



# Automated Solid Phase Extraction for Antibiotics in Animal Feed

## Introduction

It is common practice to incorporate antibiotics in animal feedstuffs such as corn, rice and maize. This method describes how to extract and cleanup a sample of Tiamulin Hydrogen Fumarate from poultry grain although it is possible this method will work for a range of antibiotics.

## Instrumentation Used for Sample Preparation



The RapidTrace for Automated Solid Phase Extraction



TurboVap LV for Automated Sample Evaporation

## Sample Preparation

A 25g sample of feedstuff is shaken for 30 minutes with Chloroform (150mL) and Sodium Carbonate (10mL) mixture to extract the antibiotic. Acetonitrile (100mL) is then added and after a brief shake a 10mL sample is filtered into a 16mm x 100mm vial ready to be cleaned up automatically by the RapidTrace. The Solid Phase Extraction is carried out using a 1mL Iolute cartridge filled with 130mg of SCX packing material. Following extraction the 1mL of eluent A (Ammonium Acetate: Acetonitrile, pH10) is neutralised with 1mL of eluent B (Ammonium Acetate: Acetonitrile pH1). A 1mL sample is transferred to a 2mL vial and 20uL is injected onto the HPLC for analysis.

## RapidTrace Overview

The RapidTrace is an automated Solid Phase Extraction instrument capable of preparing 10 samples. It has a modular design so multiple RapidTrace units can increase the throughput without risk of downtime due to the independence of each instrument. All

solvent lines are purged and primed with solvent first. A maximum of 8 solvents can be used to run a wide range of methods. The 4 independent waste solvent lines can be separated if needed. The instrument sample rack has two rows of 10 test tubes, 16mm x 100mm for holding the samples and eluent fractions. This method offers an automated, rugged and reproducible solution for cleaning up the samples to remove interferences.

## The RapidTrace Method

STEP	SOURCE	DESTINATION	VOLUME	FLOW (ML/MIN)
Condition	Acetonitrile	Organic Waste	3.0	15.0
Load	Sample	Organic Waste	5.0	2.0
Load	Sample	Organic Waste	5.0	2.0
Rinse	Methanol	Organic Waste	2.0	5.0
Rinse	Acetonitrile	Organic Waste	2.0	5.0
Dry	Nitrogen Gas		4 minutes	(20 psi)
Purge Cannula	EluentA	Cannula Waste	2.0	30.0
Collect	EluentA	Fraction 1	1.0	0.5
Purge Cannula	Water	Cannula Waste	2.0	30.0
Purge Cannula	Methanol	Cannula Waste	2.0	30.0
Purge Cannula	Acetonitrile	Cannula Waste	2.0	30.0

## Common Reagent Table for All Methods

LINE NO.	REAGENT NAME	SIP SPEED (ML/MIN)
1	Methanol	30
2	Acetonitrile	30
3	Water	30
4	Eluent	30

WASTE NAME	ABBREVIATION
Aqueous Waste	AqW
Organic Waste	OrgW
Cannula Waste	Cannula

Air Push = 2ml

Air Push Multiplier = 2



**Corporate Headquarters**  
68 Elm Street  
Hopkinton, MA 01748-1668

Tel: 508 436 9500  
Fax: 508 435 3439  
Email: [cust.support@caliperLS.com](mailto:cust.support@caliperLS.com)

[www.caliperLS.com](http://www.caliperLS.com)

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