).

Total ash (Vol. 4) Not more than 0.05% on the anhydrous basis (500°, 5h)

Lead (Vol. 4) Not more than 1 mg/kg
Determine using an AAS/ICP-AES technique appropriate to the specified level. The selection of sample size and method of sample preparation may be based on the principles of the methods described in Volume 4 (under "General

Methods, Metallic Impurities").

Microbiological criteria (Vol.4)

Total (Aerobic) plate count: not more than 300 CFU/g
Coliforms: Negative in 10 g
Yeast and moulds: Not more than 100 CFU/g

**METHOD OF ASSAY**

Determine by HPLC (Vol. 4) using the following conditions:
NOTE: Use deionized water.

Sample solution

Weigh accurately about 1000 mg of test sample into a 50-ml volumetric flask and add about 40 ml of water. Dissolve the sample completely and dilute to the mark with water.

Standard solution

Dissolve accurately weighed cyclotetraglucose standard (available under the name of cyclotetraose from Hayashibara Co., Ltd, 2-3 Shimoishii 1-chome, Okayama 700, Japan) in water to obtain a solution having known concentration of about 10 mg of cyclotetraglucose per ml.

Chromatography

Liquid chromatograph equipped with a column oven and a refractive index detector.
Column and packing: strong acidic cation exchange resin
- length: 200–400 mm
- diameter: 8–10 mm
- temperature: 80°

Mobile phase: water
Flow rate: Adjust to obtain a retention time of 55–65 min
Injection volume: 20 µl
The retention time of cyclotetraglucose is approx. 62 min.

System suitability

Upon chromatography of a solution containing about 0.4% cyclotetraglucose and 0.4% glucose, the resolution (Vol. 4) is not less than 1.0 between glucose (first peak) and cyclotetraglucose (second peak).

Procedure

Inject the sample solution into the chromatograph, and measure the area of the cyclotetraglucose peak. Repeat for the standard solution. Calculate the percentage of cyclotetraglucose in the test sample as follows:

$$\% \text{ cyclotetraglucose (dried basis)} = 100 \times (A_S/A_R)(W_R/W_S)$$

where:

$A_S$ and $A_R$ are the areas of the peaks due to cyclotetraglucose for the sample solution and standard solution, respectively.
$W_S$ and $W_R$ are the weights (mg) of the test sample and standard cyclotetraglucose, respectively, corrected for water content.