
**Tobacco — Determination of tobacco
specific nitrosamines — Method using
buffer extraction**

*Tabac — Dosage des nitrosamines spécifiques du tabac — Méthode
d'extraction par solution tampon*



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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.

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The main task of technical committees is to prepare International Standards. 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.

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ISO 22303 was prepared by Technical Committee ISO/TC 126, *Tobacco and tobacco products*.

Introduction

During the development of this International Standard, inter-laboratory tests were carried out using two different methods for the determination of tobacco specific nitrosamines; this method, using buffer extraction, and the method using alkaline dichloromethane extraction (see References [2], [3]).

These studies show that no differences occur between the results obtained by the two different methods (see Reference [4]). The method using alkaline dichloromethane extraction is described in Technical Specification ISO/TS 22304 (see Reference [1]).

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Tobacco — Determination of tobacco specific nitrosamines — Method using buffer extraction

1 Scope

This International Standard specifies the procedure for the determination of the tobacco specific nitrosamines (TSNAs): N-nitrosornicotine (NNN), N-nitrosoanatabine (NAT), N-nitrosoanabasine (NAB) and 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK) in ground leaf tobacco, manufactured tobacco and tobacco products. The determination is by means of gas chromatography.

2 Normative references

The following referenced documents are indispensable for the application 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 1042, *Laboratory glassware — One-mark volumetric flasks*

ISO 3696, *Water for analytical laboratory use — Specification and test methods*

3 Terms and definitions

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

3.1

tobacco specific nitrosamines

TSNAs

four nitrosamines found predominantly in tobacco: N-nitrosornicotine (NNN), N-nitrosoanatabine (NAT), N-nitrosoanabasine (NAB) and 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK)

4 Principle

TSNAs are extracted from ground tobacco samples using a buffer solution. The aqueous portion of the buffer is absorbed into diatomaceous earth. The TSNAs are then eluted from the diatomaceous earth with methylene chloride and concentrated in a heated water bath using nitrogen. The TSNAs are separated and quantified by gas chromatography with chemiluminescent detection. Quantification is performed by an internal standard technique.

5 Reagents

Use only reagents of recognised analytical grade.

SAFETY PRECAUTIONS — Nitrosamines are suspected carcinogens; therefore, appropriate safety precautions should be taken when preparing standards. Always wear laboratory gloves when handling standard solutions and making dilutions.