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Article

Salicylate and Procyanidin-Rich Stem Extracts of *Gaultheria procumbens* L. Inhibit Pro-Inflammatory Enzymes and Suppress Pro-Inflammatory and Pro-Oxidant Functions of Human Neutrophils *Ex Vivo*

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Supplementary Materials

Table S1. Phenolic analytes detected in *G. procumbens* stem dry extracts by UHPLC-PDA-ESI-MS^3.

Table S2. Correlation (*r*) coefficients and probability (*p*) values of linear relationships between antioxidant and anti-inflammatory activity parameters and phenolic contents of *G. procumbens* stem dry extracts.

Table S3. Antioxidant activity of *G. procumbens* stem dry extracts expressed in phenolic units.

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Table S1. Phenolic analytes detected in *G. procumbens* stem dry extracts by UHPLC-PDA-ESI-MS^3.

Peak	Analyte	Rt (min)	UV λ_{max} (nm)	$[M–H]^-$ m/z	MS^2 (% relative abundance)	MS^3 (% relative abundance)	Extract
1	protocatechuic acid hexoside	3.9	259, 293	315	225 (15); 153 (100); 107 (15)		AE, ME, BE, WE
2	protocatechuic acid *	4.2	259, 293	153			All
3	caffeic acid derivative	4.7	325	305	219 (100); 179 (73); 125 (15)		All
4	3-*O*-caffeoylquinic acid (neochlorogenic acid) *	6.0	325	353	191 (100); 179 (55)		AE, ME, BE, WE
5	3-*O*-*p*-coumaroylquinic acid derivative	6.5	310	371	353 (22); 325 (63); 281 (12); 163 (100)		All
6	procyanidin A-type dimer	9.5	280	575	499 (100); 451 (17); 407 (22); 289 (23)		All
7	procyanidin B-type dimer	10.1	280	577	**425 (100)**; 407 (57); 289 (19)	407 (100); 273 (2)	AE, ME, BE, EAE
8	procyanidin B-type dimer	10.5	280	577	**425 (100)**; 407 (56); 289 (18)	407 (100); 273 (6)	All
9	5-*O*-caffeoylquinic acid (chlorogenic acid, **CHA**) *	10.9	325	353	191 (100); 179 (5)		All
10	(+)-catechin *	11.1	280	289	245 (100); 205 (40)		All
11	unknown compound	11.9	296	385	357 (6); 287 (5); 223 (100)		All
12	methyl salicylate derivative	13.0	285	653 [a]	607 (100); 575 (10)		AE, ME, BE, WE
13	procyanidin B-type dimer	14.1	280	577	**425 (100)**; 407 (59); 289 (29)	407 (100); 273 (8)	AE, ME, WE
14	procyanidin B2 (**PB2**) *	15.3	280	577	**425 (100)**; 407 (53); 289 (14)	407 (100); 273 (7)	All
15	unknown compound	15.7	280	461	415 (100); 167 (9)		All
16	protocatechuic acid derivative	16.1	259, 293	481	463 (7); 345 (72); 327 (100); 165 (28); 153 (62)		All
17	(–)-epicatechin (**ECA**) *	16.8	280	289	245 (100); 205 (24)		All
18	gaultherin (**GT**) *	17.7	285	491 [a]	445 (15); 413 (3); 293 (72); 233 (3); 149 (5)		All
19	procyanidin A-type trimer	18.3	280	863	**711 (100)**; 573 (19); 559 (15); 451 (30); 411 (34); 289 (13)	693 (76); 559 (69); 541 (24); 407 (12)	All
20	procyanidin B-type dimer	18.7	280	577	**425 (100)**; 407 (40); 289 (52)	407 (100); 273 (10)	All
21	procyanidin A-type dimer	19.5	280	575	499 (20); 491 (22); 451 (25); **425 (100)**; 407 (96); 289 (36)	407 (100); 273 (6)	AE, ME, BE, EAE
22	procyanidin A-type trimer (**PAT**)	20.3	280	863	**711 (100)**; 693 (14); 573 (25); 559 (16); 451 (23); 411 (24); 289 (7)	693 (64); 559 (100); 541 (27); 463 (10); 407 (16); 285 (4)	All

Table S1. *Cont.*

Peak	Analyte	Rt (min)	UV λ_{max} (nm)	$[M–H]^-$ m/z	MS^2 (% relative abundance)	MS^3 (% relative abundance)	Extract
23	procyanidin B-type trimer	21.3	280	865	847 (25); 739 (73); **713 (41)**; 695 (77); 577 (98); 451 (21); 287 (17)	695 (100); 575 (22); 561 (23); 407 (22); 243 (13)	All
24	procyanidin B-type dimer	22.1	280	577	**425 (100)**; 407 (44); 289 (22)	407 (100); 273 (11)	All
25	procyanidin A-type dimer	22.6	280	575	499 (69); 491 (24); 451 (22); **425 (100)**; 407 (79); 289 (25)	407 (100); 273 (9)	All
26	lyoniresinol hexoside	22.8	280	581	565 (16); **419 (100)**; 401 (12); 373 (15)	404 (100); 373 (16)	All
27	procyanidin B-type trimer	23.6	280	865	847 (21); 739 (47); **713 (51)**; 695 (92); 577 (99); 451 (52); 287 (54)	695 (100); 575 (24); 561 (26); 407 (25); 243 (10)	All
28	procyanidin B-type dimer	25.0	280	577	**425 (100)**; 407 (19); 287 (51)	407 (100); 273 (17)	AE, ME, WE
29	unknown compound	25.1	278	597	577 (10); **553 (100)**	536 (2); 419 (100); 389 (24); 233 (13)	All
30	unknown compound	25.8	278	567	**521 (100)**	359 (100); 344 (96); 329 (28)	AE, ME, BE
31	procyanidin A-type dimer	26.5	280	575	499 (27); 451 (22); **425 (100)**; 411 (57); 289 (7)	407 (100); 273 (7)	AE, ME, BE, WE
32	procyanidin B-type trimer	27.1	280	865	577 (82); 407 (26); 289 (18)		AE, ME, BE, WE
33	quercetin 3-*O*-β-D-galactopyranoside (hyperoside, **HY**) *	27.6	254, 353	463	**301 (100)**	273 (38); 255 (20); 179 (100); 151 (64)	All
34	quercetin 3-*O*-β-D-glucopyranoside (isoquercitrin, **IQ**) *	28.6	256, 353	463	**301 (100)**	273 (54); 255 (22); 179 (100); 151 (76)	All
35	quercetin 3-*O*-β-D-glucuronopyranoside (miquelianin, **MQ**) *	29.1	256, 356	477	**301 (100)**	273 (29); 257 (19); 179 (100); 151 (58)	All
36	procyanidin A-type dimer	29.5	280	575	449 (41); **423 (100)**; 289 (19)	405 (8); 285 (100); 257 (5)	All
37	quercetin 3-*O*-α-L-arabinopyranoside (guaijaverin, **GV**) *	30.8	258, 356	433	**301 (100)**	273 (44); 255 (21); 179 (100); 151 (68)	AE, ME, BE, EAE
38	unknown compound	31.9	278	567	521 (100); 179 (33)		All
39	quercetin (**QU**) *	43.7	255, 364	301	273 (36); 255 (21); 179 (100); 151 (69)		All
40	kaempferol (**KA**) *	50.4	265, 364	285	267 (45); 257 (100); 229 (80); 163 (15); 151 (18)		AE, BE, EAE

Rt, retention times. UV λ_{max}, absorbance maxima in PDA spectra. $[M–H]^-$, pseudomolecular ions in MS spectra recorded in a negative ion mode. In bold – ions subjected to MS^3 fragmentation. Compounds marked with an asterisk (*) were identified with authentic standards. [a] $[M+HCOO]^-$.

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Table S2. Correlation (*r*) coefficients and probability (*p*) values of linear relationships between antioxidant and anti-inflammatory activity parameters and phenolic contents of *G. procumbens* stem dry extracts.

***r* (*p*) for:**	**Antioxidant activity**						**Anti-inflammatory activity**		
	DPPH	**FRAP**	**TBARS**	**$O_2^{\bullet-}$**	**$^{\bullet}OH$**	**H_2O_2**	**HYAL**	**LOX**	**COX-2**
TPC	–0.9523 (0.048) *	0.8857 (0.114)	–0.7312 (0.269)	0.1236 (0.876)	0.2291 (0.771)	–0.9127 (0.087)	–0.6998 (0.300)	–0.4797 (0.520)	–0.9729 (0.027) *
TPH	–0.9171 (0.083)	0.9773 (0.023) *	–0.8302 (0.170)	–0.2958 (0.704)	–0.1750 (0.825)	–0.8491 (0.151)	–0.7303 (0.270)	–0.7638 (0.236)	–0.8661 (0.134)
TPA	–0.3806 (0.619)	0.3443 (0.656)	–0.7292 (0.271)	–0.3806 (0.619)	–0.8987 (0.101)	–0.3804 (0.620)	–0.6502 (0.350)	–0.9257 (0.074)	–0.2147 (0.785)
TLPA	–0.7476 (0.252)	0.8691 (0.131)	–0.8364 (0.164)	–0.6647 (0.335)	–0.5705 (0.429)	–0.6772 (0.323)	–0.7005 (0.299)	–0.9448 (0.055)	–0.6300 (0.370)
TPHA	–0.6748 (0.325)	0.9158 (0.084)	–0.6978 (0.302)	–0.6378 (0.362)	–0.4982 (0.502)	–0.5740 (0.426)	–0.5322 (0.468)	–0.8490 (0.151)	–0.5703 (0.430)
TSAL	–0.9161 (0.084)	0.9419 (0.058)	–0.6902 (0.310)	0.0383 (0.962)	0.1726 (0.827)	–0.8533 (0.147)	–0.6265 (0.374)	–0.5029 (0.497)	–0.9266 (0.073)
TFL	0.1608 (0.839)	–0.5707 (0.429)	–0.1858 (0.814)	–0.1645 (0.836)	–0.3802 (0.620)	0.0308 (0.969)	–0.3085 (0.691)	–0.1445 (0.855)	0.2200 (0.780)

Activity and concentration parameters according to Figs. 2 and 3. Asterisk mean significance of the estimated linear relationship (* $p < 0.05$) for four extracts (data points $n = 4$).

Table S3. Antioxidant activity of *G. procumbens* stem dry extracts expressed in phenolic units.

Analyte	DPPH	FRAP	TBARS	$O_2^{\bullet -}$	$^{\bullet}OH$	H_2O_2
	SC_{50} (μg GAE /mL) [a]	mmol Fe^{2+}/g GAE [b]	IC_{50} (μg GAE/mL) [c]	SC_{50} (μg GAE/mL) [a]	SC_{50} (μg GAE/mL) [a]	SC_{50} (μg GAE/mL) [a]
AE	1.97	21.99	2.33	7.80	51.90	11.48
ME	1.94	19.94	2.16	8.00	41.06	10.49
BE	2.18	19.45	3.99	11.37	58.99	12.62
WE	2.14	22.68	3.72	6.12	36.92	13.55

[a] Scavenging efficiency (amount of antioxidant needed to decrease the initial concentration of the oxidant by 50%) expressed in μg of phenolics/mL of the DPPH solution (values obtained by converting the original SC_{50} values using the TPC levels); [b] values expressed per g of phenolics (obtained by converting the original FRAP values using the TPC levels); [c] inhibition concentration (amount of antioxidant needed to decrease linoleic acid peroxidation and formation of TBARS by 50%) expressed in μg of phenolics/mL of the substrate solution (values obtained by converting the original IC_{50} values using the TPC levels). For original TPC, FRAP, SC_{50} and IC_{50} parameters see Table 1 and Fig. 2.