

Introduction

- The β 2-agonists have been used worldwide as illegal growth promoters in pork production.
- Recently incidences of poisoning occurred due to high levels of the β -agonist (clenbuterol) in pork.
- Solid-Phase Extraction is an efficiency sample preparation method for extraction and purification of β 2-agonists in pork.
- This application note used Agilent's new SCX polymer cartridges to extract and enrich four β -agonists from pork and analysis by LC-MS/MS.

β 2-Agonist Compounds Used in this Study

Compound	LogP	Structure
Terbutaline	0.55	
Salbutamol	0.44	
Clebuterol	2.94	
Formoterol	1.91	

Masses Monitored in the MRM

Compound	MRM for Quantification	MRM for Confirmation
Salbutamol	240 \rightarrow 148	240 \rightarrow 222
Terbutaline	226 \rightarrow 152	226 \rightarrow 125
Clenbuterol	277 \rightarrow 203	277 \rightarrow 259
Formoterol	345 \rightarrow 149	345 \rightarrow 327



Experimental

Instrument Conditions

Analytical column: Zorbax Eclipse Plus C18 2.1 \times 50mm 1.8 μ m

Flow rate: 0.4 mL/min

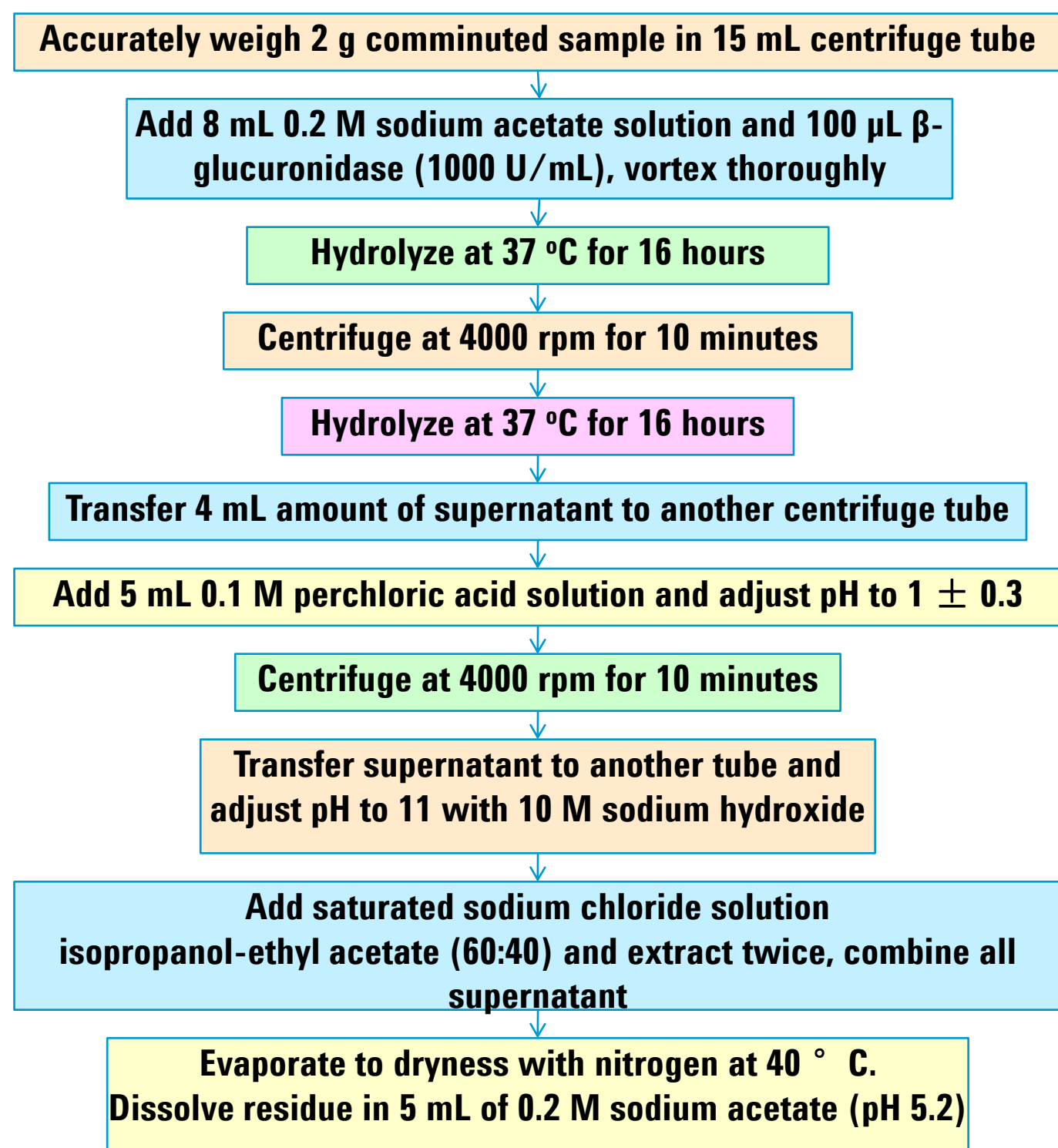
Mobile phase: A. 0.1% formic acid and 2 mmol/L ammonium acetate
B. acetonitrile

Gradient:

Time (min)	A%	B%
0	90	10
0.5	90	10
1.8	20	80
2	90	10
3.5	90	10

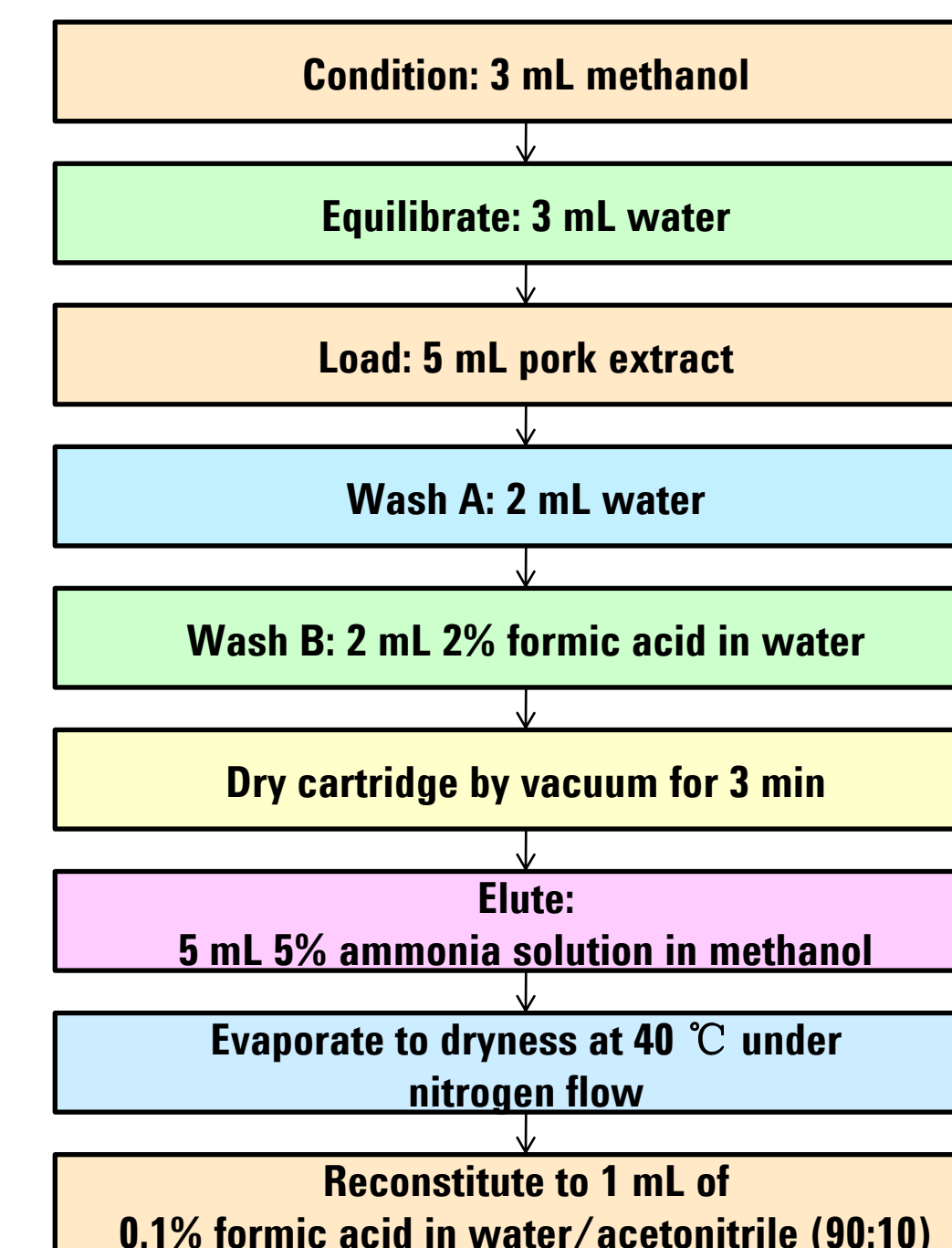
Acquisition		Source	Chromatogram	Instrument	Diagnostics
Source parameters					
Gas Temp:	325	°C		325	°C
Gas Flow:	5	l/min		5.1	l/min
Nebulizer:	45	psi		35.0036	psi
Sheath Gas Temp:	400	°C		392	°C
Sheath Gas Flow:	12	l/min		12.0	l/min
Positive		Negative			
Capillary:	4000	V	3500	V	2000
Nozzle Voltage:	0	V	1000	V	
Chamber Current:					0.19
					μ A

Sample preparation: liquid-liquid extraction

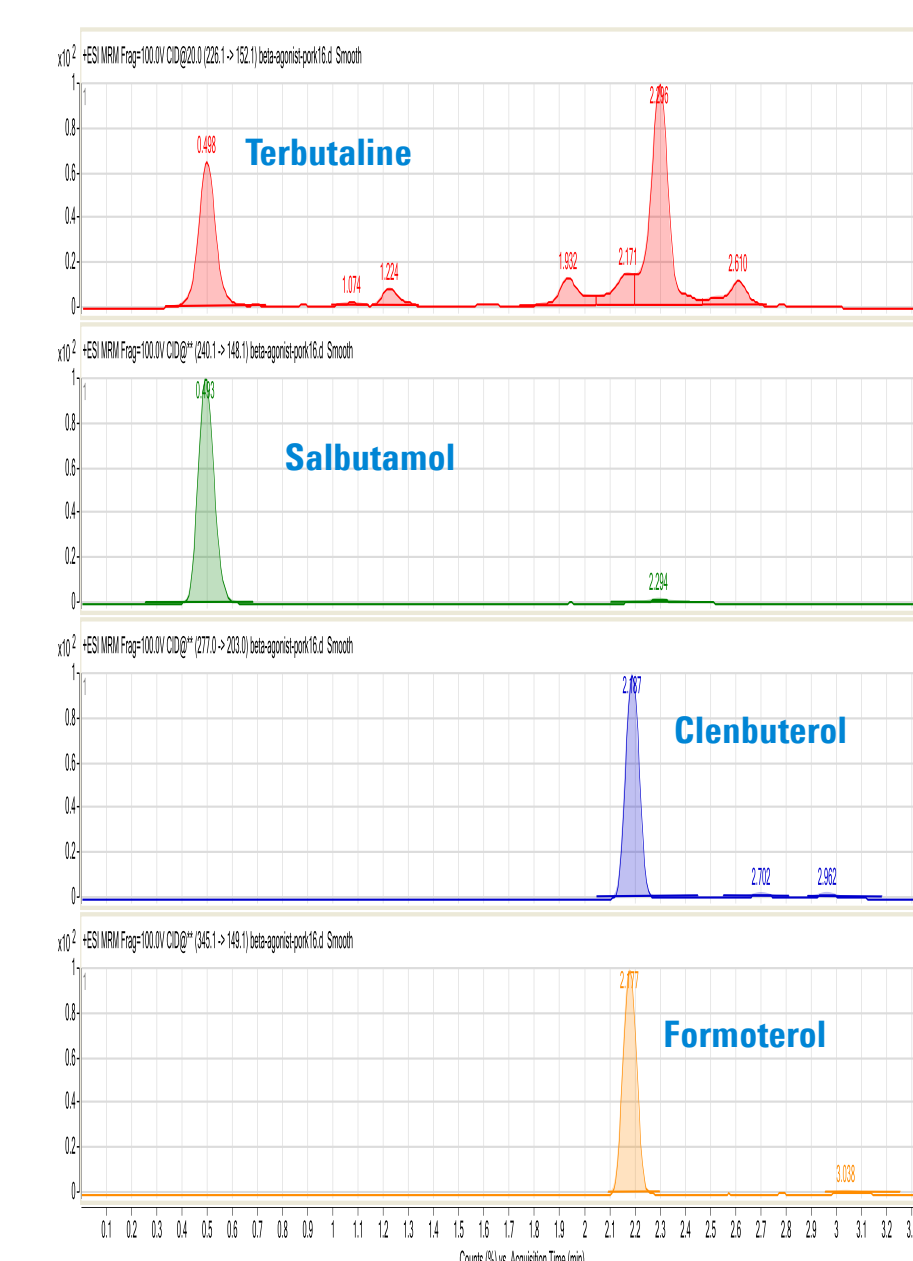


Experimental

Sample preparation: solid-phase extraction



Results and Discussion



Compound	Spiked level (ng/g pork)	Recovery (%)	RSD (n=6)
Terbutaline	0.5	88.7	5.4
	1	98.0	7.2
Salbutamol	0.5	100.6	1.8
	1	92.9	2.1
Clenbuterol	2	97.4	3.9
	0.5	82.3	5.0
Formoterol	1	91.5	6.3
	2	90.6	4.3
Formoterol	0.5	85.1	1.9
	1	83.0	4.0
	2	77.9	2.5