volumetric absorptive microsampling VAMS N-Desmethyltamoxifen analysis

Author: Smolecule Technical Support Team. **Date**: February 2026

Compound Focus: **N-Desmethyltamoxifen hydrochloride**

CAS No.: 15917-65-4

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Introduction to VAMS for Tamoxifen Metabolite Monitoring

Volumetric Absorptive Microsampling (VAMS) represents a significant advancement in therapeutic drug monitoring for breast cancer patients undergoing tamoxifen therapy. As a prodrug, tamoxifen undergoes extensive hepatic metabolism to form active metabolites including **N-desmethyltamoxifen (NDT)**, **4-hydroxytamoxifen (4HT)**, and **endoxifen (END)**, with endoxifen being particularly important due to its 30-100 times greater potency than the parent drug [1] [2].

The clinical significance of monitoring these metabolites stems from research demonstrating that endoxifen concentrations above **5.9 ng/mL** are associated with reduced recurrence risk in breast cancer patients [1] [3]. However, traditional plasma sampling methods present logistical challenges including invasiveness, requirement for cold chain storage, and larger sample volumes. VAMS technology addresses these limitations by enabling simple, volumetric collection of small blood samples (typically 10-50 µL) that remain stable at room temperature for extended periods [4].

Materials and Equipment

Chemical Reagents

- **Reference standards**: Tamoxifen, (Z)-Endoxifen, (Z)-4-Hydroxytamoxifen, and N-desmethyltamoxifen [1]
- **Internal standard**: Propranolol Hydrochloride [1]
- **HPLC-grade solvents**: Methanol, acetonitrile [1]
- **Mobile phase additive**: Formic acid (0.1%) [1]
- **Whole blood** for calibration standards and quality controls [1]

Equipment and Instruments

- **VAMS devices**: Mitra tips (10-50 µL capacity) [1]
- **LC-MS/MS system**: Waters Xevo TQD Triple Quadrupole [1]
- **UPLC column**: Acquity UPLC BEH C18 (2.1 × 100 mm; 1.7 µm) [1]
- **Sample preparation equipment**: Vortex mixer, sonication bath, nitrogen evaporator [1]

Experimental Protocol

Sample Collection Procedure

- **VAMS Sampling**: Dip the VAMS tip into fresh whole blood at a 45° angle for 2-4 seconds until the tip is completely saturated and appears uniformly red [4]
- **Drying Process**: Air-dry the collected samples horizontally for 2 hours at room temperature, ensuring tips do not contact each other or surfaces to prevent cross-contamination [1]
- **Storage**: Store dried VAMS samples in sealed plastic bags with desiccant at room temperature; demonstrated stability up to 30 days [1]

Sample Preparation and Extraction

- **Transfer**: Remove VAMS tip from handler and place in microcentrifuge tube [4]
- **Extraction**: Add 1 mL of methanol containing internal standard (propranolol, 100 ng/mL) [1]
- **Vortex**: Mix thoroughly for 1 minute [1]
- **Sonication**: Perform sonication-assisted extraction for 25 minutes [1]
- **Concentration**: Transfer 850 µL supernatant and evaporate to dryness under nitrogen stream at 55°C [1]

- **Reconstitution**: Reconstitute dried extract in 100 µL mobile phase, vortex for 20 seconds [1]
- **Injection**: Transfer 70 µL to autosampler vial; injection volume 10 µL [1]

LC-MS/MS Analysis Conditions

Table 1: Chromatographic Conditions for Tamoxifen Metabolite Analysis

Parameter	Specification
Column	Acquity UPLC BEH C18 (2.1 × 100 mm; 1.7 µm)
Mobile Phase A	0.1% Formic acid in water
Mobile Phase B	0.1% Formic acid in acetonitrile
Flow Rate	0.2 mL/min
Gradient Program	Initial 5% B (0-3 min), increase to 70% B (3-5 min)
Run Time	5 minutes
Column Temperature	50°C [5]
Injection Volume	10 µL

Table 2: Mass Spectrometry Parameters

Analyte	Precursor Ion > Product Ion (m/z)	Ionization Mode	Cone Voltage (V)	Collision Energy (V)	Retention Time (min)
N-desmethyltamoxifen	358.31 > 58.27	ESI+	42	24	2.61
Tamoxifen	372.33 > 72.28	ESI+	50	27	2.62
Endoxifen	374.25 > 58.25	ESI+	45	30	2.36

Analyte	Precursor Ion > Product Ion (m/z)	Ionization Mode	Cone Voltage (V)	Collision Energy (V)	Retention Time (min)
4-hydroxytamoxifen	388.22 > 72.28	ESI+	50	27	2.40
Propranolol (IS)	260.26 > 116.12	ESI+	35	20	2.21

Method Validation

The developed VAMS method has been comprehensively validated according to FDA and EMA guidelines [1].

Table 3: Method Validation Parameters

Validation Parameter	N-desmethyltamoxifen	Tamoxifen	Endoxifen	4-hydroxytamoxifen
LLOQ (ng/mL)	2.00	2.50	2.50	1.50
Accuracy (% bias)	≤15%	≤15%	≤15%	≤15%
Precision (% CV)	≤15%	≤15%	≤15%	≤15%
Extraction Recovery	>90%	>90%	>90%	>90%
Ion Suppression	<7%	<7%	<7%	<7%
Carryover	<6%	<6%	<6%	<6%
Stability	30 days at room temperature	30 days at room temperature	30 days at room temperature	30 days at room temperature

Data Analysis

Calibration Curve

Calibration curves are prepared using fresh spiked whole blood with concentrations covering the expected physiological range [1]. The lower limit of quantification (LLOQ) demonstrates accuracy and precision within 20% bias and CV [1].

Quality Control

Quality control samples at low, medium, and high concentrations should be analyzed alongside patient samples. For N-desmethyltamoxifen, QC concentrations of 6, 250, and 500 ng/mL are recommended [1].

Application Notes

- **Hematocrit Considerations**: While VAMS minimizes hematocrit effects compared to DBS, further investigation of hematocrit impact is recommended [3]
- **Comparative Performance**: VAMS demonstrates superior recovery compared to DBS, potentially due to differences in materials and intermolecular properties [6] [3]
- **Clinical Implementation**: The method has been successfully applied to monitor tamoxifen and metabolites in 30 ER+ breast cancer patients, proving suitable for therapeutic drug monitoring [1] [6]
- **Bridging Studies**: When implementing VAMS for clinical monitoring, conduct bridging studies between venous plasma and capillary blood to account for potential matrix differences [3]

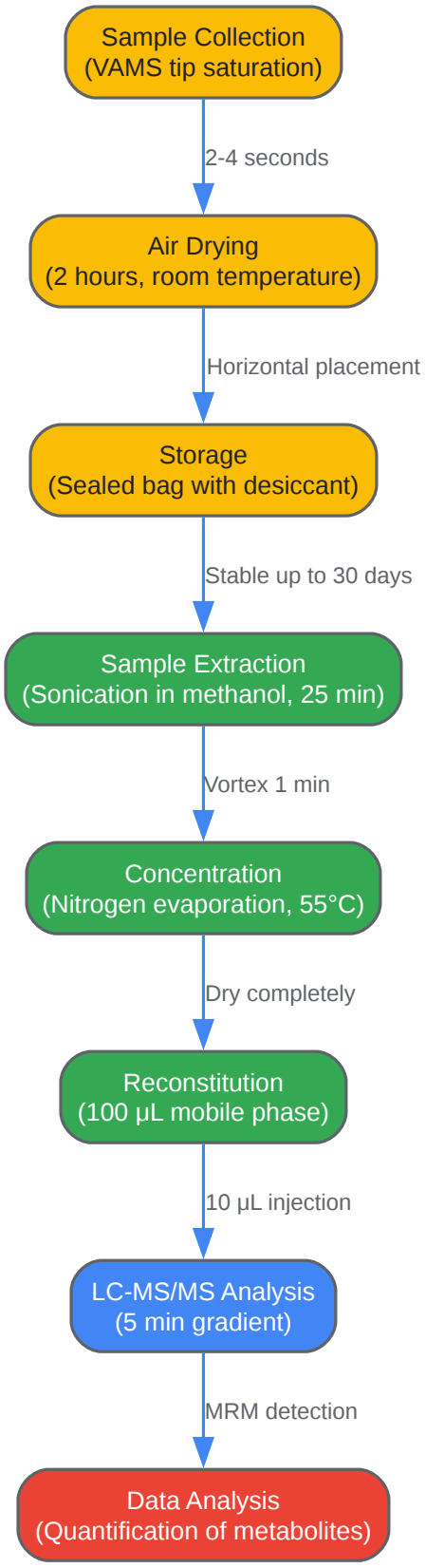
Troubleshooting

- **Incomplete Extraction**: Ensure adequate sonication time (25 minutes) and consider solvent modifications if recovery is suboptimal [1]

- **Matrix Effects**: Maintain ion suppression below 7% through proper sample cleaning and chromatographic separation [1]
- **Carryover**: Monitor carryover regularly; should remain below 6% for all analytes [1]

The experimental workflow for VAMS analysis of tamoxifen metabolites is as follows:

VAMS Experimental Workflow for Tamoxifen Metabolite Analysis



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References

1. as a New Biosampling Tool... Volumetric Absorptive Microsampling [pmc.ncbi.nlm.nih.gov]

2. Volumetric Absorptive Microsampling in Breast Cancer [dovepress.com]

3. Microsampling to Measure Tamoxifen for Breast Cancer TDM [neoteryx.com]

4. | Encyclopedia MDPI Volumetric Absorptive Microsampling [encyclopedia.pub]

5. New UPLC–MS/MS assay for the determination of ... [pmc.ncbi.nlm.nih.gov]

6. of tamoxifen and its metabolites in dried blood spot and... Analysis [scholar.ui.ac.id]

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