

ISO/FDIS 21766 ISO 21766:2021 (E)

Tobacco and tobacco products — Determination of tobacco-specific nitrosamines in tobacco products — Method using LC-MS/MS Tobacco and tobacco products — Determination of tobacco-specific nitrosamines in tobacco products — Method using LC-MS/MS

Contents

	Foreword
	Introduction
	Foreword
	Introduction
1	Scope
2	Normative references
3	Terms and definitions
4	Principle
5	Reagents
6	Apparatus
7	Preparation
7.1	Preparation of glassware
7.2	Preparation of solutions
7.2.1	Extraction solution, 100 mM ammonium acetate in water
7.2.2	HPLC Mobile Phase A: Water, resistivity $\geq 18,2 \text{ M}\Omega\cdot\text{cm}$ at 25 °C
7.2.3	HPLC Mobile Phase B: 0,1 % acetic acid in methanol
7.3	Preparation of standards
7.3.1	General
7.3.2	Preparation of internal standard solutions
7.3.2.1	Stock solution
7.3.2.2	Combined secondary internal standard solution
7.3.2.3	Internal standard spiking solution
7.3.3	Preparation of calibration standard solutions
7.3.3.1	Stock solution
7.3.3.2	Mixed TSNA standard solution (I)
7.3.3.3	Mixed TSNA standard solution (II)
7.3.3.4	TSNA calibration standards
8	Sampling
8.1	General
8.2	Sample preparation
8.3	Sample extraction
9	Sample analysis
9.1	General
9.2	Suggested HPLC parameters
9.3	MS/MS parameters
9.3.1	General
9.3.2	Quantification and qualification transitions
9.4	System suitability

	9.5	Calibration
	9.6	Calculation
10		Repeatability and reproducibility
11		Test report
1		Scope
2		Normative references
3		Terms and definitions
4		Principle
5		Reagents
6		Apparatus
7		Preparation
	7.1	Preparation of glassware
	7.2	Preparation of solutions
	7.2.1	Extraction solution, 100 mM ammonium acetate in water
	7.2.2	HPLC Mobile Phase A: Water, resistivity $\geq 18,2 \text{ M}\Omega\cdot\text{cm}$ at $25 \text{ }^\circ\text{C}$
	7.2.3	HPLC Mobile Phase B: 0,1 % acetic acid in methanol
	7.3	Preparation of standards
	7.3.1	General
	7.3.2	Preparation of internal standard solutions
	7.3.2.1	Stock solution
	7.3.2.2	Combined secondary internal standard solution
	7.3.2.3	Internal standard spiking solution
	7.3.3	Preparation of calibration standard solutions
	7.3.3.1	Stock solution
	7.3.3.2	Mixed TSNA standard solution (I)
	7.3.3.3	Mixed TSNA standard solution (II)
	7.3.3.4	TSNA calibration standards
8		Sampling
	8.1	General
	8.2	Sample preparation
	8.3	Sample extraction
9		Sample analysis
	9.1	General
	9.2	Suggested HPLC parameters
	9.3	MS/MS parameters
	9.3.1	General
	9.3.2	Quantification and qualification transitions
	9.4	System suitability
	9.5	Calibration
	9.6	Calculation
10		Repeatability and reproducibility
11		Test report
Annex A (informative) Sample clean-up using solid phase extraction (SPE)		
	A.1	Reagents and supplies
	A.2	Reagent preparation
	A.2.1	Wash No. 1: 4,5 % methanol and 0,5 % ammonium hydroxide in water
	A.2.2	Wash No. 2: 0,01 % formic acid in water
	A.2.3	Eluting solvent: 30 % water and 0,1 % acetic acid in methanol
	A.3	SPE procedure

Annex B (informative) Examples of typical chromatograms

Annex A (informative) Sample clean-up using solid phase extraction (SPE)

- A.1 Reagents and supplies**
- A.2 Reagent preparation**
 - A.2.1 Wash No. 1: 4,5 % methanol and 0,5 % ammonium hydroxide in water**
 - A.2.2 Wash No. 2: 0,01 % formic acid in water**
 - A.2.3 Eluting solvent: 30 % water and 0,1 % acetic acid in methanol**
- A.3 SPE procedure**

Annex B (informative) Examples of typical chromatograms

Page count: 0 16