

---

---

**Apple juice, apple juice concentrates and  
drinks containing apple juice —  
Determination of patulin content —**

**Part 1:**

Method using high-performance liquid  
chromatography

*Jus de pommes, concentrés de jus de pommes et boissons à base de jus  
de pommes — Détermination de la teneur en patuline —*

*Partie 1: Méthode par chromatographie en phase liquide à haute  
performance*



## 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.

Draft International Standards adopted by the technical committees are circulated to the member bodies for voting. Publication as an International Standard requires approval by at least 75 % of the member bodies casting a vote.

International Standard ISO 8128-1 was prepared by Technical Committee ISO/TC 34, *Agricultural food products*, Sub-Committee SC 3, *Fruit and vegetable products*.

ISO 8128 consists of the following parts, under the general title *Apple juice, apple juice concentrates and drinks containing apple juice — Determination of patulin content*:

- *Part 1: Method using high-performance liquid chromatography*
- *Part 2: Method using thin-layer chromatography*

Annex A of this part of ISO 8128 is for information only.

© ISO 1993

All rights reserved. No part of this publication may be reproduced or utilized in any form or by any means, electronic or mechanical, including photocopying and microfilm, without permission in writing from the publisher.

International Organization for Standardization  
Case Postale 56 • CH-1211 Genève 20 • Switzerland

Printed in Switzerland

This is a preview of ISO 8128-1:1993. Click here to purchase the full version from the ANSI store.

# Apple juice, apple juice concentrates and drinks containing apple juice — Determination of patulin content —

## Part 1:

### Method using high-performance liquid chromatography

#### 1 Scope

This part of ISO 8128 specifies a method using high-performance liquid chromatography for the determination of the patulin content of apple juice, apple juice concentrates and drinks containing apple juice.

The limit of detection of the method is 10 µg/l, based on 5 ml of ready-to-drink apple juice.

NOTE 1 ISO 8128-2 specifies a method using thin-layer chromatography.

#### 2 Principle

Extraction of patulin from a test portion using ethyl acetate, followed by partitioning of the extract with aqueous sodium carbonate solution. Qualitative and quantitative determination of the patulin content by means of high-performance liquid chromatography (HPLC) using an ultraviolet (UV) detector.

#### 3 Reagents

Use only reagents of recognized analytical grade, and water of HPLC grade.

**3.1 Solvent**, ethyl acetate.

**3.2 Mobile phase**, acetonitrile, 10 % (V/V) solution.

**3.3 Extraction solution**, 14 g/l aqueous solution of anhydrous sodium carbonate.

**3.4 Acetate buffer**, pH 4.

Mix 16,4 ml of dilute acetic acid [ $c(\text{CH}_3\text{COOH}) = 0,2 \text{ mol/l}$ ] with 3,6 ml of sodium acetate [ $c(\text{CH}_3\text{COONa}) = 0,2 \text{ mol/l}$ ].

**3.5 Acetic acid**, glacial.

**3.6 Patulin standard solution** ( $\text{C}_7\text{H}_6\text{O}_4$ ).

##### 3.6.1 Preparation

Weigh, to the nearest 0,1 mg, 10,0 mg of patulin in a 100 ml one-mark volumetric flask and dissolve it in the acetate buffer (3.4). Make up to the mark with the acetate buffer.

Pipette 10,0 ml of this solution into another 100 ml one-mark volumetric flask and make up to the mark with the acetate buffer.

The patulin content of this standard solution is 10 µg/ml approximately.

Measure the absorbance at 276 nm of this standard solution on an appropriate spectrometer using quartz cells of optical path length 10 mm.

NOTE 2 The preparation of the standard solution and the control of its purity are based on reference [3].

##### 3.6.2 Calculation of the concentration

Calculate the concentration  $\rho_{\text{ps}}$ , expressed in micrograms per millilitre, of the patulin solution (3.6.1) using the formula

$$\rho_{\text{ps}} = \frac{A \times M_r \times 1\,000 \times C}{A_{276}}$$