

Improvement in the Biosynthesis of Antioxidant-Active Metabolites in In Vitro Cultures of *Isatis tinctoria* (Brassicaceae) by Biotechnological Methods/Elicitation and Precursor Feeding

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Supplementary Materials

Table S1. Percentage yields of hydroalcoholic extracts obtained from *Isatis tinctoria* shoot cultures grown on different MS medium variants

MS Medium Variant BAP/NAA (mg/L)	Yield of extracts from <i>Isatis tinctoria</i> agitated shoot cultures (%)
2.0/1.0	43.17
1.0/0.5	36.73
1.0/1.0	51.40
1.0/0	49.17
0.5/0	48.71
0/1.0	61.20

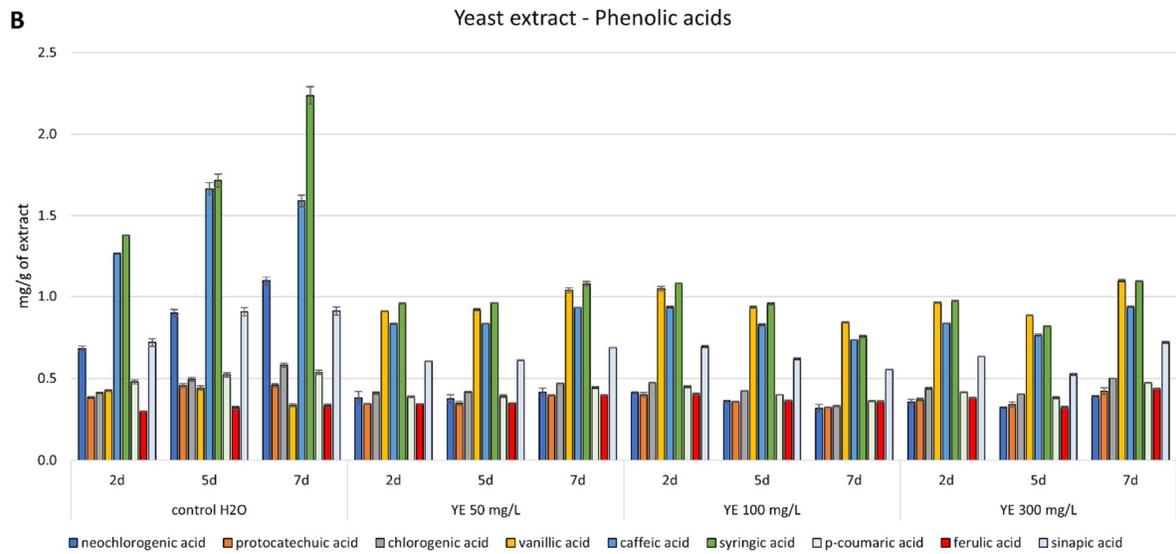
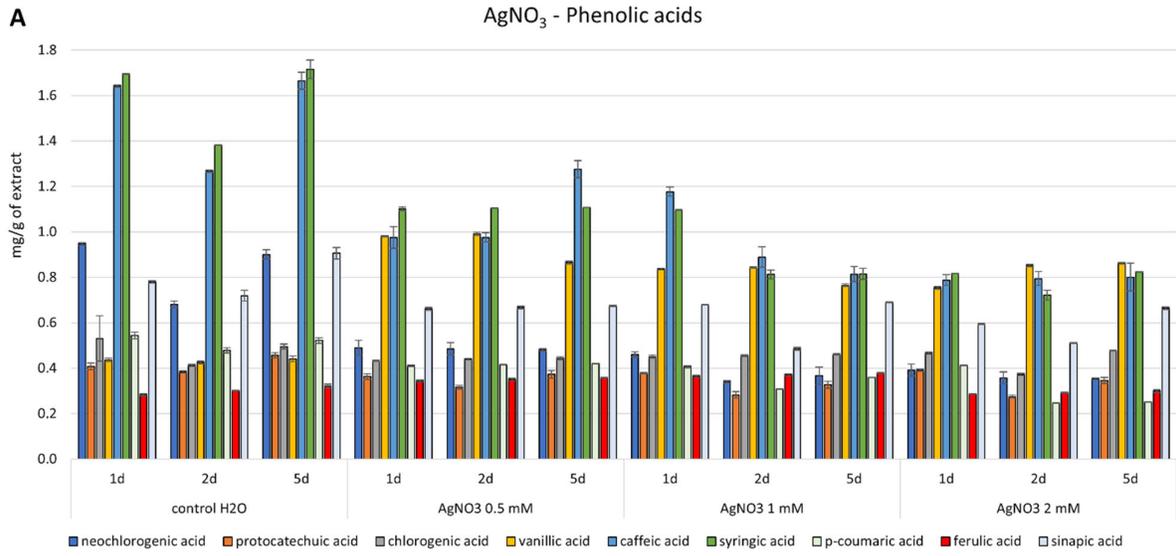
Table S2. Percentage yields of hydroalcoholic extracts obtained from *Isatis tinctoria* elicited shoot cultures (MS variant BAP/NAA 1.0/1.0 mg/L).

Elicitors		Yield of extracts from <i>Isatis tinctoria</i> elicited shoot cultures (%)			
		24 h	48 h	120 h	168 h
MeJa	10 µM	44.38	36.76	33.85	28.66
	50 µM	40.56	33.35	23.23	19.90
	100 µM	41.58	37.28	31.19	11.96
	Ctr EtOH 50%	43.04	33.25	29.46	10.25
CaCl ₂	20 mM	38.37	11.38	16.69	35.82
	50 mM	38.63	17.09	22.73	26.32
	100 mM	42.83	19.05	22.44	30.81
	Ctr H ₂ O	34.28	31.38	30.40	33.23
AgNO ₃	0.5 mM	34.25	18	19.97	
	1 mM	34.21	27.5	20.33	
	2 mM	34.33	22	7	
	Ctr H ₂ O	34.28	31.38	30.40	
YE extract	50 mg/L		30.12	10.18	15.54
	200 mg/L		29.20	29.21	33
	300 mg/L		21.93	40.68	14.12
	Ctr H ₂ O		31.38	30.40	33.23

MeJa – methyl jasmonate; YE – Yeast extract

Table S3. Percentage yields of hydroalcoholic extracts obtained from *Isatis tinctoria* precursors feeding shoot cultures (MS variant BAP/NAA 1.0/1.0 mg/L supplemented with precursor Phe and Tyr).

Precursor (g/L)		Yield of extracts from <i>Isatis tinctoria</i> elicited shoot cultures (%)		
		72 h	120 h	168 h
Phe	1	26.17	40.90	23.90
	2	30.76	38.03	30.80
	Ctr H ₂ O	14.30	30.40	33.23
Tyr	1	50.71	35.06	12.27
	2	26.96	17.54	14
	Ctr H ₂ O	14.30	30.40	33.23



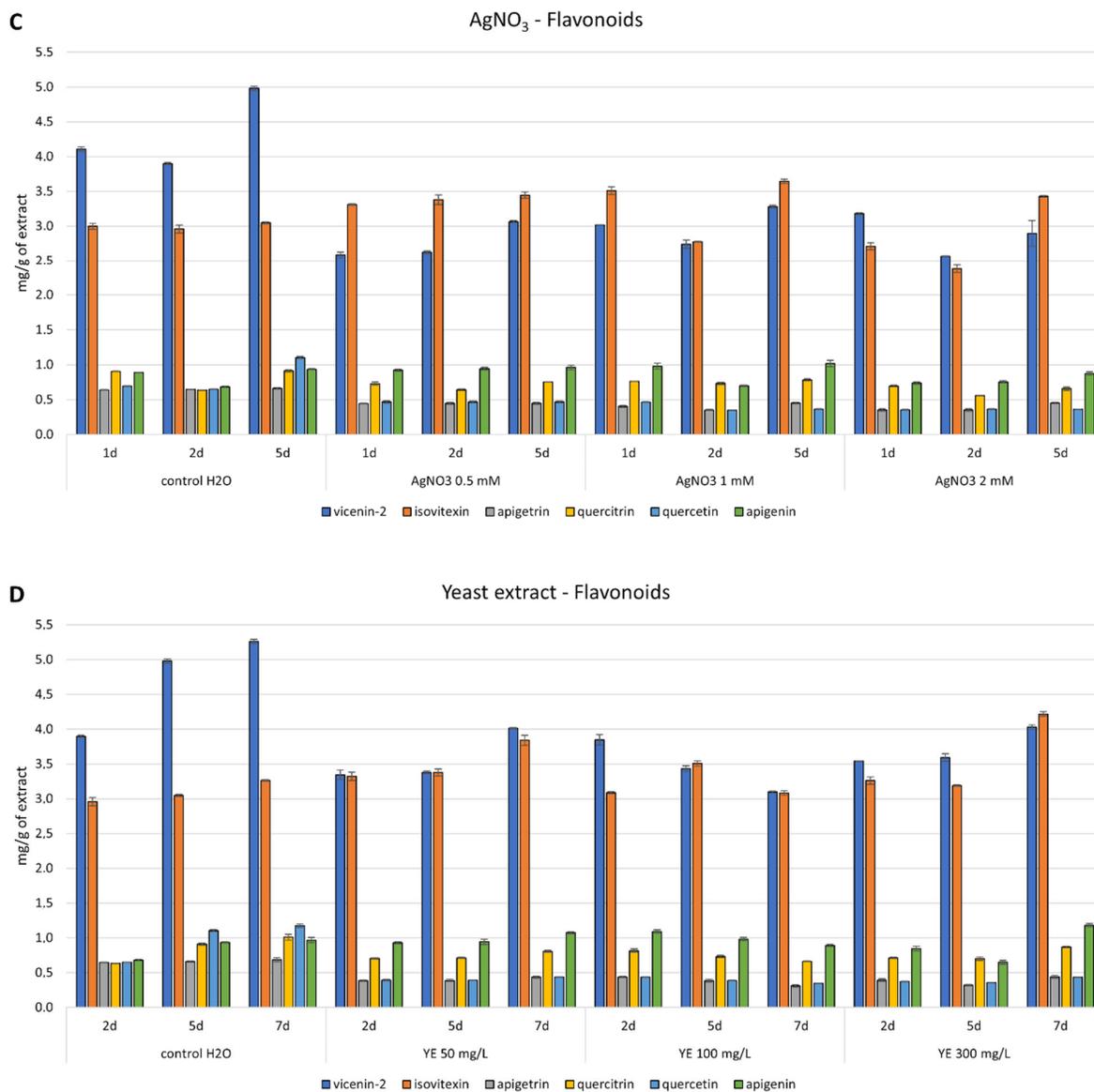


Figure S1. Content of phenolic acids and flavonoids, determined by RP-HPLC analysis, in *Isatis tinctoria in vitro* culture extracts elicited by Silver nitrate - AgNO₃(A, C), and Yeast extract – YE (B, D). Values are expressed as the mean ± SD (n = 3).