Comprehensive analysis of fatty acid metabolites produced by gut microbiota using LC-MS/MS-based lipidomics

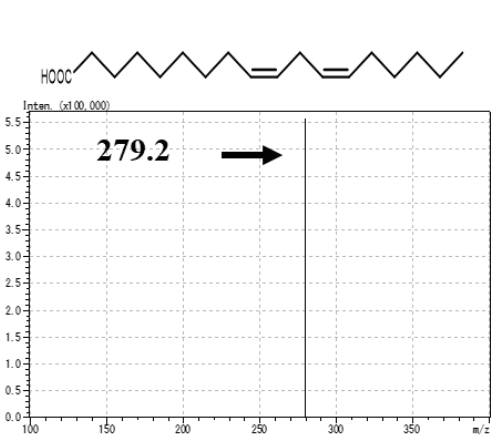
Electronic Supplementary Material

Kowa Tsuji, Wataru Shimada, Shigenobu Kishino, Jun Ogawa, Makoto Arita

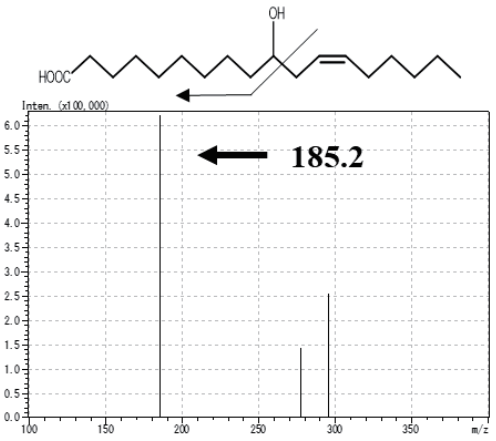
Content

Fig. S1

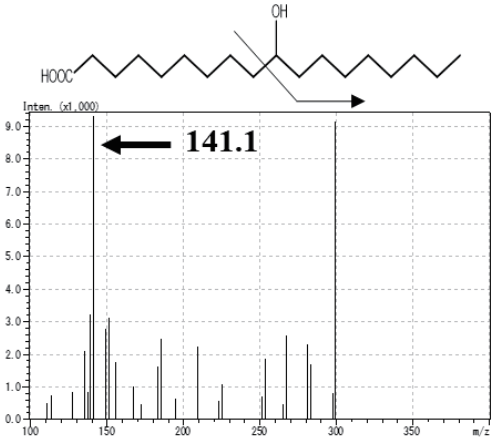
LA (Q1-Q3 : 279.2-279.2)



HYA (Q1-Q3 : 297.2-185.2)



HYB (Q1-Q3 : 299.2-141.1)



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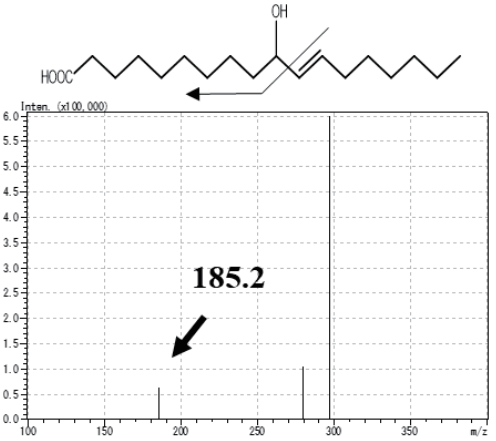

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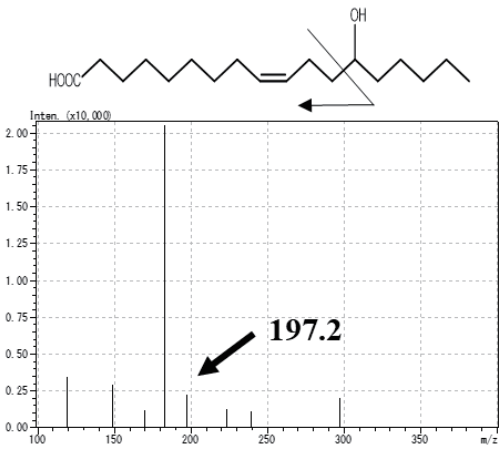

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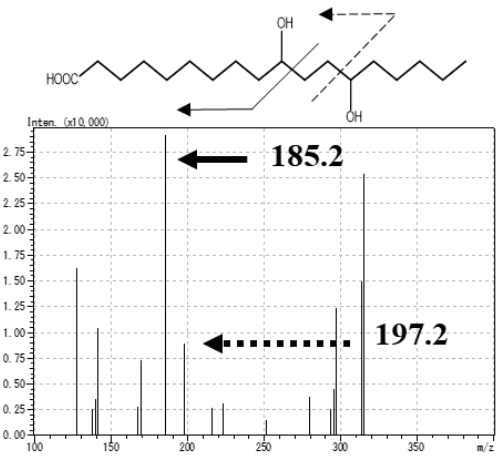

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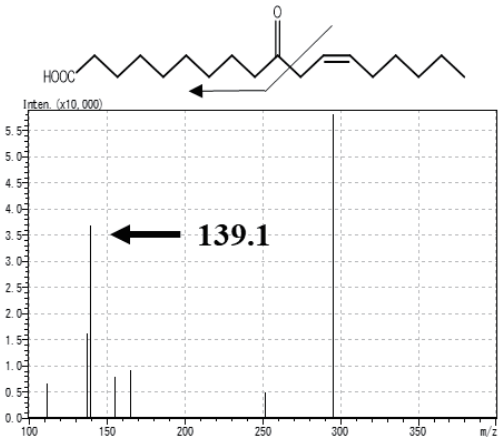

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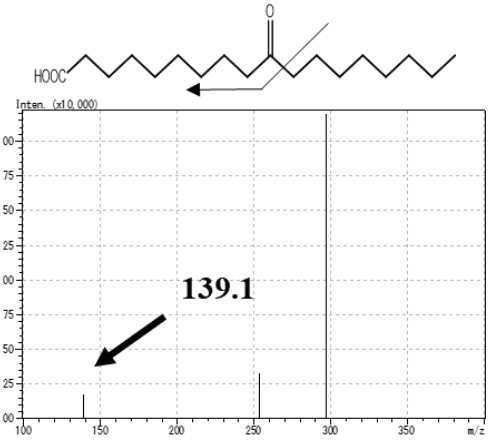

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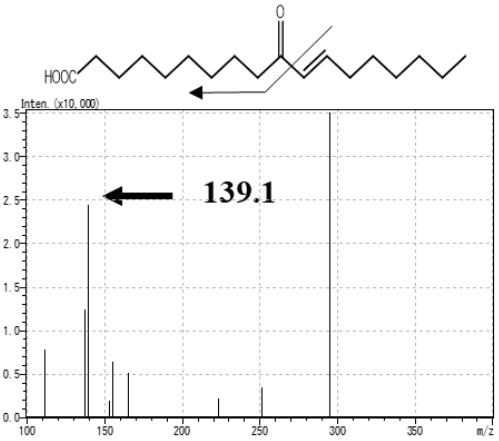

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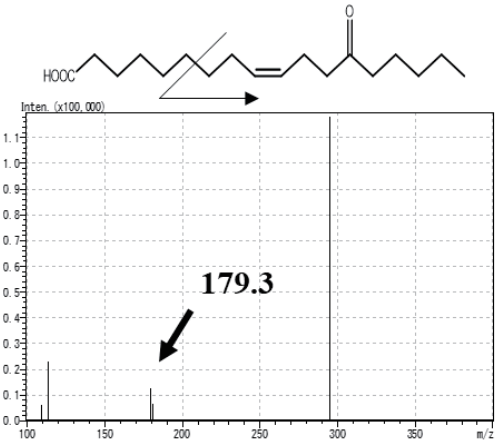

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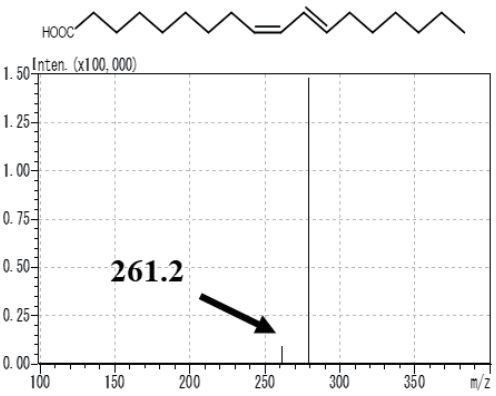

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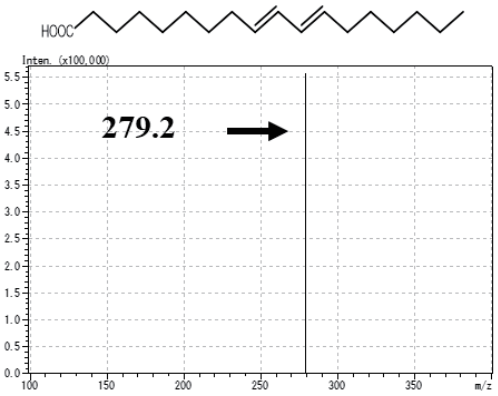

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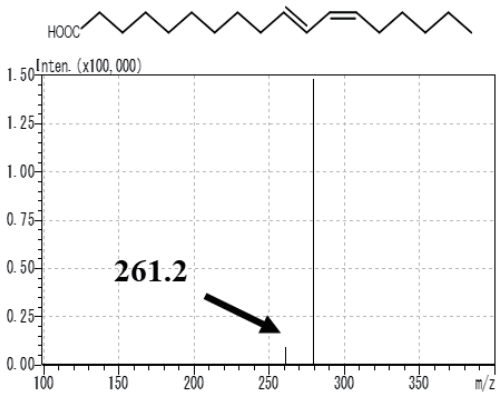

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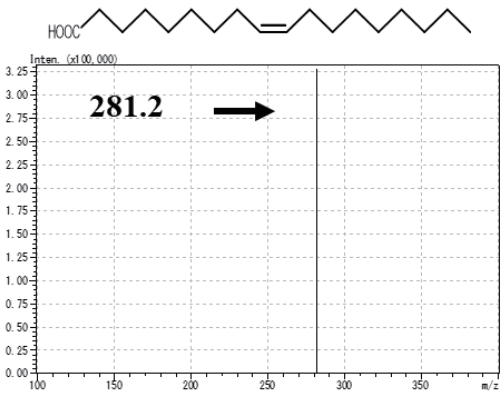

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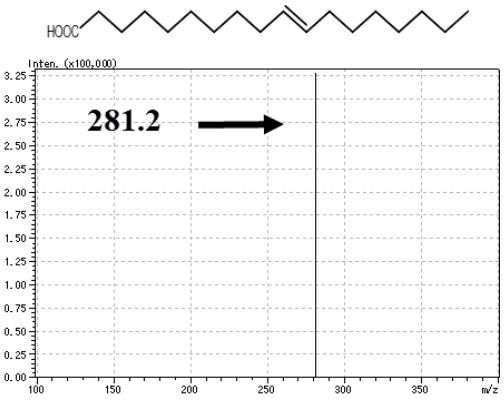

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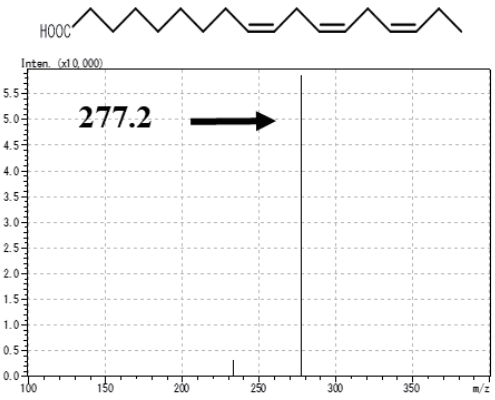

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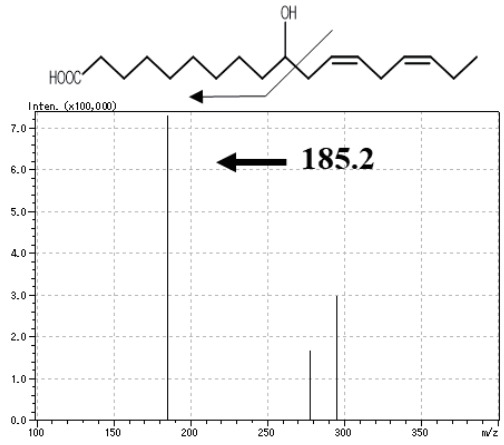

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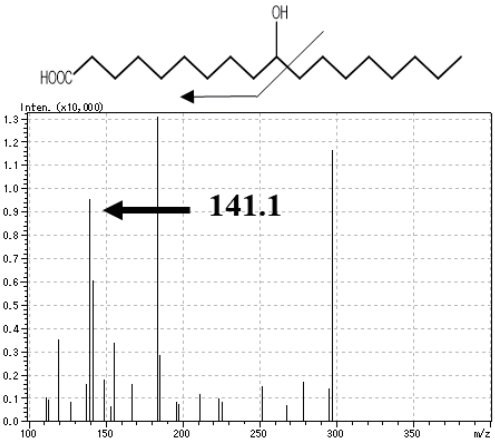

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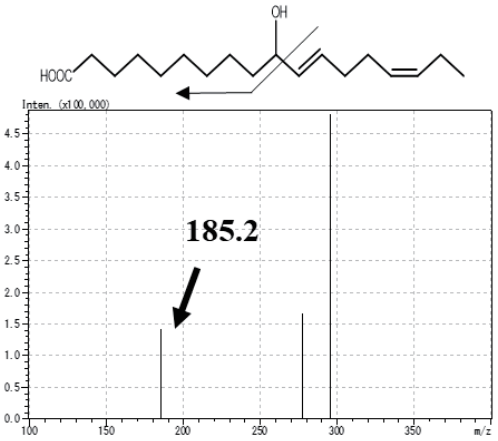

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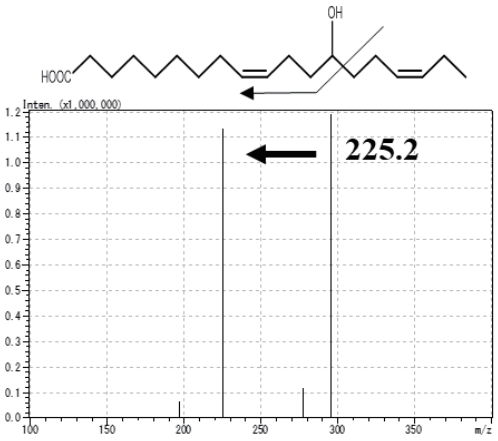

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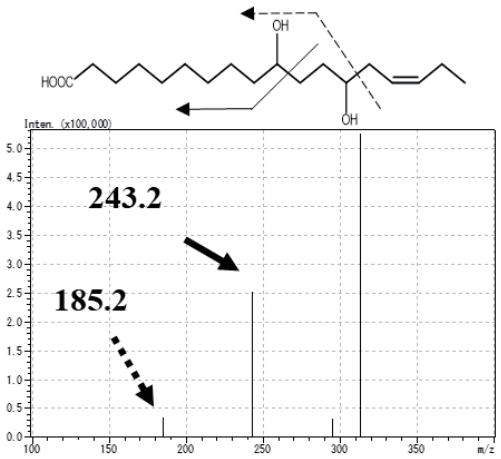

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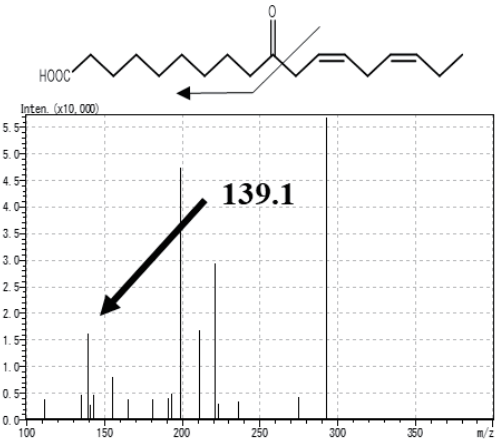

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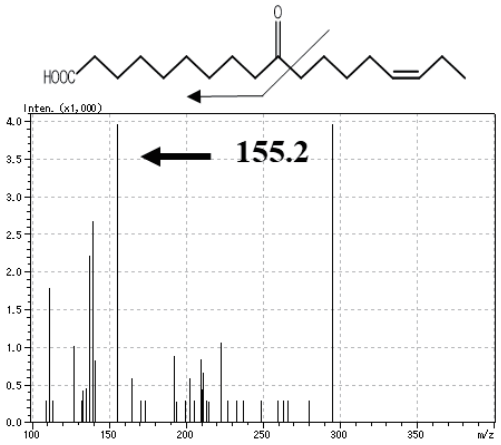

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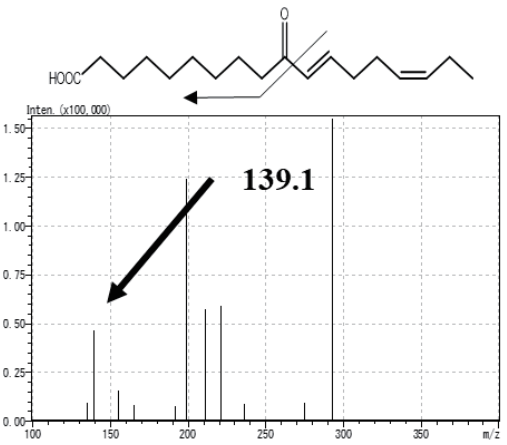

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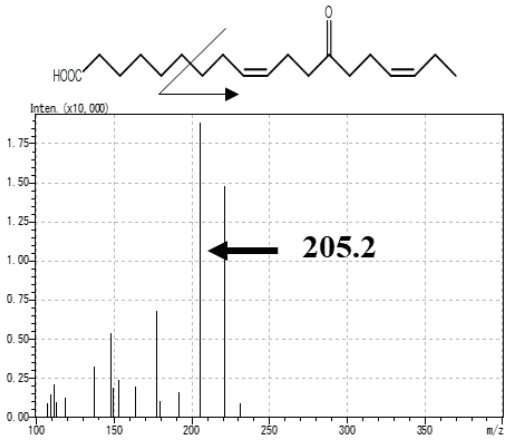

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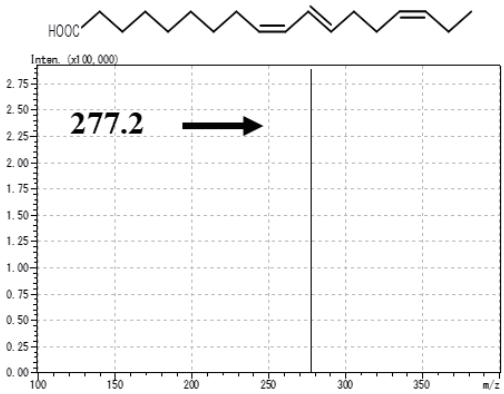

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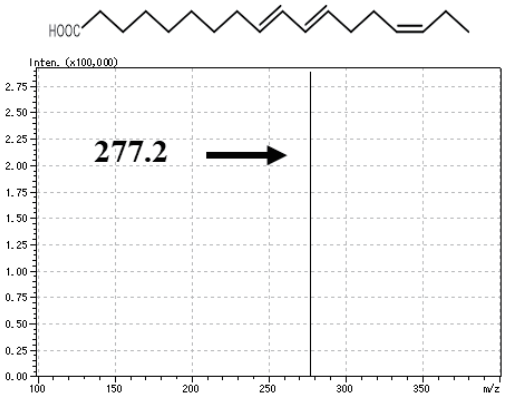

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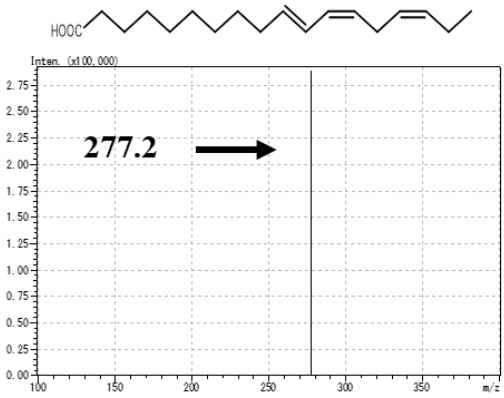

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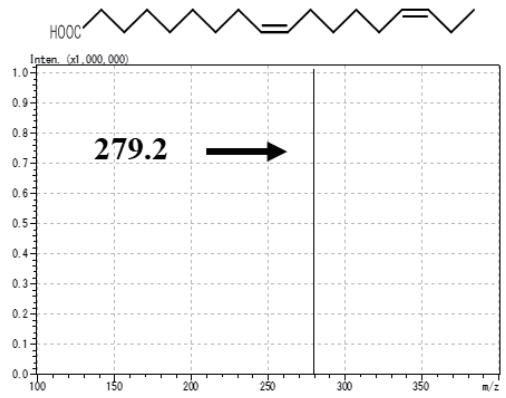

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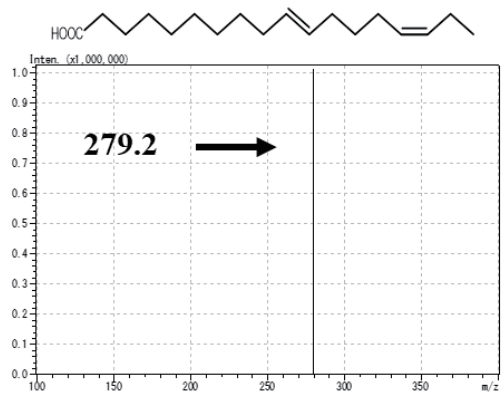

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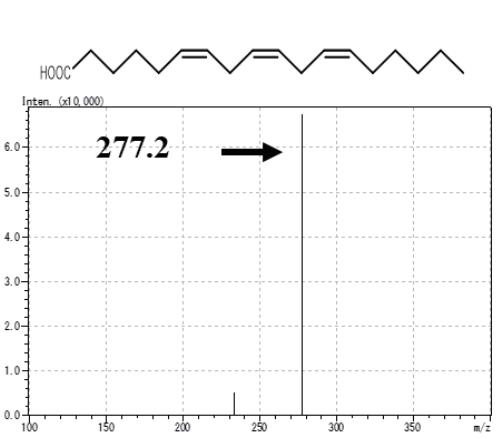
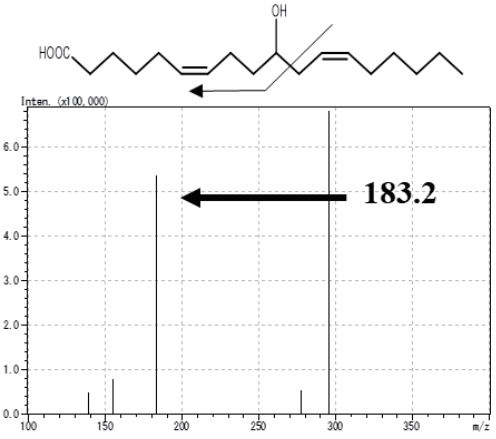
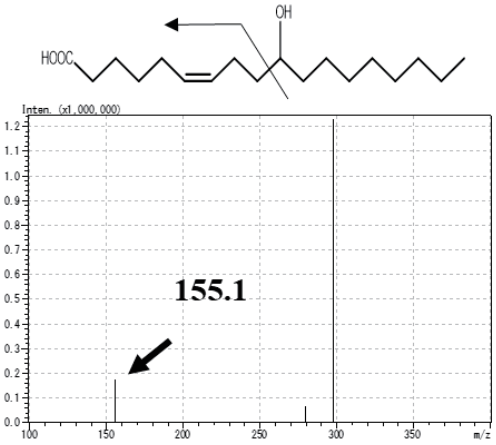


γHYA (Q1-Q3 : 295.2-183.2)



γHYB (Q1-Q3 : 297.2-155.1)



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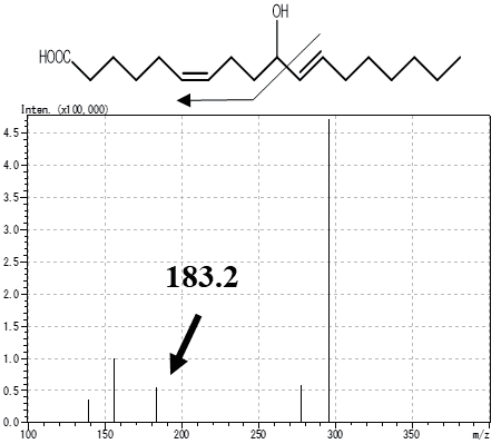

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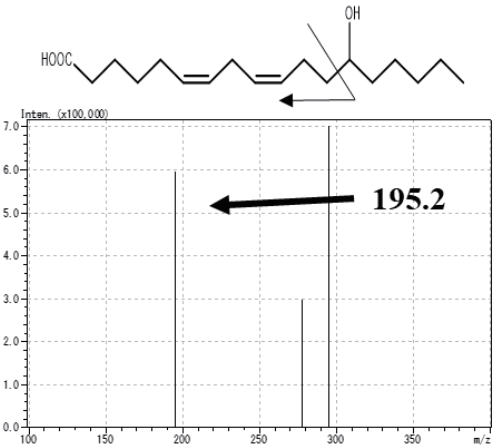

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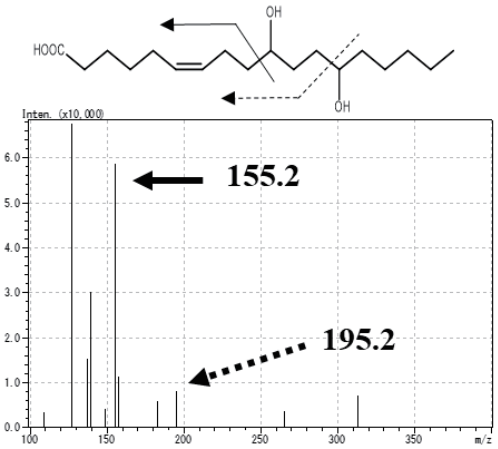

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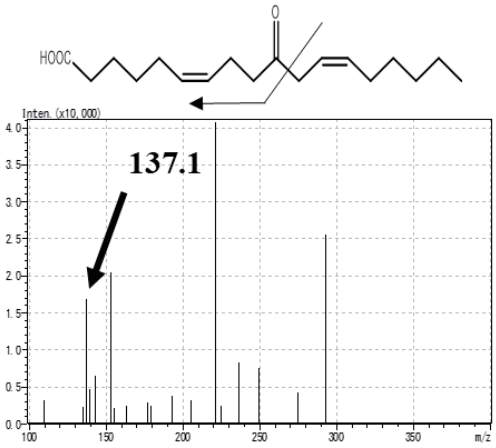

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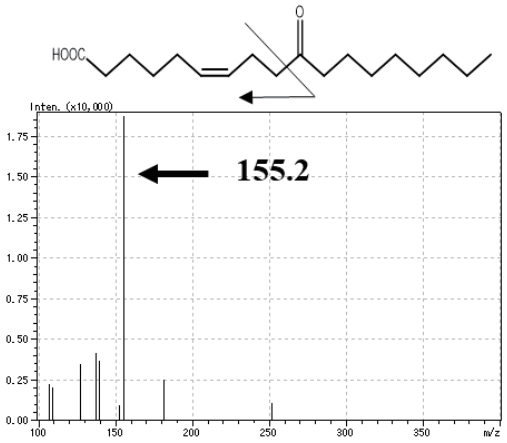

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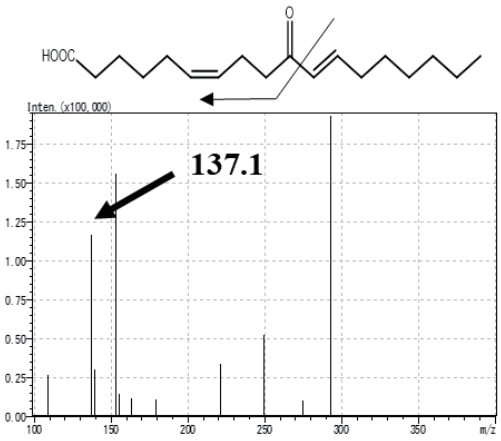

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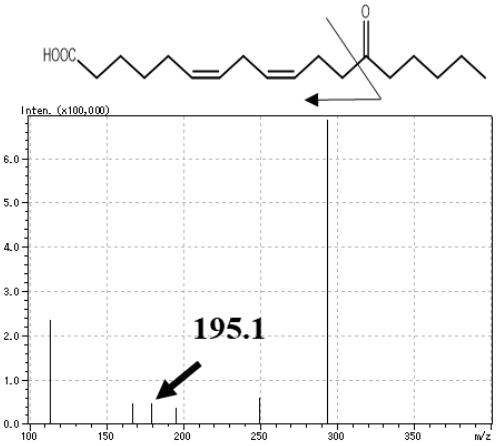
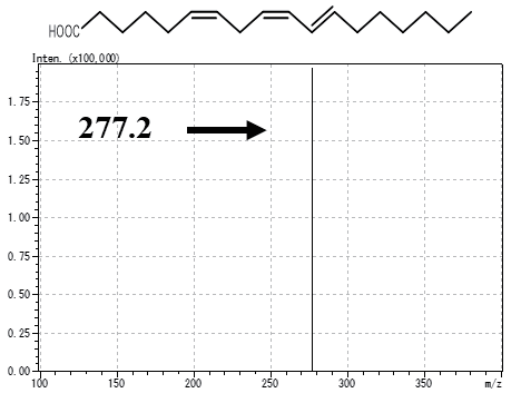


CGLA1 (Q1-Q3 : 277.2-277.2)



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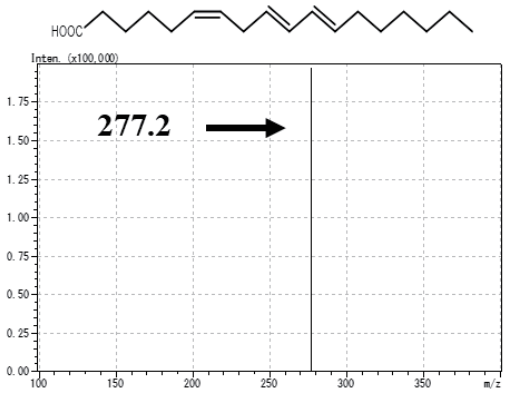

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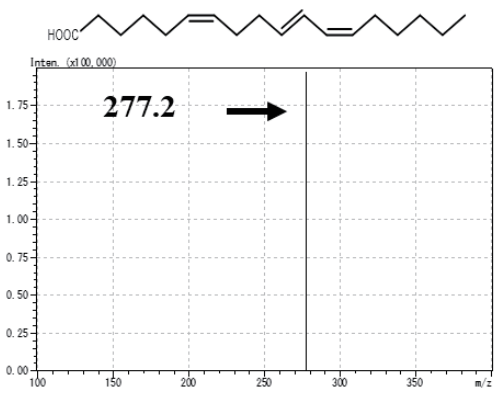

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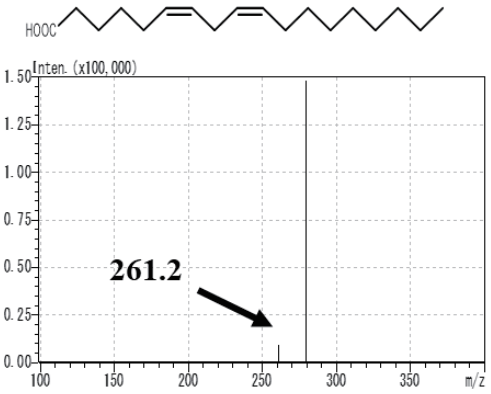

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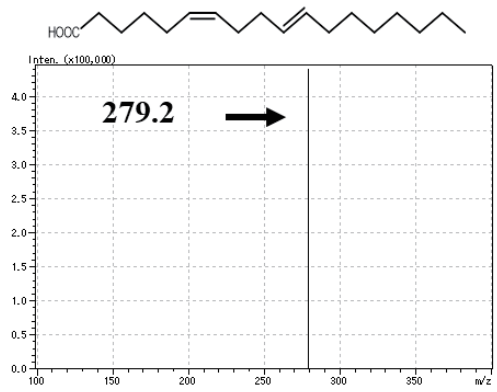
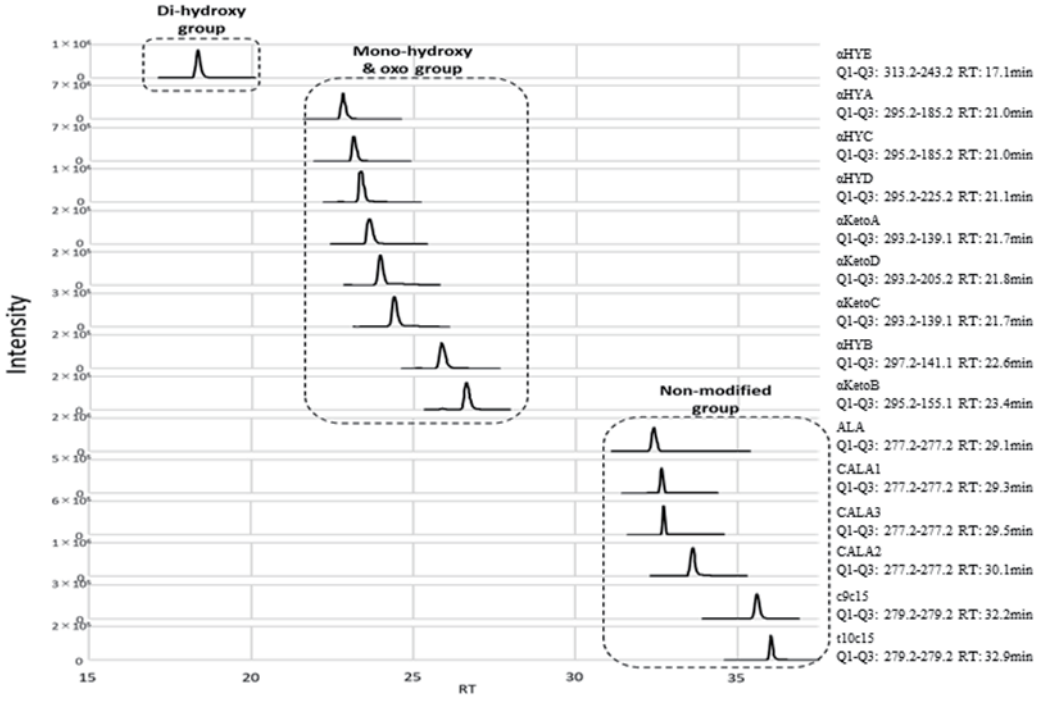


Fig. S2

A


Di-hydroxy group
Mono-hydroxy & oxo group
Non-modified group
Intensity
RT
αHYE Q1-Q3: 313.2-243.2 RT: 17.1min
αHYA Q1-Q3: 295.2-185.2 RT: 21.0min
αHYC Q1-Q3: 295.2-185.2 RT: 21.0min
αHYD Q1-Q3: 295.2-225.2 RT: 21.1min
αKetoA Q1-Q3: 293.2-139.1 RT: 21.7min
αKetoD Q1-Q3: 293.2-205.2 RT: 21.8min
αKetoC Q1-Q3: 293.2-139.1 RT: 21.7min
αHYB Q1-Q3: 297.2-141.1 RT: 22.6min
αKetoB Q1-Q3: 295.2-155.1 RT: 23.4min
ALA Q1-Q3: 277.2-277.2 RT: 29.1min
CALA1 Q1-Q3: 277.2-277.2 RT: 29.3min
CALA3 Q1-Q3: 277.2-277.2 RT: 29.5min
CALA2 Q1-Q3: 277.2-277.2 RT: 30.1min
c9c15 Q1-Q3: 279.2-279.2 RT: 32.2min
t10c15 Q1-Q3: 279.2-279.2 RT: 32.9min

B

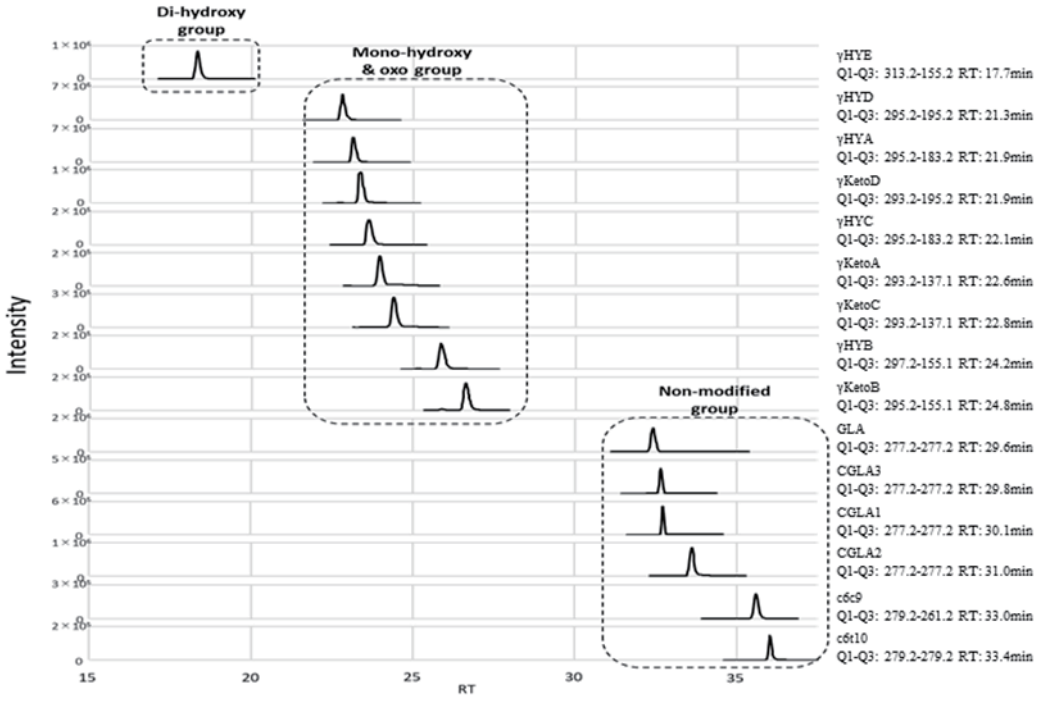

Di-hydroxy group
Mono-hydroxy & oxo group
Non-modified group
Intensity
RT
γHYE Q1-Q3: 313.2-155.2 RT: 17.7min
γHYD Q1-Q3: 295.2-195.2 RT: 21.3min
γHYA Q1-Q3: 295.2-183.2 RT: 21.9min
γKetoD Q1-Q3: 293.2-195.2 RT: 21.9min
γHYC Q1-Q3: 295.2-183.2 RT: 22.1min
γKetoA Q1-Q3: 293.2-137.1 RT: 22.6min
γKetoC Q1-Q3: 293.2-137.1 RT: 22.8min
γHYB Q1-Q3: 297.2-155.1 RT: 24.2min
γKetoB Q1-Q3: 295.2-155.1 RT: 24.8min
GLA Q1-Q3: 277.2-277.2 RT: 29.6min
CGLA3 Q1-Q3: 277.2-277.2 RT: 29.8min
CGLA1 Q1-Q3: 277.2-277.2 RT: 30.1min
CGLA2 Q1-Q3: 277.2-277.2 RT: 31.0min
c6c9 Q1-Q3: 279.2-261.2 RT: 33.0min
c6t10 Q1-Q3: 279.2-279.2 RT: 33.4min

Fig. S3


LA
HYA
HYB
HYC
HYD
HYE
KetoA
KetoB
KetoC
KetoD
CLA1
CLA2
CLA3
OA
t10
ALA
αHYA
αHYB
αHYC
αHYD
αHYE
αKetoA
αKetoB
αKetoC
αKetoD
CALA1
CALA2
CALA3
c9c15
t10c15
GLA
γHYA
γHYB
γHYC
γHYD
γHYE
γKetoA
γKetoB
γKetoC
γKetoD
CGLA1
CGLA2
CGLA3
c6c9
c6t10
pg/ μL
Area value

Table S1 Concentration of C18 fatty acid metabolites (ng/100 mg or ng/100 μL) in SI, and plasma of Saff-, Lin-, and Eve-mice

Group	Compound	SI			Plasma		
		Safflower	Linseed	Evening primrose	Safflower	Linseed	Evening primrose
LA	LA [c9c15]	7321.46±641.08	6547.75±523.09	9935.55±858.56	331.18±161.24	168.3±97.24	369.43±18.79
	HYA	1.88±0.18	1.36±0.13	4.45±1.36	0.04±0.01	0.05±0.01	0.06±0.02
	HYB	11.41±2.58	18.19±4.73	12.58±4.67	0.33±0.05	0.35±0.12	0.12±0.03
	HYC	2.58±0.98	1.08±0.36	N.D.	N.D.	N.D.	N.D.
	HYD	20.9±7.2	9.34±2.86	15.96±3.71	1.37±0.2	0.41±0.15	1.01±0.33
	HYE	3.24±1.11	0.49±0.1	0.5±0.17	N.D.	N.D.	N.D.
	KetoA	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	KetoB	19.37±6.51	16.96±3.57	4.66±0.44	0.26±0.04	0.23±0.05	0.11±0.04
	KetoC	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	KetoD	42.41±4.65	23.16±4.05	55.54±17.58	1.55±0.26	0.35±0.11	1.38±0.55
	CLA1 [CLA3]	157.25±46.19	154.06±65.2	265.34±93.98	2.05±1.95	5.23±4.19	7.37±2.94
	CLA2	N.D.	N.D.	N.D.	0.31±0.35	0.18±0.09	0.45±0.32
	OA	13731.9±1760.55	17587.99±1985.77	15613.31±923.42	488.99±307.75	491.55±250.22	607.77±209.74
	t10	239.21±71.24	446.43±99.49	302.95±34.51	N.D.	N.D.	N.D.
ALA	ALA	849.05±523.88	7995.95±1092.51	566.62±78.11	19.74±6.67	459.62±160.5	11.88±2.97
	αHYA [αHYC]	0.06±0.07	0.69±0.18	0.18±0.08	N.D.	0.02±0.01	0.01±0
	αHYB	12.9±3.45	8.86±2.89	1.3±0.22	N.D.	0.06±0.01	N.D.
	αHYD	1.15±1.19	11.89±2.63	0.58±0.23	0.02±0.01	0.22±0.06	0.03±0.03
	αHYE	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	αKetoA [αKetoC]	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	αKetoB	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	αKetoD	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	CALA1	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	CALA2 [CGLA1]	93.1±25.58	167.25±19.13	376.17±64.8	12.11±5.1	50.65±27.62	18.58±17.09
	CALA3 [GLA]	471.27±96.19	533.71±88.68	4178.28±1093.04	48.44±16.1	17.33±9.28	195.55±25.91
	c9c15 [LA]	7321.46±641.08	6547.75±523.09	9935.55±858.56	331.18±161.24	168.3±97.24	369.43±18.79
	t10c15	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
GLA	GLA [CALA3]	471.27±96.19	533.71±88.68	4178.28±1093.04	48.44±16.1	17.33±9.28	195.55±25.91
	γHYA [γHYC]	0.18±0.03	0.16±0.32	1.71±0.49	0.01±0.01	N.D.	0.06±0.01
	γHYB	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.

γHYC	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
γHYD	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
γHYE	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
γKetoA	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
γKetoB	N.D.	N.D.	0.18±0.37	N.D.	N.D.	N.D.
γKetoC	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
γKetoD	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
CGLA1 [CALA2]	93.1±25.58	167.25±19.13	376.17±64.8	12.11±5.1	50.65±27.62	18.58±17.09
CGLA2	11.07±1.75	10.14±4.32	18.3±7.56	0.87±0.33	N.D.	N.D.
CGLA3	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
c6c9	N.D.	N.D.	N.D.	4.62±1.34	3.7±2.55	10.43±4.42
c6t10	N.D.	N.D.	N.D.	0.83±0.28	0.6±0.44	1.53±0.39

Data are expressed as the mean ± SE, n = 5. N.D.: not detected because concentrations were lower than the limit of detection. [ ]: not separated metabolite.

Table.S2 Concentration of C18 fatty acid metabolites (ng/100 mg) in liver and kidney of Saff-, Lin-, and Eve-mice

Group	Compound	Liver			Kidney		
		Safflower	Linseed	Evening primrose	Safflower	Linseed	Evening primrose
LA	LA [c9c15]	1877.26±261.04	1059.87±596.04	1566.69±119.38	1059.01±188.93	1002.98±163.4	1218.13±215.03
	HYA	0.14±0.01	0.19±0.04	0.12±0.01	0.2±0.1	0.1±0.05	0.15±0.03
	HYB	0.83±0.15	0.64±0.12	0.44±0.02	0.66±0.16	0.31±0.07	0.31±0.07
	HYC	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	HYD	1.6±0.18	0.58±0.16	1.48±0.18	0.97±0.22	0.22±0.07	0.64±0.28
	HYE	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	KetoA	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	KetoB	0.69±0.11	0.69±0.1	0.55±0.12	1.27±0.43	0.69±0.29	0.64±0.18
	KetoC	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	KetoD	0.61±0.16	0.41±0.1	0.48±0.16	0.71±0.35	0.27±0.1	0.45±0.18
	CLA1 [CLA3]	13.54±8.57	9.49±10.22	18.82±14.64	N.D.	N.D.	N.D.
	CLA2	2.6±1.86	0.92±0.7	0.79±0.45	0.77±0.5	0.25±0.15	0.73±0.35
	OA	4175.37±792.13	3098.26±1558.89	3193.99±651.32	1463.29±346.87	2222.65±411.98	1957.65±472.28
	t10	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
ALA	ALA	30.83±5.67	1699.92±308.64	25.79±13.9	16.27±4.15	972.48±239.15	15.17±4.22
	αHYA [αHYC]	N.D.	0.03±0.01	N.D.	N.D.	0.03±0.01	N.D.
	αHYB	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	αHYD	0.03±0.01	0.24±0.12	0.02±0	0.02±0.01	0.17±0.12	0.01±0
	αHYE	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	αKetoA [αKetoC]	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	αKetoB	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	αKetoD	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	CALA1	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
	CALA2 [CGLA1]	57.22±9.96	151.29±115.16	88.82±28.8	13.19±5.58	N.D.	5.61±11.22
	CALA3 [GLA]	258.16±32.73	98.82±47.03	557.7±185.22	105.91±16.99	48.16±10.24	284.27±58.27
	c9c15 [LA]	1877.26±261.04	1059.87±596.04	1566.69±119.38	1059.01±188.93	1002.98±163.4	1218.13±215.03
	t10c15	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
GLA	GLA [CALA3]	258.16±32.73	98.82±47.03	557.7±185.22	105.91±16.99	48.16±10.24	284.27±58.27
	γHYA [γHYC]	0.03±0.02	N.D.	0.08±0.01	N.D.	N.D.	0.03±0

γHYB	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
γHYC	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
γHYD	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
γHYE	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
γKetoA	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
γKetoB	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
γKetoC	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
γKetoD	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
CGLA1 [CALA2]	57.22±9.96	151.29±115.16	88.82±28.8	13.19±5.58	N.D.	5.61±11.22
CGLA2	3.94±0.53	N.D.	5.97±3.26	N.D.	N.D.	N.D.
CGLA3	20.79±8.16	43.72±32.47	30.18±20.2	N.D.	N.D.	N.D.
c6c9	44.94±8.98	16.56±11.02	27.1±21.96	12.83±3.3	5.59±1.85	10.76±2.91
c6t10	10.2±2.96	3.5±2.29	11.92±3.15	6.14±2.3	3.16±1.66	7.22±2.19

Data are expressed as the mean ± SE, n = 5. N.D.: not detected because concentrations were lower than the limit of detection. [ ]: not separated metabolite.