Castanea sativa Mill. leaf: UHPLC-ESI-Q*q*TOF analysis and effects on *in vitro* rumen fermentation and methanogenesis

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Supplement Materials

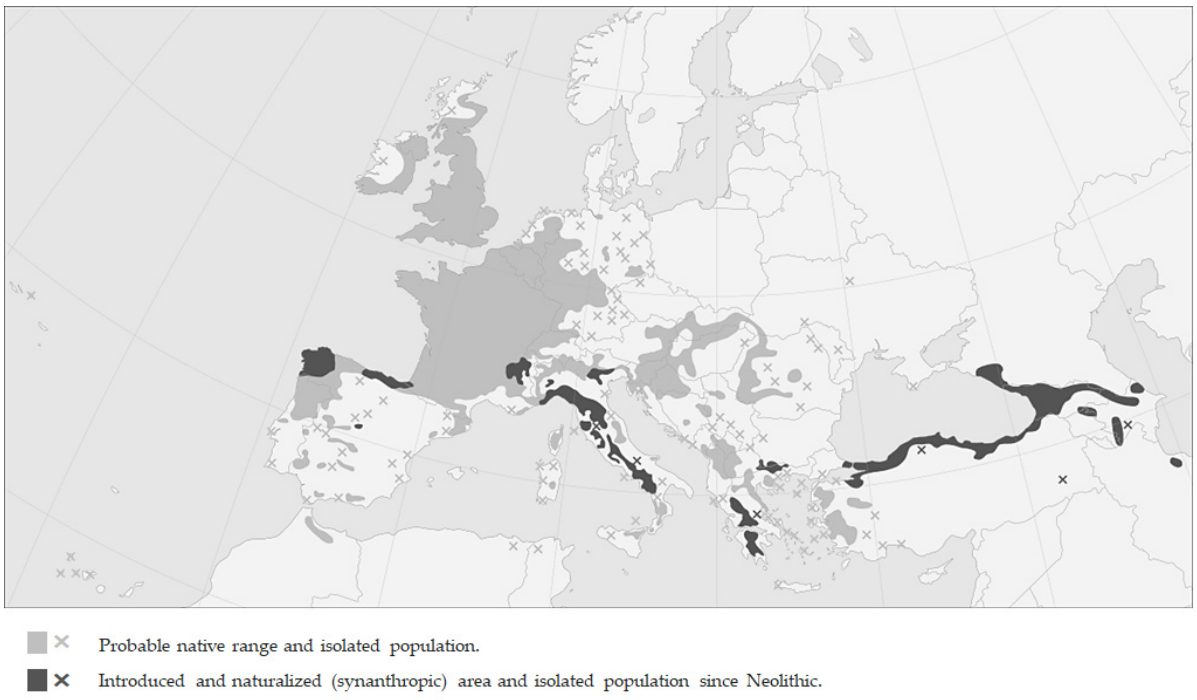

Figure S1. Worldwide distribution of Castanea sativa Mill. (readapted from https://www.euforgen.org/species/castanea-sativa/)

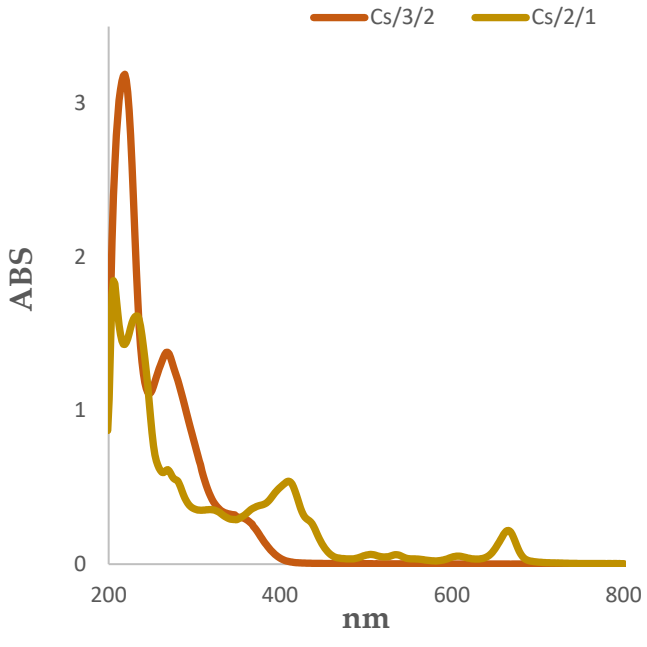

Figure S2. UV/Visible spectra of fractions Cs/2/1 and Cs/3/2.

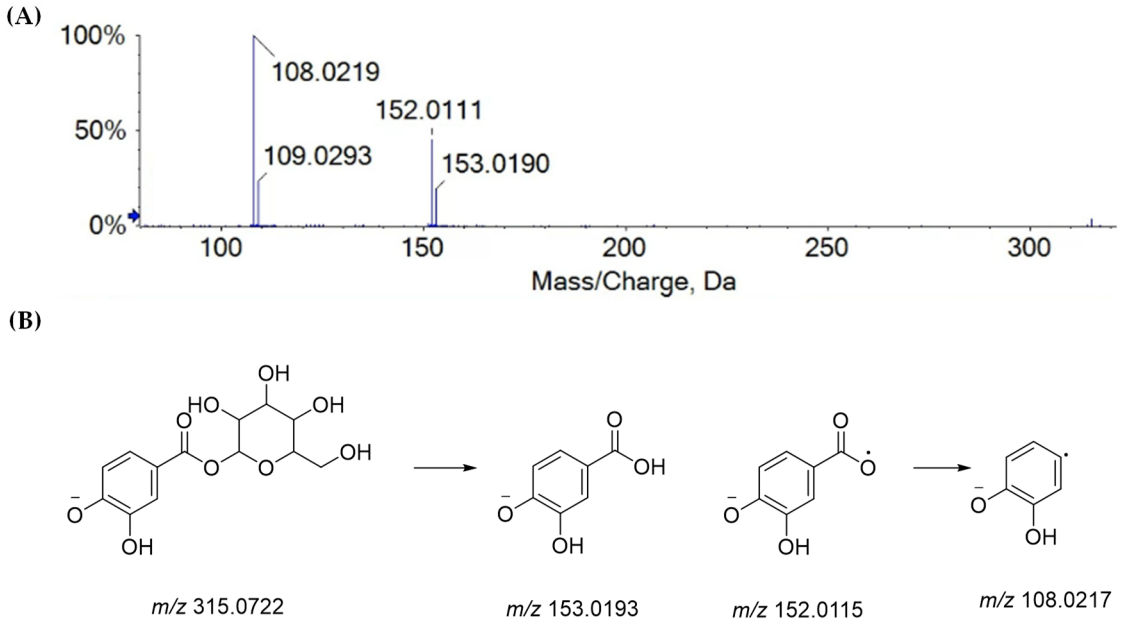

Figure S3. (A) TOF-MS/MS spectrum of compound **10**, **(B)** putative fragmentation patterns; theoretical mass is reported under each structure.

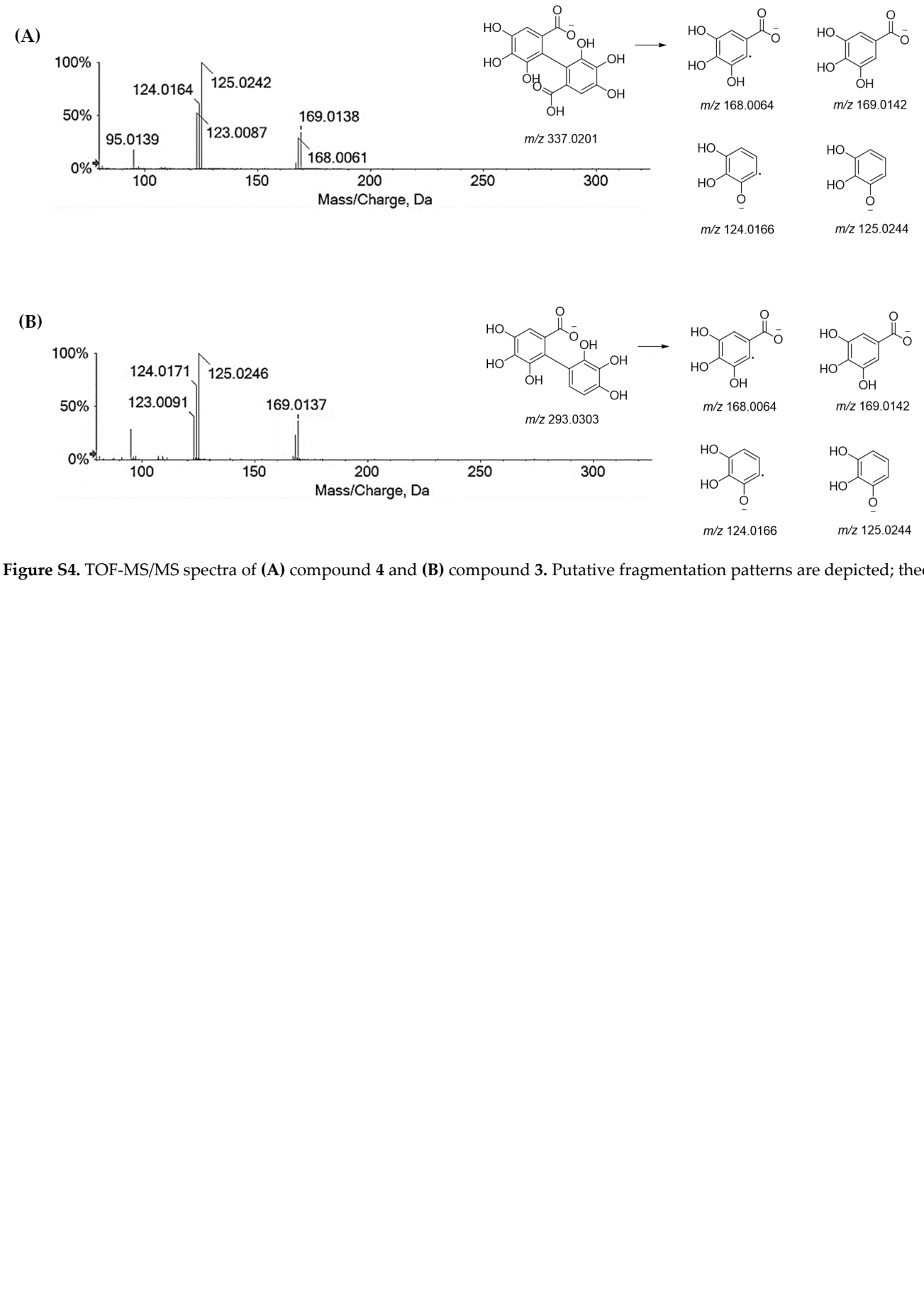

Figure S4. TOF-MS/MS spectra of **(A)** compound **4** and **(B)** compound **3.** Putative fragmentation patterns are depicted; theoretical mass is reported under each structure.

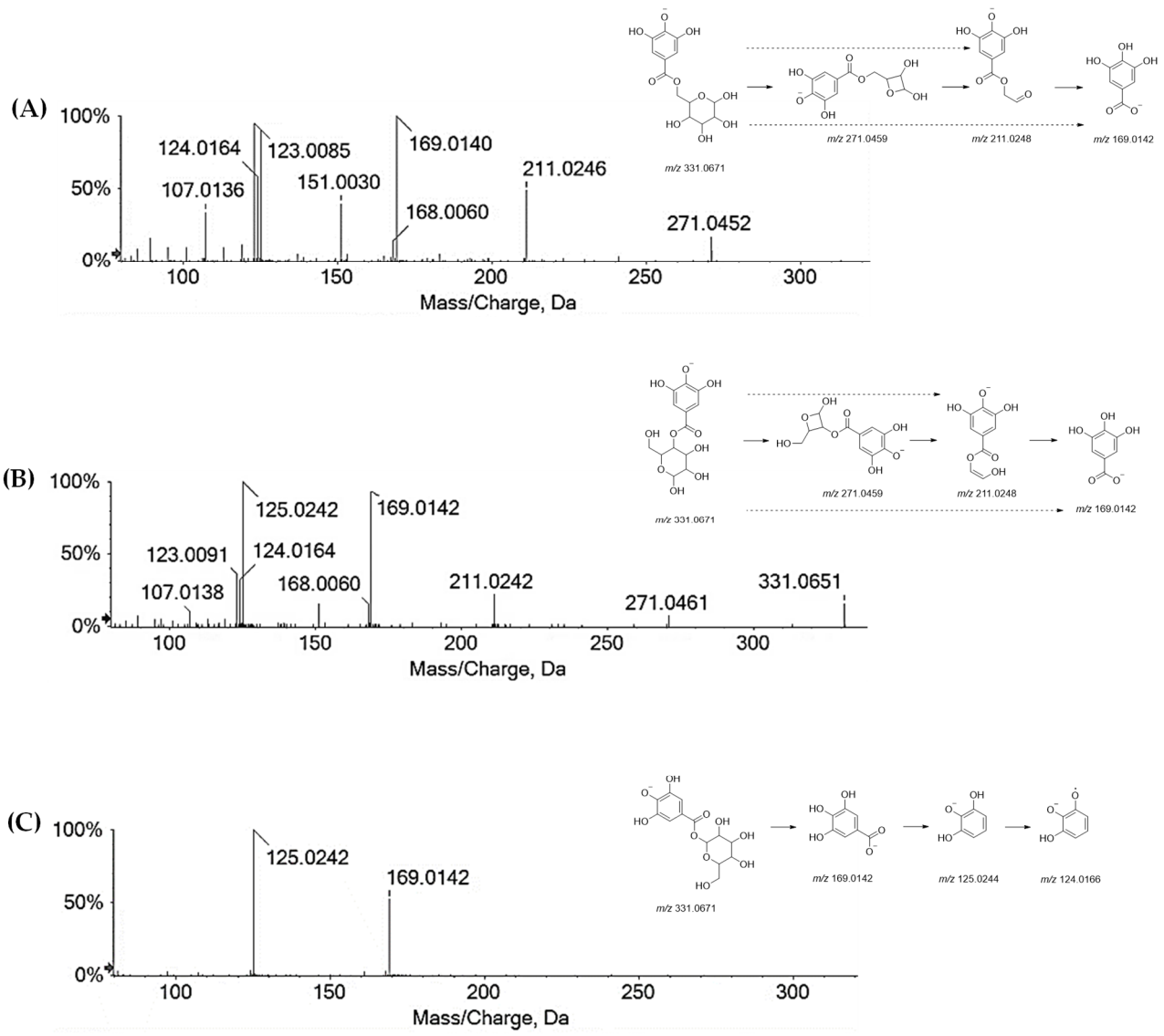

Figure S5. Galloyl hexose isomers (**2, 6, 12**) TOF-MS/MS spectra. **(A)** Compound **2**; **(B)** compound **6**; **(C)** compound **12**. Putative fragmentation patterns are depicted; theoretical mass is reported under each structure.

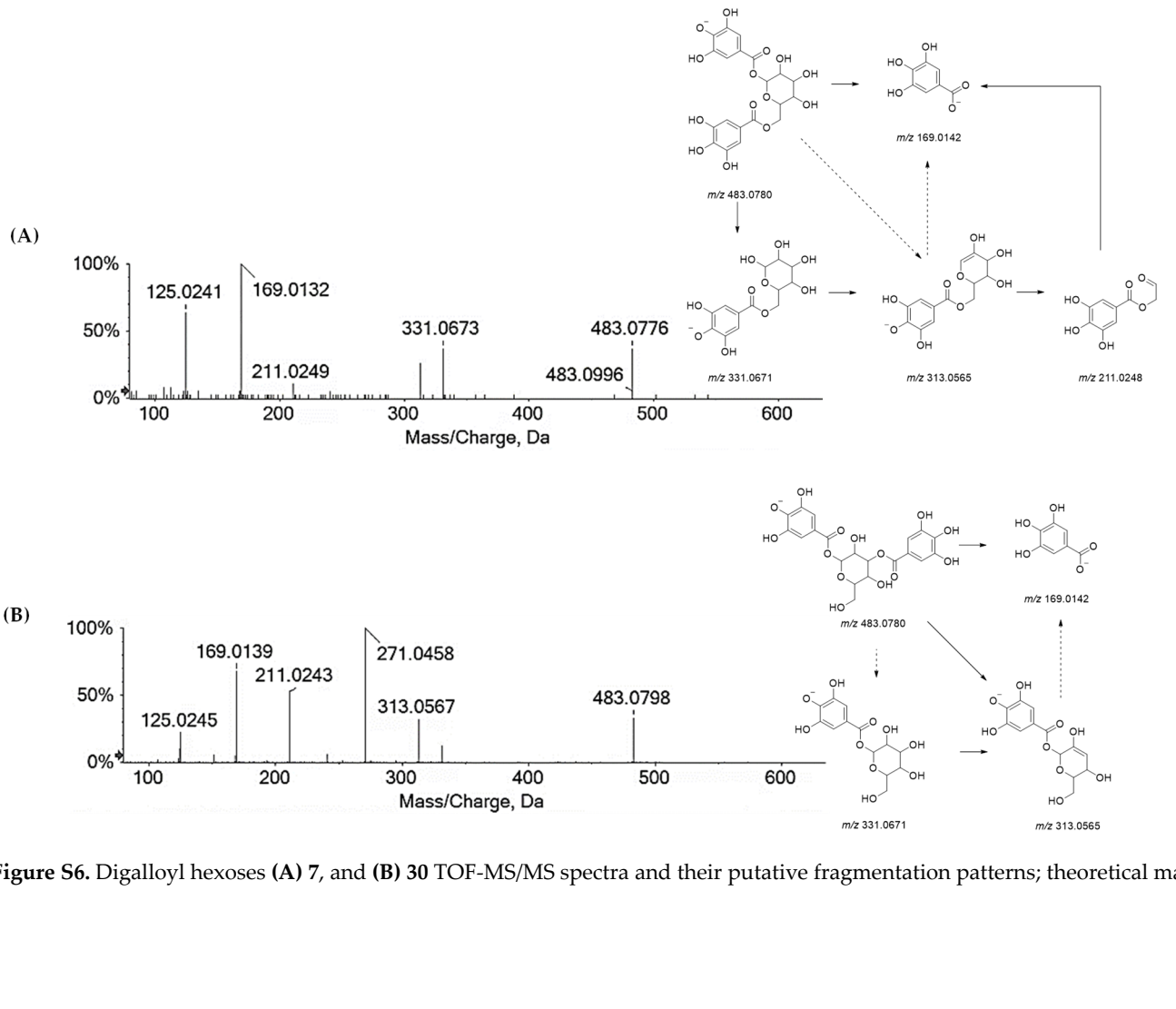

Figure S6. Digalloyl hexoses **(A) 7**, and **(B) 30** TOF-MS/MS spectra and their putative fragmentation patterns; theoretical mass is reported under each structure.

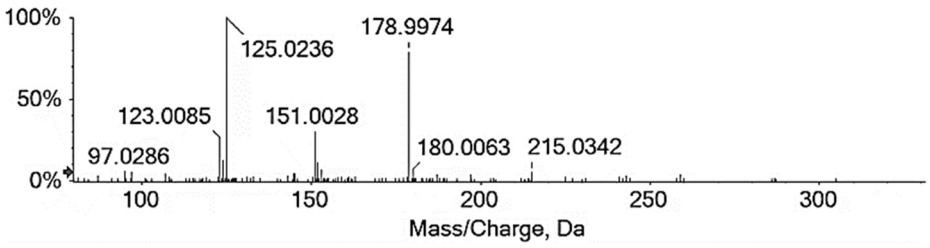

Figure S7. TOF-MS/MS spectra of compound **17**, tentatively identified as galloyl dihydroxybenzoic acid.

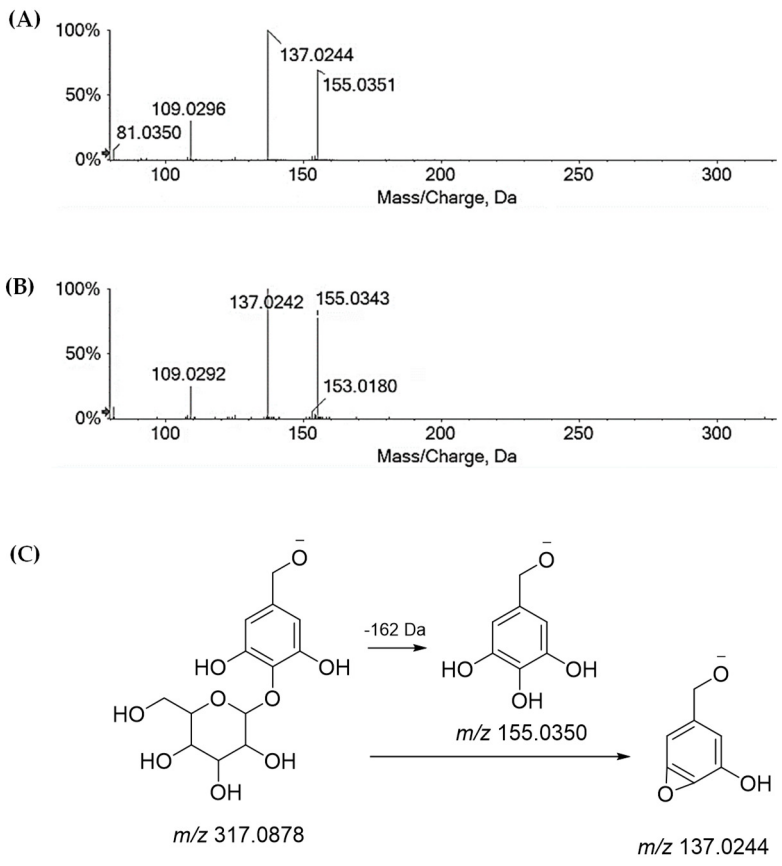

Figure S8. TOF-MS/MS spectra of compounds **8 (A)** and **11 (B)**. In panel **(C)** the putative fragmentation pattern of compound **8** is depicted.

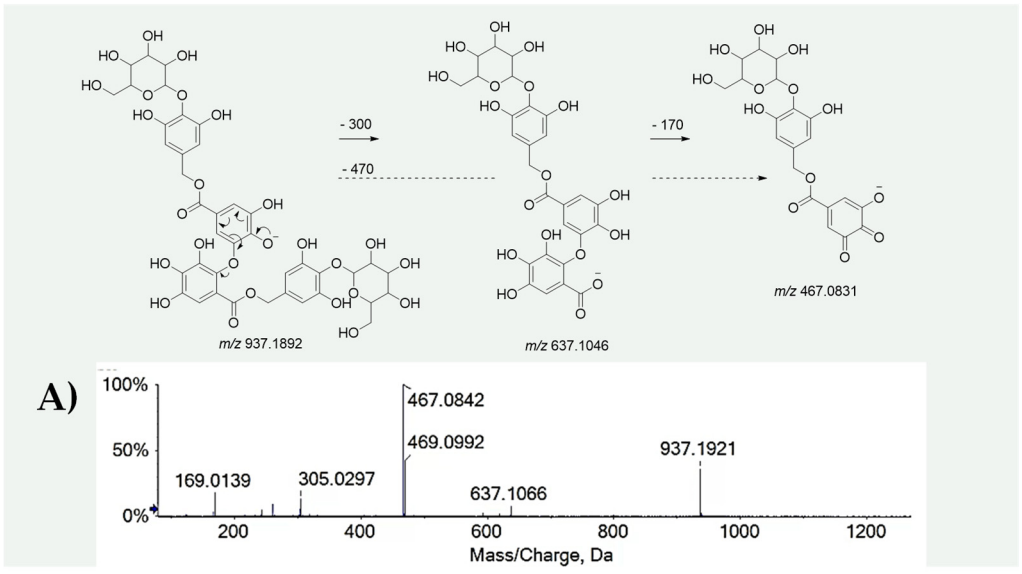

Figure S9. TOF-MS/MS spectra of **(A)** chestanin **(49)** and **(B)** isochestanin **(58).** Putative fragmentation patterns are depicted; theoretical mass is reported under each structure.

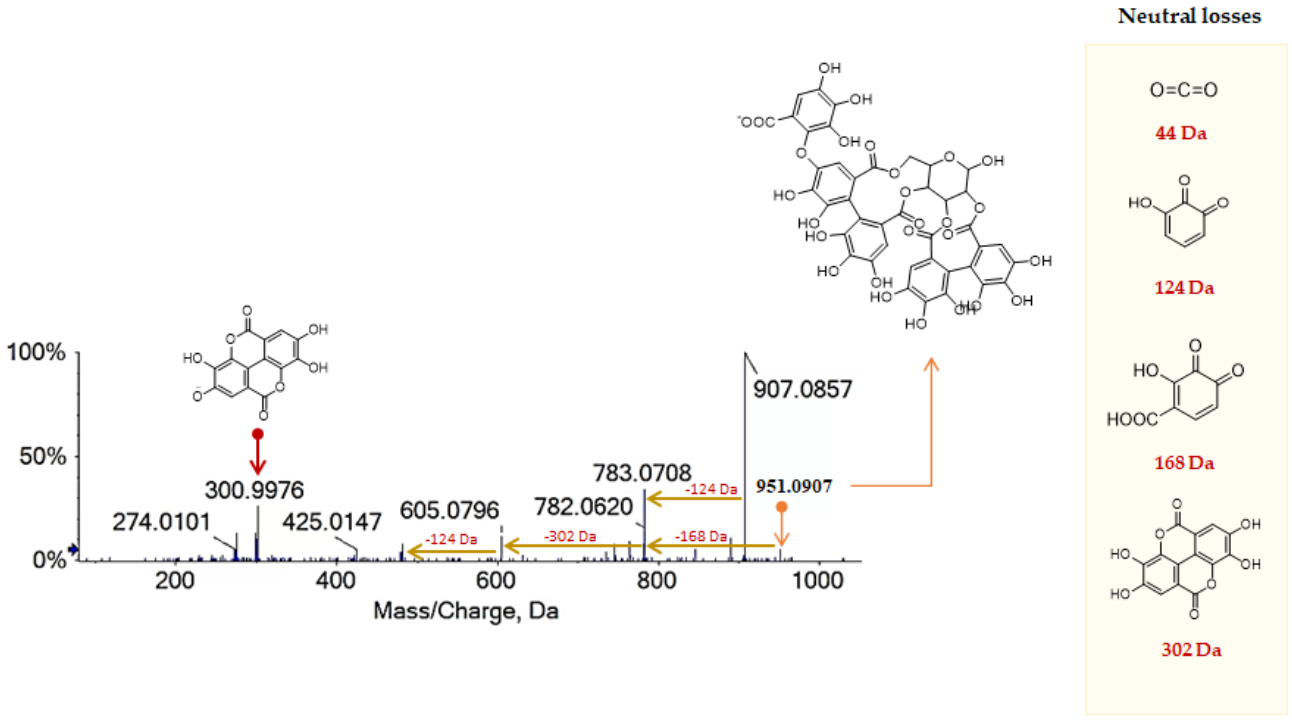

Figure S10. TOF-MS/MS spectra of compound **23** with $[M-H]^-$ ion at *m/z* 951.0907. Structures of observed neutral losses are depicted.

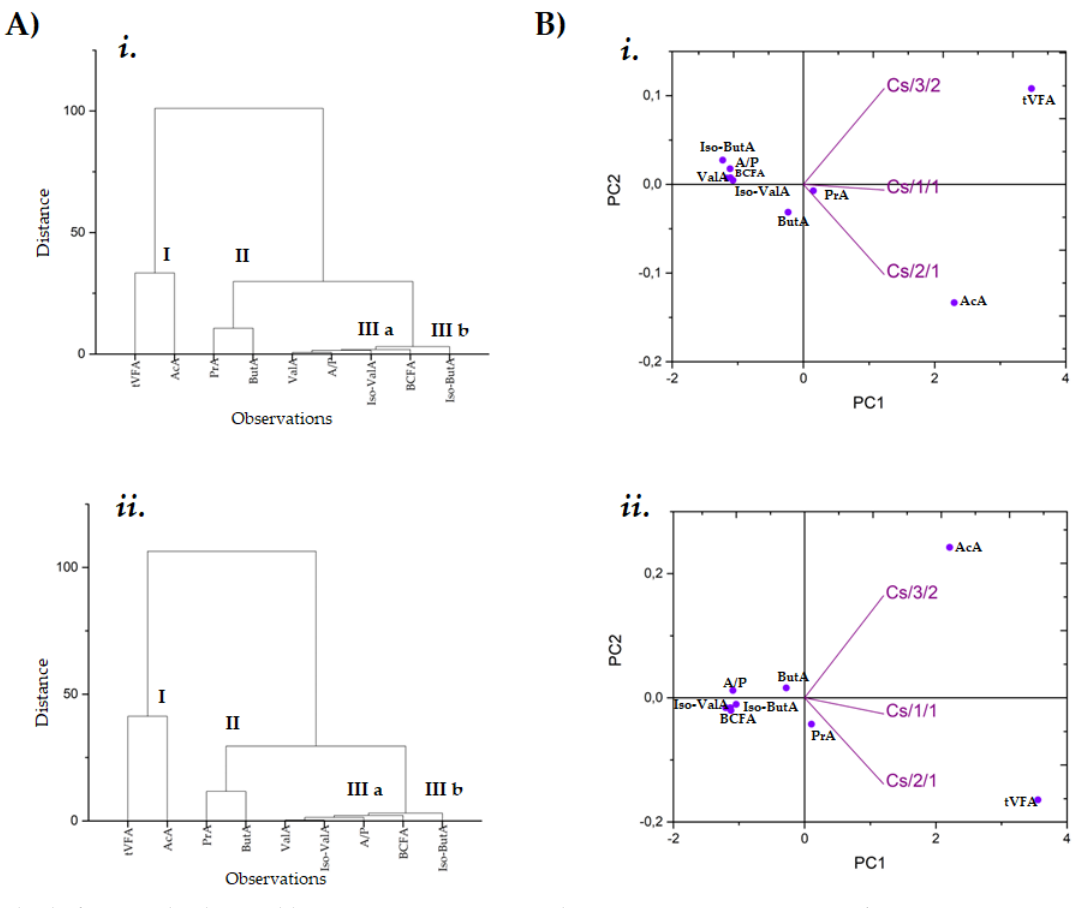

Figure S11. (**A**) Dendrograms of different volatile fatty acids obtained by treatment at 50 (***i.***) and 200 mg (***ii.***); (**B**) (***i.***) PCA (% variance on PC1 99.8; on PC2 0.2) of VFAs at 50 mg-dose level; (***ii.***) PCA (% variance on PC1 99.6; on PC2 0.4) of VFAs 200 of Cs/1/1, Cs/2/1 and Cs/3/2 fractions.

Table S1. Compounds tentatively identified in the chestnut Cs/1/1 alcoholic extract and its Cs/2/1 and Cs/3/2 fractions. Rt = retention time; RDB = ring double bond equivalent value. Base peak fragments are reported in bold.

Peak	Rt	Tentative assignment	Formula	$[M-H]^-$ found (*m*/*z*)	error (ppm)	RDB	MS/MS fragment ions (*m*/*z*) and relative intensity
1	0.288	Quinic acid	$C_7H_{12}O_6$	191.0562	0.5	2	191.0543; 127.0296; 93.0343; 87.0086; **85.0295**
2	0.602	Galloyl hexose I	$C_{13}H_{16}O_{10}$	331.0670	-0.2	6	331.0675; 271.0452; 211.0246; 169.0140; 151.0030; **125.0243**; 124.0164; 107.0136
3	0.680	Pyrogallol gallic acid	$C_{13}H_{10}O_8$	293.0302	0.1	9	169.0130; 168.0056; **125.0238**; 124.0161; 123.0081; 95.0132
4	0.680	Hexahydroxydiphenic acid	$C_{14}H_{10}O_{10}$	337.0204	0.8	10	169.0142; **125.0243**; 124.0163; 95.0137
5	0.721	Gallic acid	$C_7H_6O_5$	169.0151	5.0	5	**125.0239**; 124.0159
6	0.776	Galloyl hexose II	$C_{13}H_{16}O_{10}$	331.0669	-0.5	6	331.0651; 271.0461; 211.0242; 169.0142; 151.0034; **125.0242**; 124.0164; 107.0138
7	1.008	Digalloyl hexose	$C_{20}H_{20}O_{14}$	483.0785	1.0	11	483.0776; 331.0673; 313.0544; 211.0249; **169.0132**; 125.0241
8	1.124	3,4,5-trihydroxybenzyl hexoside I (e.g. crenatin)	$C_{13}H_{18}O_9$	317.0879	0.3	5	155.0340; **137.0242;** 109.0293
9	1.163	NHTP-HHDP-hexose I (e.g.,castalagin/vescalagin)	$C_{41}H_{26}O_{26}$	933.0640; 466.0328 $[M-2H]^{2-}$	1.8	29	**933.0680;** 915.0558; 871.0690; 631.0606; 569.0601; 467.0282; 300.9986
10	1.260	Dihydroxybenzoic acid hexoside	$C_{13}H_{16}O_9$	315.0724	0.8	6	152.0111; 109.0293; **108.0219**
11	1.417	3,4,5-trihydroxybenzyl hexoside II	$C_{13}H_{18}O_9$	317.0877	-0.3	5	155.0343; **137.0242**; 109.0292
12	1.535	Galloyl hexose III	$C_{13}H_{16}O_{10}$	331.0674	1.0	6	169.0142; **125.0244**
13	1.632	Bis-HHDP-hexose I (e.g.,pedunculagin/casuariin)	$C_{34}H_{24}O_{22}$	783.0700	1.7	23	783.0735; 481.0643; **300.9993**; 275.0199
14	1.768	Galloyl shikimic acid	$C_{14}H_{14}O_9$	325.0554	-3.4	8	325.0520; **169.0132**; 125.0273;124.0161
15	1.827	NHTP-HHDP-hexose II (e.g.,castalagin/vescalagin)	$C_{41}H_{26}O_{26}$	933.0641; 466.0326 $[M-2H]^{2-}$	0.2	29	613.0440; 467.0292; **300.9996**; 299.9908; 275.0205; 257.0080; 229.0135; 169.0138
16	2.087	Galloyl shikimic acid	$C_{14}H_{14}O_9$	325.0563	-0.6	8	271.1001; 169.0130; **125.0235**; 124.0159; 93.0351
17	2.187	Galloyl dihydroxybenzoic acid	$C_{14}H_{10}O_8$	305.0305	0.7	10	178.9974; 151.0028; **125.0236**
18	2.385	Galloyl shikimic acid	$C_{14}H_{14}O_9$	325.0566	-0.1	8	325.0532; 169.0141; **125.0237**; 124.0265; 107.0138
19	3.314	Bis-HHDP-hexose II (e.g.,pedunculagin/casuariin)	$C_{34}H_{24}O_{22}$	783.0696	1.2	23	783.0720; 481.0624; **300.9984**; 275.0188; 249.0396; 169.013
20	3.929	*p*-Coumaric acid hexoside I	$C_{15}H_{18}O_8$	325.0926	-0.9	7	163.0470; **119.0481**
21	4.589	Galloyl-chebuloyl-HHDP-hexose I (e.g., chebulagic acid)	$C_{41}H_{30}O_{27}$	953.0901	-0.1	27	953.0915; 909.1024; **785.0858**; 765.0585; 425.0136; 300.9979; 299.9902; 275.0186
22	4.609	*p*-Coumaric acid hexoside II	$C_{15}H_{18}O_8$	325.0926	-0.9	7	163.0390; **119.0502**
23	4.966	HHDP-valoneoyl-hexose	$C_{41}H_{28}O_{27}$	951.0761	1.7	28	951.0875; **907.0857;** 889.0935; 783.0708; 605.0796; 481.0693; 425.0147; 300.9976
24	4.988	Caffeic acid hexoside	$C_{15}H_{18}O_9$	341.0871	-1.2	7	179.0329; **135.0447**
25	5.268	Digalloyl-HHDP-hexose I	$C_{34}H_{26}O_{22}$	785.0857	1.8	22	785.0889; 615.0645; 483.0799; 419.0620; **300.9987;** 275.0193; 249.0400
26	5.628	5-*O*-Caffeoyl quinic acid	$C_{16}H_{18}O_9$	353.0884	1.7	8	**191.0560**; 85.0299
27	6.004	Galloyl-HHDP-hexose	$C_{27}H_{22}O_{18}$	633.0742	1.8	17	633.0739; 463.0520; **300.9978**; 275.0179; 249.0408
28	6.004	Galloyl-bis-HHDP-hexose I (e.g., stachyurin/casuarinin)	$C_{41}H_{28}O_{26}$	935.0818	2.1	28	**935.0792**; 917.0989; 873.0778; 855.0683; 783.0718; 633.0721; 571.0715; 300.9969; 299.0169; 275.0179

29	6.244	Methylvaloneoyl–NHTP–hexose (e.g.,vescavaloninic/castavaloninic acid methyl ester)	$C_{49}H_{32}O_{31}$	557.0405 $[M-2H]^{2-}$	1.8	34	933.0706; 631.0572; 466.0307; 425.0188; 300.9994; 275.0215; **181.0134**; 153.0201
30	6.165	Digalloyl hexose	$C_{20}H_{20}O_{14}$	483.0788	1.6	11	483.0798; 331.0666; 313.0567; **271.0458;** 211.0243; 169.0139; 125.0245
31	6.560	Galloyl-bis-HHDP-hexose II (e.g., stachyurin/casuarinin)	$C_{41}H_{28}O_{26}$	935.0801	0.5	28	**935.0826**; 917.0730; 873.0830; 783.0713; 633.0740; 571.0753; 419.0628; 300.9979; 299.0187; 275.0184
32	6.500	Galloyl-chebuloyl-HHDP-hexose II (e.g., chebulagic acid)	$C_{41}H_{30}O_{27}$	953.0909	0.8	27	**953.0966**; 935.0828; 909.1031; 785.0875; 633.0771; 300.9982; 275.0194
33	6.546	Trigalloyl hexose I	$C_{27}H_{24}O_{18}$	635.0892	0.3	16	635.0875; 483.0810; 465.0678; **313.0540**; 169.0131
34	6.819	Chesnatin	$C_{27}H_{26}O_{18}$	637.1064	2.8	15	637.1036; 469.1002; **467.0857**; 305.0299; 260.0327; 169.0143; 166.9984
35	7.080	3-O-*p*-Coumaroyl quinic acid	$C_{16}H_{18}O_{8}$	337.0925	-1.2	8	**191.0561**; 173.0450; 119.0500; 93.0342
36	7.218	3-O-*p*-Coumaroyl quinic acid	$C_{16}H_{18}O_{8}$	337.0933	1.2	8	**191.0556**; 173.0450; 119.0503; 93.0345
37	7.297	Digalloyl-HHDP-hexose II	$C_{34}H_{26}O_{22}$	785.0856	1.7	22	785.0884; 633.0755; 483.0795; **300.9991**; 275.0196; 249.0403
38	7.417	Digalloyl deoxyhexose	$C_{20}H_{20}O_{13}$	467.0843	2.5	11	467.0840; 449.0731; **423.0933**; 315.0715; 297.0611; 169.0137; 152.0112; 125.0238
39	8.110	Isochesnatin	$C_{27}H_{26}O_{18}$	637.1063	2.6	15	593.1180; 469.1004; 293.0301; 261.0301; **169.0142**
40	8.327	Galloyl phenol hexoside I (e.g.,cretanin)	$C_{20}H_{22}O_{13}$	469.0996	1.8	10	**169.0150**; 125.0250
41	8.581	Galloyl-bis-HHDP-hexose III	$C_{41}H_{28}O_{26}$	935.0797; 467.0375 $[M-2H]^{2-}$	0.1	28	391.0301; **300.9982**; 299.9900; 275.0184; 169.0137
42	8.721	Galloyl phenol hexoside II	$C_{20}H_{22}O_{13}$	469.0997	2.0	10	**169.0142**; 125.0244
43	8.919	Trigalloyl hexose II	$C_{27}H_{24}O_{18}$	635.0905	2.4	16	635.0923; **483.0795**; 465.0689; 313.0571; 271.0450; 169.0140
44	9.000	Trigalloyl-valoneoyl-HHDP-hexose (e.g.,rugosin A)	$C_{48}H_{34}O_{31}$	552.0490 $[M-2H]^{2-}$	1.1	32	937.1942; 785.0853; 767.9712; 749.0615; 615.0636; 425.0152; **300.9987**; 299.9907; 169.0137; 125.0242
45	9.455	Trigalloyl-HHDP-hexose	$C_{41}H_{30}O_{26}$	937.0947; 468.0454 $[M-2H]^{2-}$	-0.6	27	767.0768; 615.0654; 463.0522; **300.9995**; 283.9963; 275.0199; 245.0086; 231.0301; 169.0149; 125.0253
46	9.595	Quercetin pentosyl-hexoside	$C_{26}H_{28}O_{16}$	595.1318	2.3	13	595.1314; 343.0462; 301.0346; **300.0268;** 271.0232
47	9.704	Tetragalloyl hexose	$C_{34}H_{28}O_{22}$	787.1007	1.0	21	787.1060; **635.0927**; 617.0824; 573.0914; 465.0691; 313.0555; 295.0452; 169.0141
48	9.704	Ellagic acid pentoside	$C_{19}H_{14}O_{12}$	433.0415	0.6	13	433.0398; 300.9979; **299.9902;** 244.0003
49	9.754	Chestanin	$C_{40}H_{42}O_{26}$	937.1894	0.3	20	937.1921; 637.1066; 469.0992; **467.0824**; 305.0297; 169.0139
50	9.969	Quercetin hexuronide I	$C_{21}H_{18}O_{13}$	477.0686	2.6	13	477.0687; **301.0356**; 283.0236; 255.0294; 227.0338; 151.0036
51	9.911	Quercetin 3-*O*-hexoside I	$C_{12}H_{20}O_{12}$	463.0893	2.4	12	463.0894; 301.0351; **300.0276**; 271.0243; 255.0295; 151.0033
52	10.009	Rutin	$C_{27}H_{30}O_{16}$	609.1479	2.9	13	609.1484; 301.0352; **300.0277**; 271.0244; 2550296
53	10.067	Quercetin 3-*O*-hexoside II	$C_{12}H_{20}O_{12}$	463.0892	2.2	12	463.0901; 301.0357; **300.0280**; 271.0252; 255.0299; 243.0298
54	10.315	Quercetin hexuronide II	$C_{21}H_{18}O_{13}$	477.0680	1.1	13	477.0687; **301.0356**; 283.0236; 255.0300; 227.0338; 151.0036
55	10.315	Quercetin 3-*O*-hexoside III	$C_{12}H_{20}O_{12}$	463.0890	1.7	12	463.0908; 301.0357; **300.0276**; 271.0246; 255.0296; 151.0033
56	10.379	Benzyl-dihydroxybenzoate-*O*-pentosil-hexoside I	$C_{25}H_{30}O_{13}$	537.1624	1.9	11	429.1045; 297.0591; 243.0656; **153.0198;** 135.0086
57	10.515	Quercetin 3-*O*-pentoside	$C_{20}H_{18}O_{11}$	433.0786	1.5	12	433.0773; 301.0346; **300.0270**; 271.0240; 255.0289
58	10.592	Isochestanin	$C_{40}H_{42}O_{26}$	937.1904	1.3	20	937.1939; 469.1001; **467.0853**; 303.0303; 169.0143
59	10.635	Benzyl-dihydroxybenzoate-*O*-hexoside	$C_{20}H_{22}O_{9}$	405.1192	0.2	10	405.1164; 297.0611; 243.0643; 153.0195; **135.0082**; 109.0284
60	10.810	Kaempferol deoxyhexosyl-hexoside I	$C_{27}H_{30}O_{15}$	593.1533	3.5	13	593.1545; **285.0405**; 284.0330; 255.0301
61	10.887	Isorhamnetin deoxyhexoside I	$C_{21}H_{20}O_{11}$	447.0941	1.8	12	447.0949; 315.0140; 301.0358; 300.0280; 285.0408; 284.0333; 271.0246; **255.0302**; 227.0350

62	11.099	Isorhamnetin deoxyhexosyl-hexoside I	$C_{28}H_{32}O_{16}$	623.1632	2.2	13	623.1650; **315.0513**: 314.0435; 300.0273; 299.0195; 271.0244
63	11.176	Kaempferol deoxyhexosyl-hexoside II	$C_{27}H_{30}O_{15}$	593.1526	2.4	13	593.1557; **285.0409**; 284.0328; 255.0299
64	11.196	Isorhamnetin 3-*O*-hexoside I	$C_{22}H_{22}O_{12}$	477.1046	1.6	12	477.1059; 315.0503; **314.0435**; 300.0270; 299.0187; 271.0248
65	11.293	Isorhamnetin deoxyhexoside II	$C_{21}H_{20}O_{11}$	447.0935	0.5	12	447.0932; 315.0142; 314.0065; 300.0266; 299.9910; 285.0394; 284.0319; 271.0240; **255.0296**; 227.0344
66	11.293	Isorhamnetin hexuronide	$C_{22}H_{20}O_{13}$	491.0841	2.0	13	491.0817; 315.0507; **300.0268**; 271,0237; 255.0290
67	11.510	Isorhamnetin deoxyhexosyl-hexoside II	$C_{28}H_{32}O_{16}$	623.1633	2.3	13	623.1647; **315.0508**: 314.0428; 300.0273; 299.0195; 271.0244
68	11.570	Isorhamnetin 3-*O*-hexoside II	$C_{22}H_{22}O_{12}$	477.1051	2.6	12	477.1049; 315.0495; **314.0422**; 300.0273; 299.0194; 285.0405; 271.0248
69	11.980	Trimethylellagic acid hexose	$C_{23}H_{24}O_{13}$	551.1057 $[M+FA]^-$	n.c.	n.c.	343.0457; **328.0216**; 312.9983; 297.9743
70	12.181	Kaempferol acetylhexoside I	$C_{23}H_{22}O_{12}$	489.1058	4.0	13	489.1065; 285.0399; **284.0325**; 255.0295; 227.0346
71	12.402	Quercetin	$C_{15}H_{10}O_7$	301.0354	-0.3	11	301.0348; 273.0396; 227.0344; **151.0031;** 107.0152
72	12.737	Luteolin	$C_{15}H_{10}O_6$	285.0398	-2.3	11	285.0394; 241.0495; 175.0400; **133.0297**
73	12.819	Kaempferol acetylhexoside II	$C_{23}H_{22}O_{12}$	489.1051	2.6	13	489.1072; 285.0400; **284.0331**; 255.0300; 227.0346
74	13.160	Benzyl-dihydroxybenzoate-*O*-pentosil-hexoside II	$C_{25}H_{30}O_{13}$	537.1635	4.0	11	537.1937; **243.0661**; 152.0110; 108.0214
75	13.729	Quercetin *p*-coumaroyl-hexoside	$C_{30}H_{26}O_{14}$	609.1258	1.3	18	609.1258; 463.0855; **301.0337**; 300.0256; 271.0243; 151.0034
76	13.749	Quercetin *p*-coumaroyl-deoxyhexosyl-hexoside I	$C_{36}H_{36}O_{18}$	755.1854	3.3	19	755.1878; 609.1496: 591.1388; **301.0350**; 300.0270; 271.0248
77	13.769	Kaempferol *p*-coumaroyl-hexoside I	$C_{30}H_{26}O_{13}$	593.1326	2.4	13	593.1316; 447.0942; 307.0810; **285.0396**; 284.0319; 255.0289
78	13.968	Quercetin hexuronide butyl-ester	$C_{25}H_{26}O_{14}$	533.1316	2.9	13	533.1324; 357.0983; 301.0350; **300.0278;** 271.0243; 255.0291
79	13.968	Quercetin *p*-coumaroyl- deoxyhexosil-hexoside II	$C_{36}H_{36}O_{18}$	755.1850	3.3	19	755.1866; 609.1486: 591.1374; **301.0353**; 300.0276; 285.0399; 271.0248; 255.0291
80	13.984	Kaempferol	$C_{15}H_{10}O_6$	285.0403	-0.6	11	**285.0401**; 255.0296; 227.0348; 107.0150
81	14.174	Trimethylellagic acid deoxyhexose	$C_{23}H_{22}O_{12}$	535.1106 $[M+FA]^-$	n.c.	n.c.	343.04600; **328.0226**; 312.9990; 269.9802
82	14.174	Trimethylellagic acid I	$C_{17}H_{12}O_8$	343.0455	-1.3	12	328.0242; 312.9971; **297.9752;** 269.9794
83	14.366	Kaempferol deoxyhexosyl-*p*-coumaroyl-hexoside I	$C_{36}H_{36}O_{17}$	739.1894	1.9	19	739.1915; 593.1530; 575.1424; 453.1399; **285.0401**; 284.0318
84	14.436	Traumatic acid	$C_{12}H_{20}O_4$	227.1291	1.0	3	**183.1388**
85	14.561	Kaempferol *p*-coumaroyl-hexoside II	$C_{30}H_{26}O_{13}$	593.1311	1.7	13	447.0943; **285.0404**; 284.0330; 255.0296
86	14.643	Kaempferol deoxyhexosyl-*p*-coumaroyl-hexoside II	$C_{36}H_{36}O_{17}$	739.1899	2.9	19	739.1955; 593.1567; 575.1453; 453.1426; **285.0417**; 284.0335
87	15.139	Trimethylellagic acid II	$C_{17}H_{12}O_8$	343.0459	-0.1	12	328.0222; 312.9985; **297.9753;** 269.9796
88	15.563	Kaempferol acetyl *p*-coumaroyl-hexoside	$C_{32}H_{28}O_{14}$	635.1432	4.0	19	635.1441; 489.1068; **285.0404**; 284.0324
89	15.799	Methylkaempferol	$C_{16}H_{12}O_6$	299.0559	-0.7	11	299.0550; **284.0320**; 256.0368; 227.0336; 133.0298
90	16.019	3,7,24-trihydroxy-cycloartene-28-oic acid hexoside	$C_{36}H_{58}O_{10}$	695.4037 $[M+FA]^-$	n.c.	8	695.4039; 649.3972; **487.3450**
91	17.088	Kaempferol di-*p*-coumaroyl hexoside I	$C_{39}H_{32}O_{15}$	739.1695	3.6	24	739.1724; 593.1351; 453.1211; 307.0823; **285.0409;** 284.0326
92	17.284	Kaempferol di-*p*-coumaroyl hexoside II	$C_{39}H_{32}O_{15}$	739.1694	3.3	24	739.1715; 593.1337; 453.1211; **285.0409**; 284.0323
93	17.313	Acacetin	$C_{16}H_{12}O_5$	283.0611	-0.3	11	283.0594; **268.0363**; 211.0385; 151.0027; 117.0342
94	17.343	3,7-dihydroxynorcycloartane 24,28-dioic acid (e.g., castaartancrenoic acid A)	$C_{27}H_{42}O_6$	461.2921	2.7	7	**461.2908**; 443.2804; 417.2991; 415.2841
95	17.856	Cycloartane-type triterpene	$C_{30}H_{50}O_6$	505.3544	1.9	6	**505.3553;** 487.3429; 469.3327; 459.2758; 443.3201
96	18.337	Kaempferol acetyl di-*p*-coumaroyl hexoside I	$C_{41}H_{34}O_{16}$	781.1792	2.3	25	781.1803; 635.1434; 495.1325**; 285.0401;** 284.0320; 145.0287
97	18.594	Kaempferol acetyl di-*p*-coumaroyl hexoside II	$C_{41}H_{34}O_{16}$	781.1797	2.9	25	781.1797; 635.1417; 495.1289**; 285.0394;** 284.0312; 145.0293
98	18.752	Cycloartane-type triterpene	$C_{27}H_{42}O_5$	445.2956	-0.8	7	**445.2959**; 427.2842; 399.2893; 383.2949
99	19.845	Cycloartane-type triterpene	$C_{30}H_{48}O_5$	487.3435	1.2	7	**487.3439;** 469.3326

100	19.954	Kaempferol di-acetyl di-*p*-coumaroyl hexoside I	$C_{43}H_{36}O_{17}$	823.1879	-0.1	26	823.1903; 677.1529; 659.1410; 617.1293; 537.1411**; 285.0396**; 284.0313; 145.0294
101	20.044	Cycloartane-type triterpene	$C_{30}H_{48}O_5$	487.3437	1.6	7	**487.3435**; 469.3325; 441.3378; 383.2937
102	20.144	Kaempferol di-acetyl di-*p*-coumaroyl hexoside II	$C_{43}H_{36}O_{17}$	823.1885	0.6	26	823.1916; 677.1551; 617.1326; 557.1125; **285.0406;** 284.0325
103	20.256	Cycloartane-type triterpene	$C_{30}H_{48}O_5$	487.3438	1.8	7	**487.3444;** 469.3328
104	20.425	Pentacyclic triterpene	$C_{30}H_{48}O_6$	503.3383	1.0	7	503.3394**; 471.3130;** 469.3319; 453.3015
105	20.606	DGMG (18:3)	$C_{33}H_{56}O_{14}$	675.3601	0.5	6	415.1457; 397.1344; **277.2172**; 235.0804
106	20.721	9-oxooctadeca-10,12-dienoic acid	$C_{18}H_{32}O_3$	293.2123	0.3	4	293.2123; **275.2015**; 231.2077; 183.1384
107	20.721	Pentacyclic triterpene	$C_{30}H_{48}O_6$	503.3385	1.4	7	503.3399; 485.3282; 471.3130; 445.2957; 401.2708; 387.2543; 359.2230; **319.1914**
108	21.100	9-oxooctadeca-10,12,15 trienoic acid	$C_{18}H_{28}O_3$	291.1965	-0.2	5	**291.1948;** 273.1847; 247.2077; 211.1332; 195.1379
109	21.318	l-PA (18:3)	$C_{21}H_{37}O_7P$	431.2212	1.8	4	277.2166; **152.9959**
110	21.846	Pentacyclic triterpene	$C_{30}H_{48}O_4$	471.3501 517.3558 [M+FA]-	4.5	7	**471.3513**
111	22.313	l-PA (16:0)	$C_{19}H_{39}O_7P$	409.2358	-0.6	1	409.2335; 255.2325; **152.9957**
112	22.513	Pentacyclic triterpene	$C_{30}H_{48}O_4$	471.3487	1.5	7	**471.3478**
113	22.654	Pentacyclic triterpene	$C_{30}H_{48}O_5$	487.3440	2.9	7	487.3430; 469.3330; 455.3172; 385.2745; **303.1957**
114	22.695	Pentacyclic triterpene	$C_{30}H_{48}O_4$	471.3480	0.1	7	**471.3480**
115	22.784	Pentacyclic triterpene	$C_{30}H_{48}O_5$	487.3433	0.8	7	487.3433; **455.3179**
116	22.975	Linolenic acid	$C_{18}H_{30}O_2$	277.2172	-0.4	4	**277.2137**
117	23.115	Pentacyclic triterpene	$C_{30}H_{48}O_4$	471.3483	0.6	7	**471.3482;** 453.3371; 425.3433; 407.3324
118	23.215	Pentacyclic triterpene (e.g. ursolic or oleanolic acid)	$C_{30}H_{48}O_3$	455.3537	1.4	7	**455.3552**
119	23.496	Linoleic acid	$C_{18}H_{32}O_2$	279.2324	-2.0	3	**279.2314**

Table S2. Effects of *C. sativa* L. extracts at different doses (50 and 200 mg) on fermentation end products after 24 h of incubation. Total VFA: total volatile fatty acids (acetate + propionate + butyrate + iso-butyrate + valerate + iso-valerate); AcA = acetic acid; PrA = propionic acid; ButA = Butyric acid; ValA = valeric acid; iso-ButA = iso-butyric acid; iso-ValA = iso-valeric acid; BCFA= branched chain fatty acids (iso-butyrate + iso-valerate/tVFA); A/P=Acetate/Propionate. Along the row * $p < 0.05$, ** $p < 0.01$ and *** $p < 0.001$; NS: not significant; MSE: mean square error.

	Control diet	**Cs/1/1**		**Cs/2/1**		**Cs/3/2**		**MSE**
		50mg	**200mg**	**50mg**	**200mg**	**50mg**	**200mg**	
pH	6.32	6.34^{NS}	6.24^{NS}	6.35^{NS}	6.34^{NS}	6.35^{NS}	6.71^{NS}	0.02
Total VFA (mmol/L)	41.6	55.0***	54.3***	47.4***	58.5***	49.5**	38.7***	0.74
AcA (% VFA)	62.4	56.2***	55.9***	59.7*	56.6***	64.7^{NS}	61.4^{NS}	0.58
PrA (% VFA)	17.6	21.3***	22.6***	17.7^{NS}	19.5*	17.1^{NS}	18.2^{NS}	0.23
ButA (% VFA)	13.9	19.6***	18.8***	19.7***	20.9***	15.6*	18.3***	0.16

ValA (% VFA)	3.70	1.97***	2.11***	1.79***	1.87***	1.52***	1.44***	0.02
iso-ButA (% VFA)	0.61	0.50^{NS}	0.24**	0.51^{NS}	0.50^{NS}	0.50^{NS}	0.28**	0.005
iso-ValA (% VFA)	1.67	0.44***	0.38***	0.51***	0.54***	0.45***	0.36***	0.001
BCFA (%VFA)	2.29	0.97***	0.62***	1.02***	1.04***	0.95***	0.64***	0.004
A/P (%VFA)	3.53	2.64***	2.47***	3.37^{NS}	2.90**	3.77^{NS}	3.37^{NS}	0.01

Table S3. Values of Pearson's coefficient correlation, between antiradical ($DPPH^{\bullet}$, $ABTS^{\bullet+}$) activities, reducing activity (PFRAP), total phenol content (TPC), total flavonoid content (TFC) with fermentation parameters at the dose level of 50 mg. tVFA: total volatile fatty acids; AcA = acetic acid; PrA = propionic acid; ButA = Butyric acid; ValA = valeric acid; iso-ButA = iso-butyric acid; iso-ValA = iso-valeric acid; BCFA: branched chain fatty acids (iso-butyrate + iso-valerate/tVFA); A/P=Acetate/Propionate; OMD: organic matter degradability; OMCV: cumulative volume of gas related to incubated organic matter. R_{max}: maximum fermentation rate; T_{max}: time at which R_{max} occurs.

	TFC	*TPC*	*ABTS*	*DPPH*	*FRAP*	*pH*	*tVFA*	*AcA*	*PrA*	*ButA*	*ValA*	*iso-ButA*	*iso-ValA*	*BCFA*	*A/P*	*OMD*	*OMCV*	R_{max}	T_{max}
TFC	1,000																		
TPC	0,935	1,000																	
ABTS	0,985	0,982	1,000																
DPPH	0,968	0,994	0,997	1,000															
FRAP	0,927	1,000	0,978	0,991	1,000														
pH	-0,949	-0,999	-0,989	-0,998	-0,998	1,000													
tVFA	0,998	0,913	0,973	0,952	0,904	-0,929	1,000												
AcA	0,207	-0,153	0,036	-0,044	-0,174	0,111	0,263	1,000											
PrA	0,607	0,849	0,735	0,786	0,860	-0,826	0,560	-0,652	1,000										
ButA	-0,873	-0,989	-0,944	-0,967	-0,992	0,982	-0,843	0,297	-0,918	1,000									
ValA	-0,602	-0,280	-0,456	-0,384	-0,260	0,321	-0,648	-0,906	0,269	0,136	1,000								
Iso-ButA	0,608	0,287	0,462	0,390	0,266	-0,327	0,653	0,903	-0,262	-0,143	-1,000	1,000							
Iso-ValA	-0,677	-0,894	-0,794	-0,840	-0,903	0,874	-0,634	0,579	-0,996	0,950	-0,179	0,172	1,000						
BCFA	-0,383	-0,686	-0,536	-0,602	-0,701	0,654	-0,329	0,825	-0,967	0,785	-0,507	0,501	0,939	1,000					
A/P	-0,473	-0,755	-0,618	-0,678	-0,768	0,726	-0,421	0,764	-0,987	0,843	-0,418	0,412	0,969	0,995	1,000				
OMD	0,888	0,993	0,954	0,975	0,996	-0,987	0,860	-0,267	0,905	-1,000	-0,167	0,174	-0,940	-0,765	-0,826	1,000			
OMCV	0,979	0,844	0,930	0,898	0,832	-0,866	0,989	0,401	0,434	-0,756	-0,751	0,756	-0,514	-0,188	-0,285	0,776	1,000		
R_{max}	-0,914	-0,998	-0,970	-0,986	-0,999	0,995	-0,889	0,207	-0,877	0,996	0,227	-0,234	0,917	0,724	0,790	-0,998	-0,813	1,000	
T_{max}	0,578	0,251	0,429	0,356	0,230	-0,292	0,624	0,918	-0,298	-0,106	-1,000	0,999	0,209	0,533	0,446	0,137	0,731	-0,197	1,000

Table S4. Values of Pearson's coefficient correlation, between antiradical ($DPPH^{\bullet}$, $ABTS^{\bullet+}$) activities, reducing activity (PFRAP), total phenol content (TPC), total flavonoid content (TFC) with fermentation parameters at the dose level of 200 mg. tVFA: total volatile fatty acids; AcA = acetic acid; PrA = propionic acid; ButA = Butyric acid; ValA = valeric acid; iso-ButA = iso-butyric acid; iso-ValA = iso-valeric acid; BCFA: branched chain fatty acids (iso-butyrate + iso-valerate/tVFA); A/P=Acetate/Propionate; OMD: organic matter degradability; OMCV: cumulative volume of gas related to incubated organic matter. R_{max}: maximum fermentation rate; T_{max}: time at which R_{max} occurs.

	TFC	*TPC*	*ABTS*	*DPPH*	*FRAP*	*pH*	*tVFA*	*AcA*	*PrA*	*ButA*	*ValA*	*iso-ButA*	*iso-ValA*	*BCFA*	*A/P*	*OMD*	*OMCV*	R_{max}	T_{max}

TFC	1,00																		
TPC	0,935	1,000																	
ABTS	0,985	0,982	1,000																
DPPH	0,968	0,994	0,997	1,000															
FRAP	0,927	1,000	0,978	0,991	1,000														
pH	-0,340	-0,651	-0,497	-0,564	-0,667	1,000													
tVFA	-0,951	-0,779	-0,884	-0,844	-0,766	0,033	1,000												
AcA	0,765	0,944	0,864	0,902	0,950	-0,866	-0,528	1,000											
PrA	-0,512	-0,783	-0,652	-0,710	-0,796	0,982	0,221	-0,945	1,000										
ButA	-0,488	-0,147	-0,331	-0,255	-0,126	-0,655	0,734	0,189	-0,500	1,000									
ValA	-0,895	-0,995	-0,959	-0,978	-0,997	0,723	0,714	-0,972	0,841	0,049	1,000								
iso-ButA	-0,949	-0,776	-0,881	-0,840	-0,762	0,027	1,000	-0,523	0,215	0,738	0,710	1,000							
iso-ValA	-0,537	-0,801	-0,674	-0,731	-0,814	0,976	0,249	-0,954	1,000	-0,474	0,856	0,244	1,000						
BCFA	-0,272	-0,595	-0,433	-0,503	-0,612	0,997	-0,039	-0,828	0,966	-0,707	0,672	-0,045	0,958	1,000					
A/P	0,652	0,878	0,773	0,821	0,888	-0,935	-0,385	0,987	-0,985	0,344	-0,921	-0,380	-0,990	-0,907	1,000				
OMD	0,521	0,185	0,367	0,292	0,164	0,625	-0,760	-0,151	0,466	-0,999	-0,087	-0,763	0,440	0,680	-0,307	1,000			
OMCV	0,603	0,281	0,457	0,385	0,261	0,545	-0,820	-0,052	0,376	-0,991	-0,185	-0,824	0,349	0,604	-0,211	0,995	1,000		
R_{max}	-0,550	-0,810	-0,686	-0,741	-0,823	0,972	0,265	-0,959	0,999	-0,460	0,864	0,259	1,000	0,953	-0,992	0,426	0,334	1,000	
T_{max}	0,966	0,995	0,996	1,000	0,993	-0,573	-0,838	0,906	-0,717	-0,245	-0,980	-0,835	-0,738	-0,512	0,827	0,282	0,375	-0,748	1,000