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Infant formula and adult nutritionals — Determination of vitamin E and vitamin A by normal phase high performance liquid chromatography

Formules infantiles et produits nutritionnels pour adultes — Détermination de la teneur en vitamine E et de la teneur en vitamine A par chromatographie liquide à haute performance en phase normale

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ISO copyright office
Ch. de Blandonnet 8 • CP 401
CH-1214 Vernier, Geneva, Switzerland
Tel. +41 22 749 01 11
Fax +41 22 749 09 47
copyright@iso.org
www.iso.org

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Foreword

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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).

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For an explanation on the meaning of ISO specific terms and expressions related to conformity assessment, as well as information about ISO's adherence to the WTO principles in the Technical Barriers to Trade (TBT) see the following URL: Foreword - Supplementary information

The committee responsible for this document is ISO/TC 34, *Food products* in collaboration with AOAC INTERNATIONAL. It is being published by ISO and separately by AOAC INTERNATIONAL. The method described in this International Standard is equivalent to the AOAC Official Method 2012-10: *Infant formula and adult nutritionals — Determination of vitamin E and vitamin A by normal phase high performance liquid chromatography*.

Infant formula and adult nutritionals — Determination of vitamin E and vitamin A by normal phase high performance liquid chromatography

WARNING — The use of this International Standard can involve hazardous materials, operations and equipment. This International Standard does not purport to address all the safety problems associated with its use. It is the responsibility of the user of this International Standard to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use.

1 Scope

This International Standard specifies a method for the simultaneous quantitative determination of vitamin E (α-tocopherol and α-tocopheryl acetate) and vitamin A (13-*cis* and all-*trans* isomers of retinyl palmitate and retinyl acetate) present in all forms of infant and adult formulas (powders, ready-to-feed liquids and liquid concentrates).

Retinol is not used for fortification purposes and therefore is not addressed in this method. The innate amount in products is insignificant.

Stereoisomers of vitamin E, α-tocopherol and α-tocopheryl acetate, are not differentiated in this method.

2 Terms and definitions

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

2.1
adult nutritional
nutritionally complete, specially formulated food, consumed in liquid form, which may constitute the sole source of nourishment, made from any combination of milk, soy, rice, whey, hydrolysed protein, starch and amino acids, with and without intact protein

2.2
infant formula
breast-milk substitute specially manufactured to satisfy, by itself, the nutritional requirements of infants during the first months of life up to the introduction of appropriate complementary feeding

[SOURCE: Codex Standard 72-1981]

3 Principle

This procedure utilizes the proteolytic enzyme, papain, to hydrolyze the hydrophilic protein coating of fat micelles in milk or soy-based infant formulations in an aqueous solution. The hydrophobic contents of the micelles are then extracted quantitatively into iso-octane in a single extraction. The extract is analysed by normal phase HPLC using an analytical column with gradient elution. Quantification of α-tocopherol and α-tocopheryl acetate is done using fluorescence detection with excitation and emission wavelengths at 280 nm and 310 nm. Retinyl palmitate (*cis* and *trans*) and retinyl acetate (*cis* and *trans*) are quantified using UV detection at 325 nm.