molecules

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Article

Development of an HPLC-MS/MS method for the determination of silybin in human plasma, urine and breast tissue.

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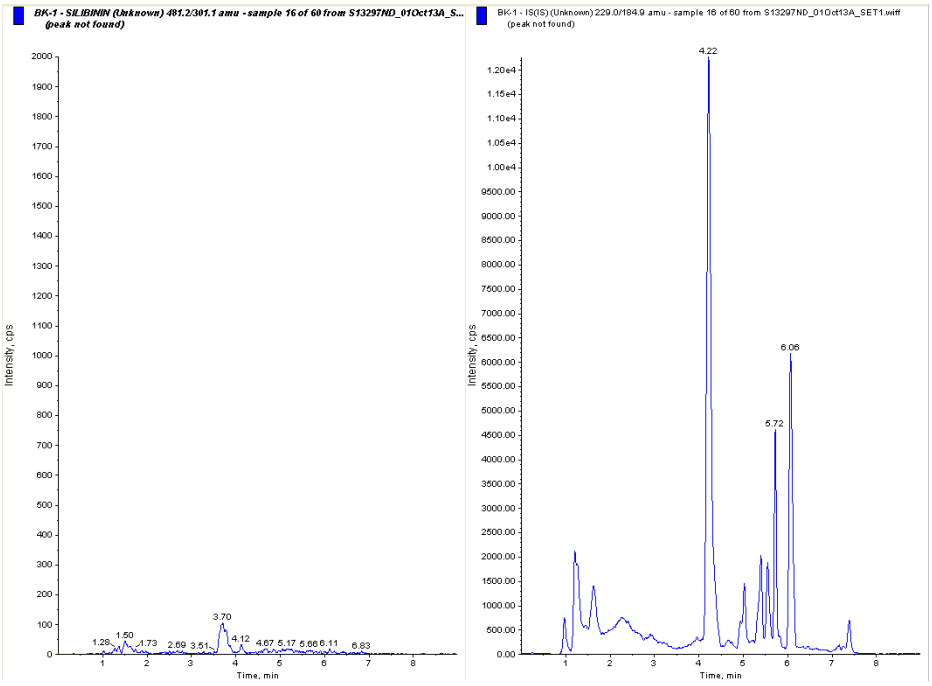
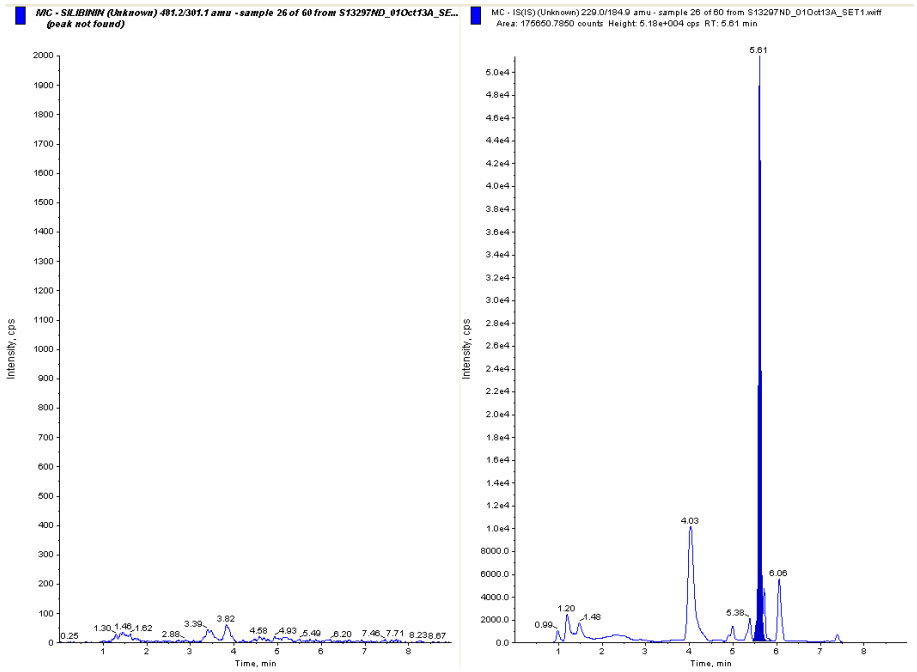
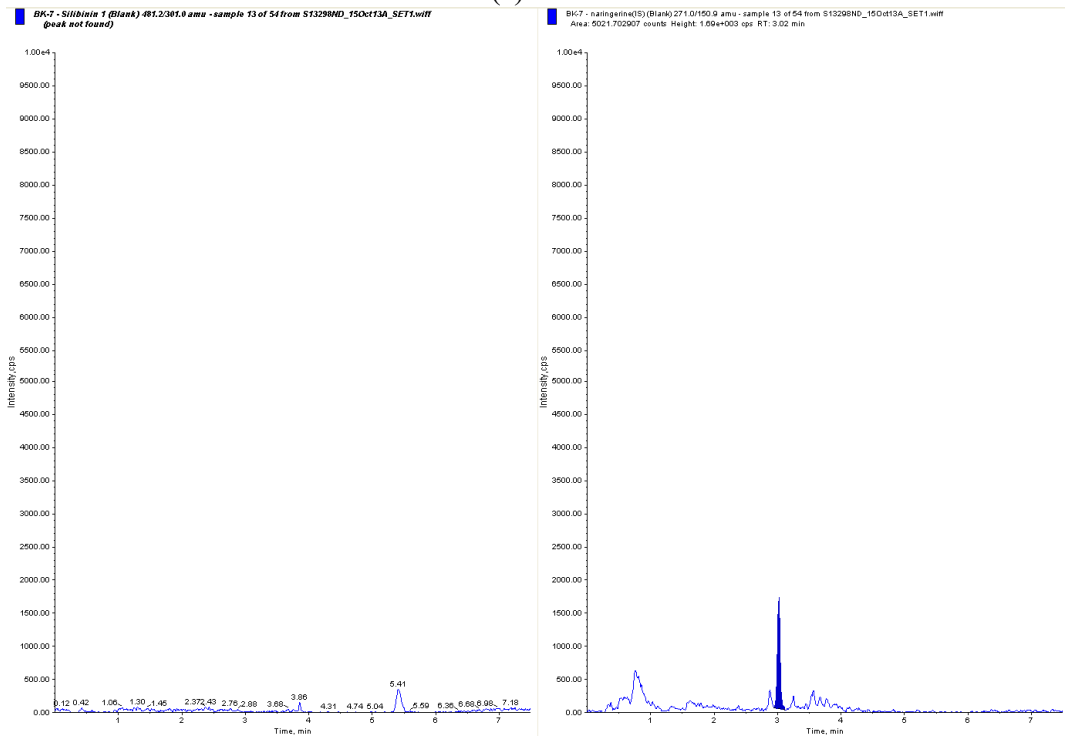
(a)



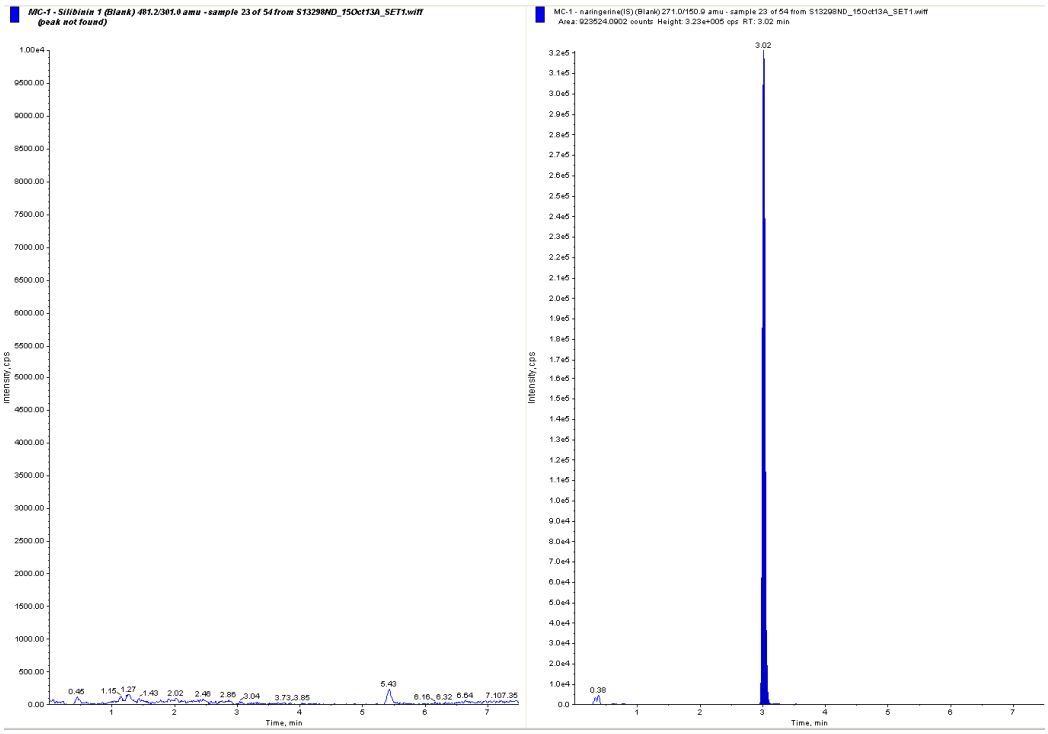
(b)


MC - SILIBININ (Unknown) 481.2/301.1 amu - sample 26 of 60 from S13297ND_01Oct13A_SE...
(peak not found)
MC - IS(IS) (Unknown) 229.0/184.9 amu - sample 26 of 60 from S13297ND_01Oct13A_SET1.wiff
Area: 175650.7850 counts Height: 5.18e+004 cps RT: 5.61 min
Intensity, cps
Time, min
0.25
1.30
1.46
1.62
2.88
3.39
3.82
4.58
4.93
5.49
6.20
7.46
7.71
8.23
8.67
0.99
1.20
1.48
4.03
5.38
5.61
6.06

(a)


BK-7 - Silibinin 1 (Blank) 481.2/301.0 amu - sample 13 of 54 from S13298ND_15Oct13A_SET1.wiff
(peak not found)
BK-7 - naringenine(IS) (Blank) 271.0/150.9 amu - sample 13 of 54 from S13298ND_15Oct13A_SET1.wiff
Area: 5021.702907 counts Height: 1.69e+003 cps RT: 3.02 min
Intensity, cps
Time, min
0.12
0.42
1.06
1.30
1.45
2.37
2.43
2.76
2.88
3.68
3.86
4.31
4.74
5.04
5.41
5.59
6.35
6.68
6.98
7.18

(b)



Supplementary Figure 2. MRM chromatogram corresponding to a pig muscle blank sample (Panel A) and a pig muscle zero sample (Panel B). In each panel: on the left MRM chromatogram corresponding to silibinin, on the right MRM chromatogram corresponding to the internal standard.

Supplementary Table 1. Total and free Silybin levels in plasma, urine, breast cancer, and adjacent unaffected breast tissue in patients who received silybin for 28 days [17].

		Median	Q1	Q3	Min	Max	P-value
TOT-SIL (plasma) ng/mL	Baseline	0	0	0	0	0	
	Before last administration	901	651	1481	214	4574	
	2 hrs after last administration	14538	13147	16828	7654	31121	0.0004¥
SIL (plasma) ng/mL	Baseline	0	0	0	0	0	
	Before last administration	69	13	159	1	523	
	2 hrs after last administration	5847	4526	6454	1818	10861	0.0004¥
TOT-SIL (urine) ng/mL*	Baseline	0	0	0	0	0	
	At surgery	7131	2015	21095	90	26573	
SIL (urine) ng/mL*	Baseline	0	0	0	0	0	
	At surgery	212	71	359	7	747	
TOT-SIL (tissue) ng/g	Tumor	131	35	869	0	1375	
	Normal breast tissue	11	0	34	0	48	0.018
SIL (tissue) ng/g	Tumor	33	4	158	0	177	0.013
	Normal breast tissue	0	0	3.79	0	19	

Twelve breast cancer patients received silybin-phosphatidylcholine, 2.8 g daily for 4 weeks prior to surgery. Silybin levels were measured before (SIL) and after (TOT-SIL) enzymatic hydrolysis by high-performance liquid chromatography (HPLC)-MS/MS in biologic samples (plasma, urine, breast cancer, and surrounding normal tissue). Fasting blood samples were taken at baseline, before the last administration, and 2 hours later. All patients were fully compliant and completed the treatment program.

*: creatinine normalization; end: before the last administration; end-2h: 2 hours after the last administration.

¥ P for the difference between Before last administration and 2 hrs after last administration.