TERFENADINE AND METABOLITES BY REVERSE-PHASE SPE AND LC/MS/MS

HPLC METHOD

COLUMN:	XTerra® MS C_{18}, 2.1 x 30 mm, 3.5 µm
PART NUMBER:	186000398
MOBILE PHASE A:	Water + 0.1M NH_4 formate pH 9.5
MOBILE PHASE B:	MeOH + 0.1M NH_4 formate pH 9.5
FLOW RATE:	0.4 mL/min
GRADIENT:	Time Profile

Time (min)	%A	%B
0	95	5
1	5	95

TEMPERATURE:	Ambient
LC INSTRUMENT:	Alliance® 2795

MS CONDITIONS

MS INSTRUMENT:	Waters Micromass Quattro™	
ION SOURCE:	ESI (+)	
SOURCE TEMP:	150 °C	
GAS CELL:	2.0 e-3 bar Argon	
CONE VOLTAGE:	35 volts	
CAPILLARY VOLTAGE:	3.5 Kv	
DRYING GAS FLOW:	500 L/hr	
CONE GAS FLOW:	50 L/hr	
DESOLVATION TEMP:	350 °C	
MRM TRANSITION:	Protriptyline (IS)	m/z 263.9 → 190.8
	Terfenadine	m/z 472.2 → 436.2
	Terfenadine-alcohol	m/z 488.2 → 452.2
	Terfenadine-carboxylate	m/z 502.2 → 466.2

OASIS® HLB GENERIC EXTRACTION PROTOCOL

Conditions for Oasis® HLB µElution Plate
Part Number 186001828BA

CONDITION:
200 µL MeOH

EQUILIBRATE:
200 µL Water

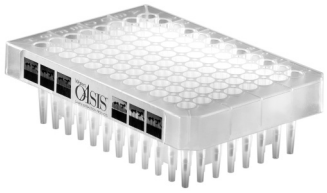
MIX/LOAD:
250 µL spiked rat plasma +
250 µL IS (10 pg/µL) solution

WASH:
200 µL 5% MeOH in water

ELUTE:
25 µL ACN:IPA 40:60 + 2% FA

DILUTE: 50 µL water,
INJECT: 20 µL

NEW! OASIS® µELUTION PLATE



METHOD VALIDATION OF TERFENADINE AND METOBOLITES ON OASIS® HLB µELUTION PLATE

Conc. (ng/mL)	0.5	1.0	5	10	20	100	200
TERFENADINE							
Day 1 (N=6) (C.V.)	0.50 (2)	0.96 (5)	4.99 (5)	10.11 (5)	19.26 (2)	103.25 (4)	197.18 (1)
Day 2 (N=6)	0.51 (2)	0.92 (3)	4.96 (2)	10.05 (4)	19.76 (5)	103.80 (5)	196.50 (1)
Day 3 (N=6)	0.50 (2)	0.97 (6)	5.00 (6)	9.87 (6)	19.37 (5)	106.73 (3)	195.23 (1)
Day 4 (N=6)	0.50 (2)	0.90 (5)	5.24 (5)	9.49 (2)	19.49 (6)	106.89 (2)	194.62 (1)
TERFENADINE-CARBOXYLATE							
Day 1 (N=6) (C.V.)	0.49 (3)	1.02 (5)	5.05 (5)	10.12 (5)	19.71 (6)	101.69 (3)	196.39 (2)
Day 2 (N=6)	0.51 (1)	0.94 (3)	4.82 (3)	10.08 (5)	20.26 (4)	99.98 (5)	202.19 (4)
Day 3 (N=6)	0.50 (2)	0.97 (5)	5.04 (5)	9.89 (4)	20.53 (6)	98.50 (4)	196.57 (4)
Day 4 (N=6)	0.50 (3)	0.99 (3)	5.07 (3)	9.86 (6)	20.14 (3)	99.03 (5)	199.20 (3)
TERFENADINE -ALCOHOL							
Day 1 (N=6) (C.V.)	0.51 (2)	0.97 (4)	5.19 (5)	10.24 (4)	19.81 (4)	102.05 (3)	196.55 (4)
Day 2 (N=6)	0.51 (1)	0.93 (2)	5.06 (2)	10.27 (4)	20.40 (3)	99.69 (6)	199.17 (5)
Day 3 (N=6)	0.51 (1)	0.94 (3)	4.77 (2)	10.02 (5)	20.07 (5)	99.73 (5)	198.60 (2)
Day 4 (N=6)	0.51 (2)	0.92 (2)	5.19 (3)	9.78 (4)	20.26 (4)	101.14 (5)	193.32 (6)

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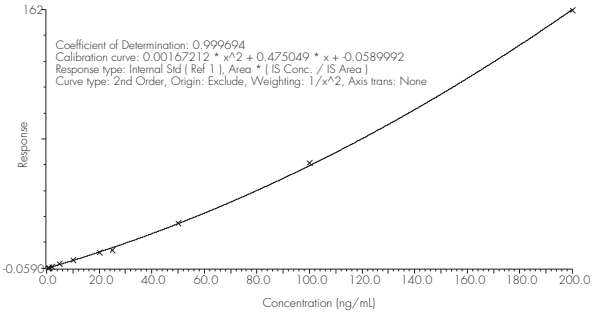
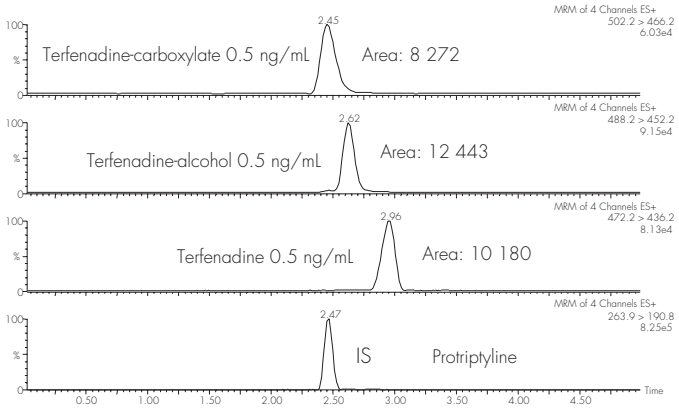
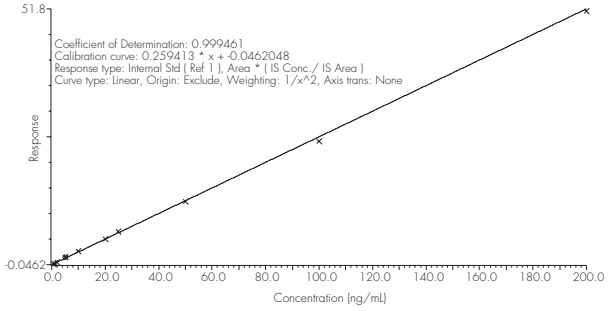
CALIBRATION CURVE OF TERFENADINE ON OASIS® HLB µELUTION PLATE



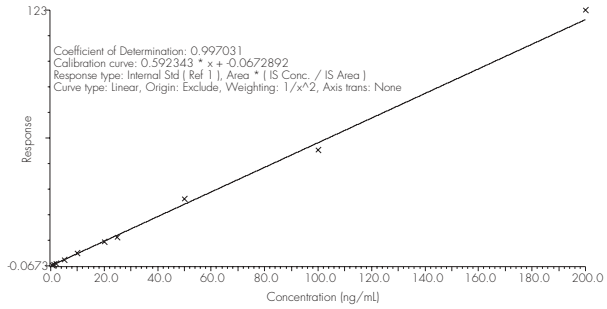
HPLC- MS/MS ANALYSIS



CALIBRATION CURVE OF TERFENADINE-CARBOXYLATE ON OASIS® HLB µELUTION PLATE



CALIBRATION CURVE OF TERFENADINE-ALCOHOL ON OASIS® HLB µELUTION PLATE



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