

Electronic Supplementary Information

Bioavailability and metabolism of hydroxycinnamates in rats fed durum wheat aleurone fractions.

Luca Calani^{1,2#}, Fayçal Ounnas^{3#}, Patricia Salen³, Christile Demeilliers⁴, Letizia Bresciani^{1,2},
Francesca Scazzina², Furio Brighenti², Camilla Melegari⁵, Alan Crozier⁶, Michel de
Lorgeril³, Daniele Del Rio^{1,2*}.

¹LS9 Interlab Group, The Laboratory of Phytochemicals in Physiology, Department of Food Science, University of Parma, Parma, Italy

²Human Nutrition Unit, Department of Food Science, University of Parma, Parma, Italy.

³Laboratoire TIMC-IMAG CNRS UMR 5525, Cœur et Nutrition, Faculté de Médecine, Université Joseph Fourier, Grenoble, France.

⁴Laboratory of Fundamental and Applied Bioenergetics (LBFA), Université Grenoble Alpes, Grenoble, France

⁵Barilla G. e R. F.lli, Via Mantova, 166, 43122 Parma, Italy

⁶Plant Products and Human Nutrition Group, School of Medicine, College of Biomedical and Life Sciences, University of Glasgow, Glasgow, UK

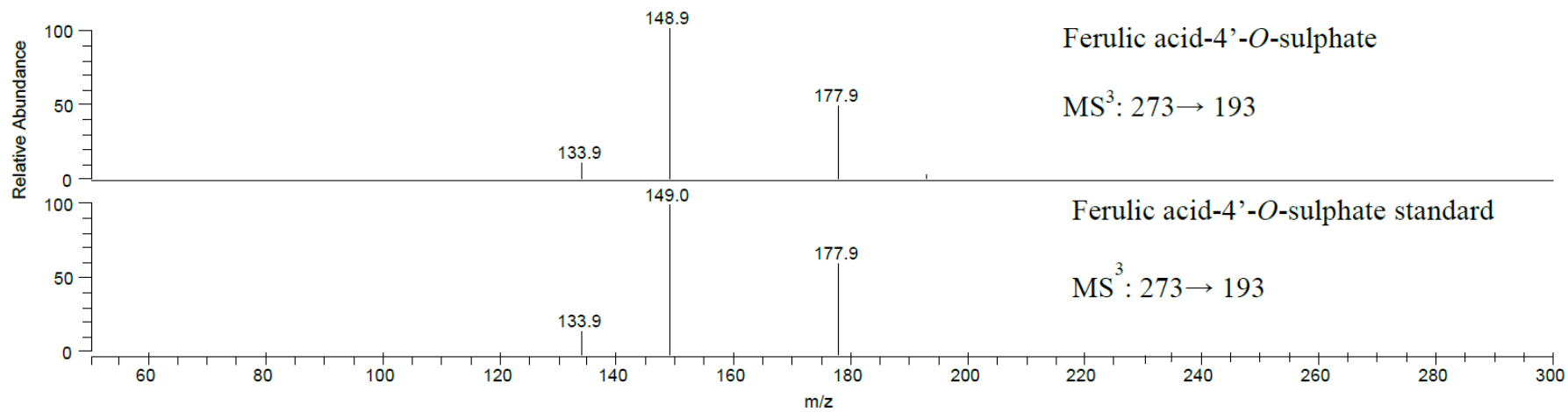
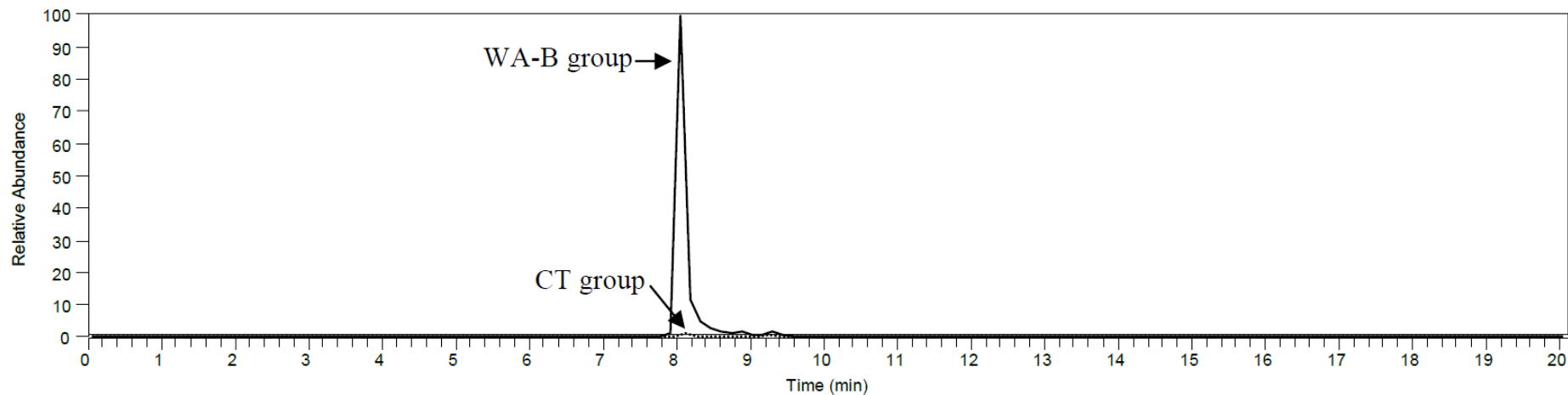
Supplemental Table 1. UHPLC-MSⁿ identification of phenolic compounds in the aleurone fractions ingested by rats.

Rt (min)	Compound	[M-H] ⁻ (m/z)	MS ² ions (m/z)	MS ³ ions (m/z)
4.73	4-Hydroxybenzoic acid	137	93	
8.52	4'-Coumaric acid	163	119	
6.52	Caffeic acid	179	135	
9.37	<i>trans</i> -Ferulic acid	193	149 (100), 178 (63), 134 (21)	
9.59	Ferulic acid isomer	193	134 (100), 149 (63), 178 (30)	
9.66	Diferulic acid	385	341 (100), 317 (3), 249 (1)	[341]: 326 (100), 297 (71), 282 (25), 217 (9)
9.70	<i>trans</i> -Sinapic acid	223	208 (100), 179 (61), 164 (13)	
10.04	Sinapic acid isomer	223	179 (100), 208 (30), 164 (13)	
10.72	Triferulic acid	577	533 (100), 509 (49), 441 (33), 531 (29), 489 (28), 513 (10)	[533]: 409 (100), 445 (76), 367 (15)
10.94	Diferulic acid	385	341 (100), 297 (33), 317 (3), 249 (2), 323 (1)	[341]: 297 (100), 326 (40), 282 (10), 323 (3)
12.06	Triferulic acid	577	489 (100), 441 (39), 509 (31), 531 (19), 533 (12)	[533]: 417 (100), 193 (46), 409 (3)
12.66	Diferulic acid	385	341 (100), 317 (44), 249 (21), 339 (10), 367 (4), 349 (3)	[341]: 309 (100), 297 (4), 296 (1)
12.99	Diferulic acid	385	341 (100), 317 (7), 297 (6), 249 (2), 339 (2), 367 (1)	[341]: 326 (100), 297 (93), 282 (23), 323 (3)
13.00	Triferulic acid	577	533 (100), 341 (35), 489 (33), 461 (271), 441 (19), 509 (18), 531 (9)	[533]: 341 (100), 297 (4), 445 (2) [341]: 297 (100), 326 (83), 282 (20), 217 (14)
13.55	Diferulic acid	385	341 (100), 326 (12), 370 (10), 282 (4), 297 (3), 342 (2), 367 (2), 249 (1)	[341]: 326 (100), 282 (34), 297 (16), 249 (3), 323 (2)
13.94	diferulic acid	385	313 (100), 193 (26), 341 (8), 317 (2), 249 (1)	[341]: 297 (100), 326 (77), 309 (57), 282 (17)

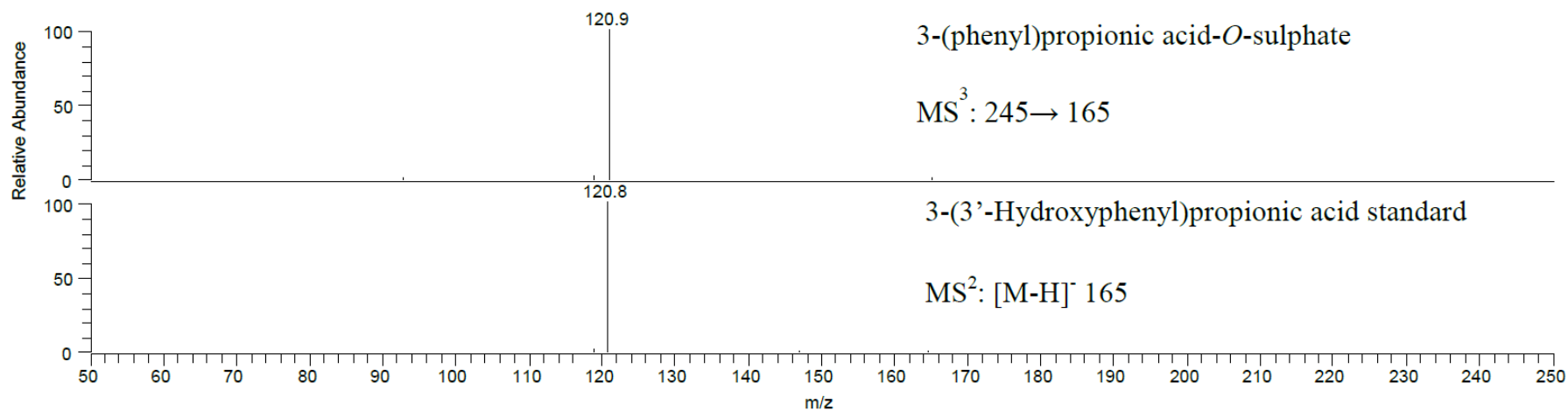
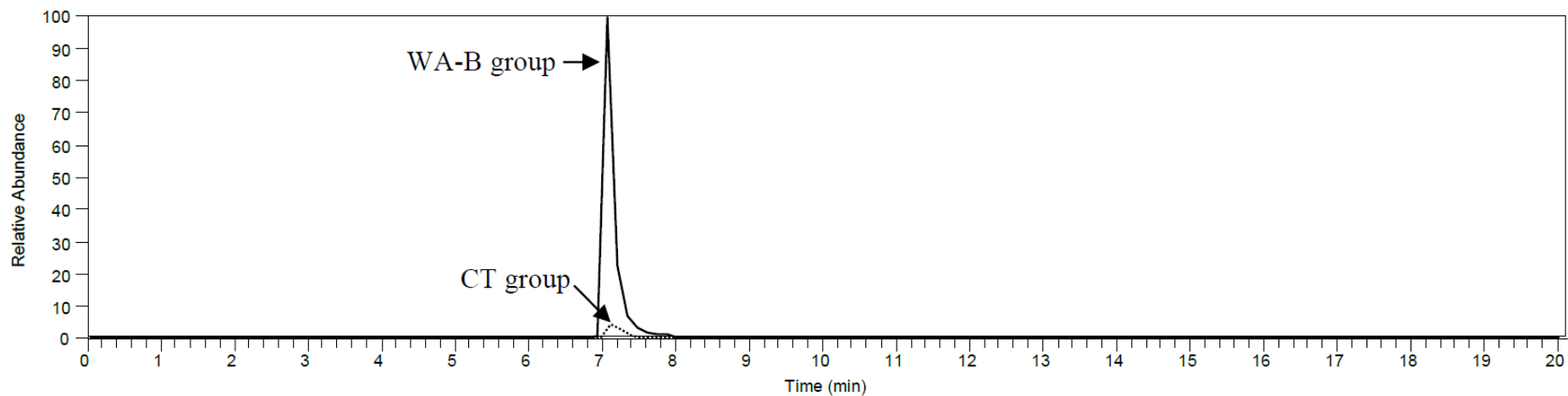
14.16	diferulic acid	385	341 (100), 317 (1)	[193]: 149 (100), 178 (57), 134 (25) [341]: 297 (100), 326 (70), 282 (17), 323 (4)
14.38	diferulic acid	385	341 (100), 313 (23), 193 (13), 317 (11), 249 (6), 316 (2)	[341]: 326 (100), 297 (31), 282 (28), 323 (3) [193]: 149 (100), 178 (68), 134 (22)
14.94	triferulic acid	577	385 (100), 441 (14), 531 (9), 341 (5), 533 (4)	[385]: 341 (100), 370 (14), 326 (12), 282 (5), 297 (2), 367 (1) [341]: 326 (100), 282 (26), 297 (5),
15.05	triferulic acid	577	533 (100), 355 (17), 509 (7), 441 (5), 311 (5), 461 (3)	[533]: 311 (100), 296 (7), 193 (1), 341 (1)

The relative ionic abundance for each ion is reported in round brackets. The ions in square brackets were fragmented in MS³ experiments.

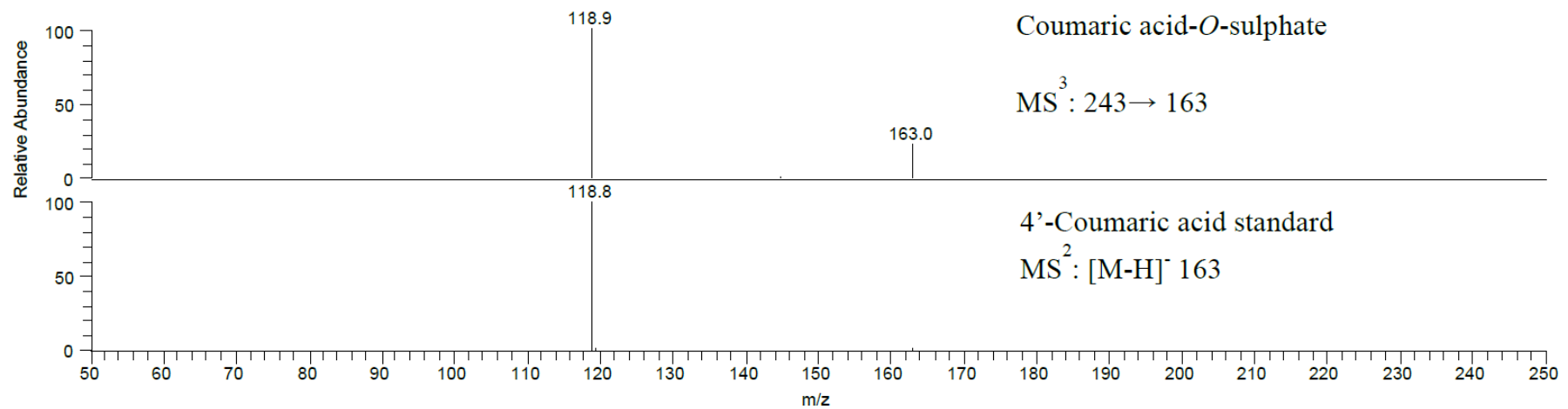
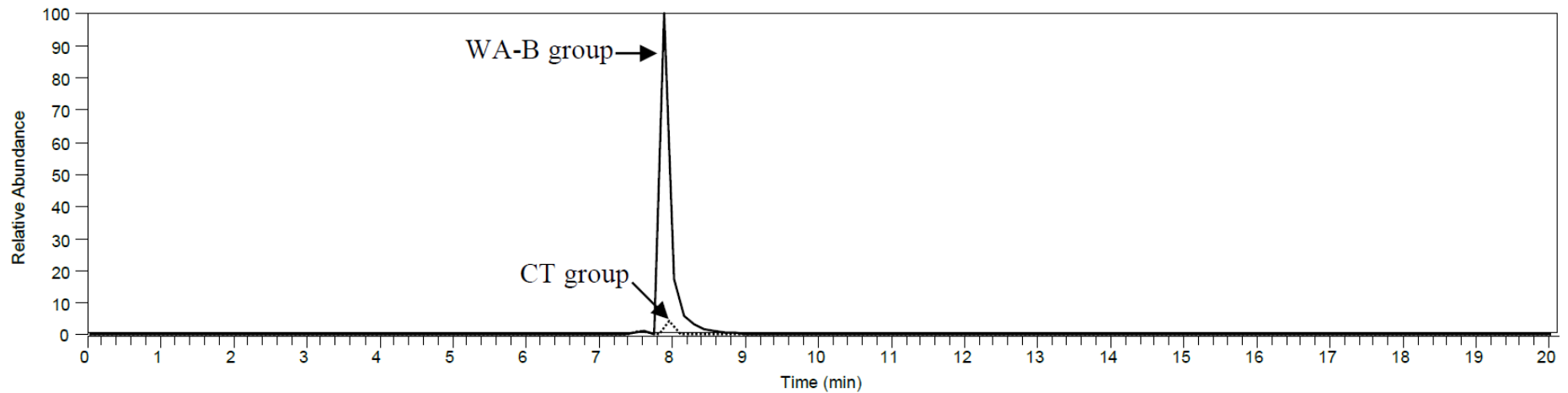
Supplemental Figure 1. Comparison of chromatographic traces of ferulic acid-4'-*O*-sulphate in WA-B group and CT group and comparison of MS³ ion spectrum of the same urinary compound with those of corresponding standard.



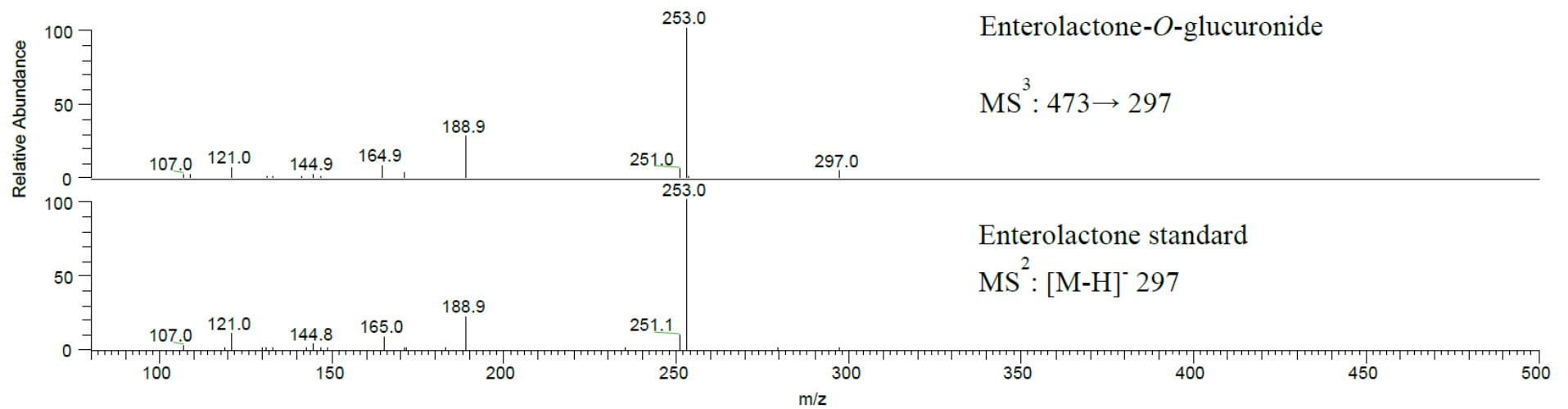
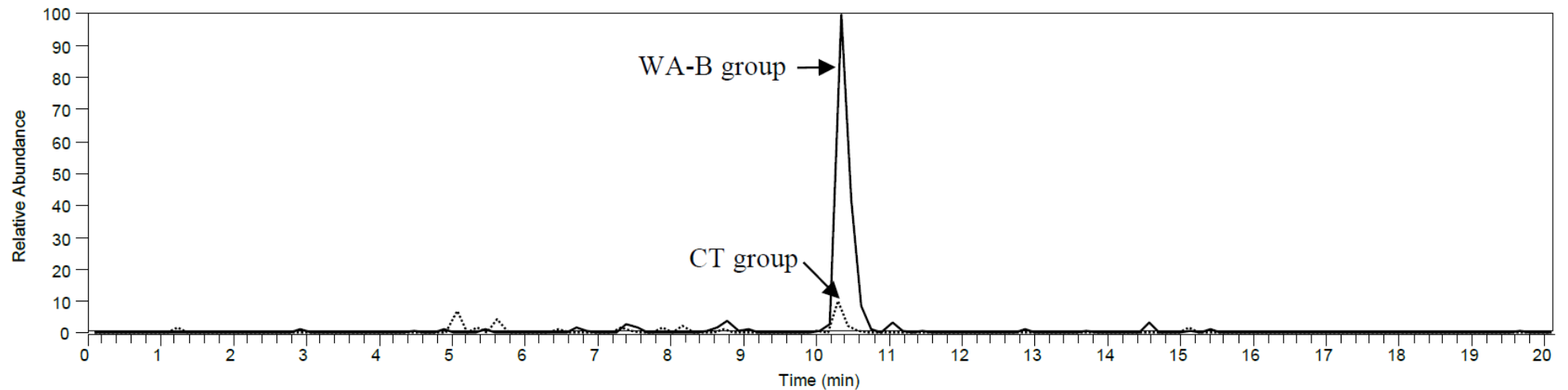
Supplemental Figure 2. Comparison of chromatographic traces of 3-(phenyl)propionic acid-*O*-sulphate in WA-B group and CT group and comparison of MS³ ion spectrum of the same urinary compound with MS² ion spectrum of 3'-hydroxyphenylpropionic acid standard.



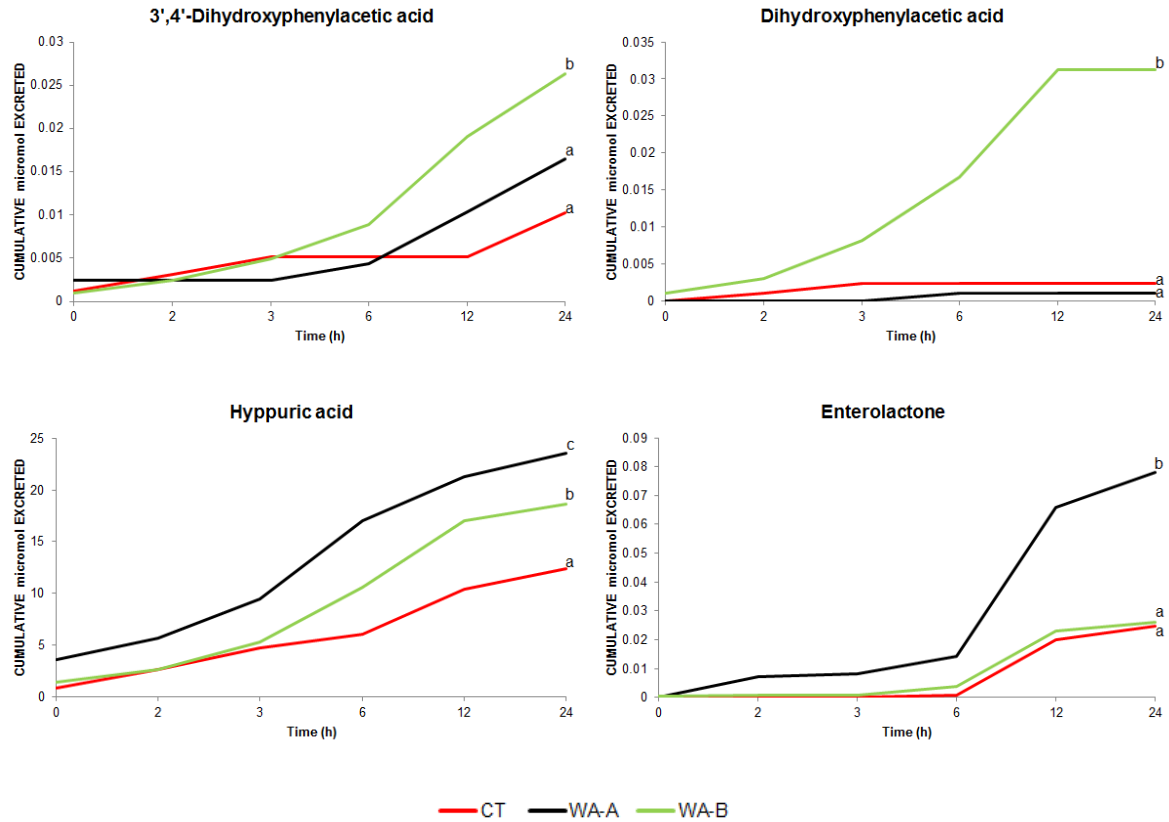
Supplemental Figure 3. Comparison of chromatographic traces of coumaric acid-*O*-sulphate in WA-B group and CT group and comparison of MS³ ion spectrum of the same urinary compound with MS² ion spectrum of 4'-coumaric acid standard.



Supplemental Figure 4. Comparison of chromatographic traces of enterolactone-*O*-glucuronide in WA-B group and CT group and comparison of MS³ ion spectrum of the same urinary compound with MS² ion spectrum of enterolactone standard.



Supplemental Figure 5. Cumulative urinary excretion of some phenolic metabolites in the acute feeding. CT: control group. WA-A: wheat aleurone A group. WA-B: wheat aleurone B group. Data are expressed as mean values of μmol excreted, with $n=4$ for each group. Letters are included for values significantly different among groups ($p<0.05$).



Supplemental figure 6. Excretion in the 24 h-urine of some phenolic metabolites in the chronic feeding. CT: control group. WA-A: wheat aleurone A group. WA-B: wheat aleurone B group. Data are expressed as mean values of $\mu\text{mol} \pm \text{SEM}$, with $n=6$ for each group. Letters are included for values significantly different among groups ($p<0.05$).

