

Supplementary Materials

Discovery of potential biomarkers in human melanoma cells with different metastatic potential by metabolic and lipidomic profiling

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Supplementary Table S1. Global metabolic profiles of human primary melanocytes (HEMn-LP) and melanoma cell lines (A375, A2058) using GC-MS. ANOVA was performed to detect statistically significant differences between samples ($p < 0.05$). Different letters indicate statistically significant differences between metabolite levels. RT, retention time; TMS, trimethylsilylation; ND, not detected.

No.	Compound	RT (min)	Fragmentation ion (m/z)	TMS	HEMn-LP	A375	A2058
Alcohol							
1	Myo-inositol	28.93	191, 217, 305 , 318	6	1.29 ± 0.42^a	964.61 ± 75.51^b	35.38 ± 7.50^a
2	Glucitol	26.27	103, 205, 217, 319	6	ND	0.62 ± 0.14^b	1.02 ± 0.42^c
Amino acid							
3	Alanine	7.01	100, 116 , 190, 218	2	63.39 ± 31.37^a	14.09 ± 1.14^b	34.51 ± 8.45^b
4	β -Alanine	15.13	133, 174 , 248, 290	3	ND	12.03 ± 1.26^b	13.97 ± 2.90^b
5	Aspartic acid	17.43	100, 202, 218, 232	3	96.47 ± 20.16^a	129.69 ± 4.65^b	127.16 ± 22.50^b
6	Cysteine	18.22	59, 100, 132, 220	3	1.14 ± 0.39^a	3.14 ± 0.48^b	8.13 ± 2.15^c
7	Glutamine	23.12	156 , 203, 245, 347	3	ND	0.60 ± 0.20^b	1.68 ± 0.76^c
8	Glutamic acid	19.78	128, 156, 218, 246	3	76.16 ± 14.04^a	669.94 ± 25.81^b	458.06 ± 116.12^c
9	Glycine	7.47	59, 102 , 176, 204	2	78.19 ± 15.56^a	156.19 ± 15.15^b	230.97 ± 44.23^c
		12.03	86, 100, 174 , 248	3			
10	Isoleucine	11.73	100, 117, 158 , 218	2	26.03 ± 6.86^a	15.24 ± 2.95^b	35.00 ± 10.04^c
11	Leucine	8.31	75, 86 , 146, 188	1	6.56 ± 1.48^a	2.20 ± 0.65^b	3.95 ± 1.23^c
12	Lysine	26.08	128, 156, 174 , 317	4	4.23 ± 1.25^a	12.54 ± 1.72^b	21.59 ± 9.23^c
13	Methionine	17.34	61, 128, 156, 176	2	4.34 ± 0.65^a	5.89 ± 0.67^a	8.91 ± 3.25^b
14	Ornithine	23.99	142 , 174, 200, 420	4	0.32 ± 0.11^a	1.63 ± 0.24^b	3.16 ± 1.17^c
15	Proline	11.81	100, 133, 142 , 216	2	51.32 ± 15.75^a	148.97 ± 16.49^b	103.73 ± 29.16^c
16	Pyroglutamic acid	17.37	59, 156 , 230, 258	2	5.28 ± 0.94^a	17.40 ± 2.28^b	33.00 ± 9.31^c

17	Serine	13.45	100, 133, 204 , 218	3	1305.91 ± 306.68 ^a	43.10 ± 5.43 ^b	54.72 ± 15.26 ^b
18	Threonine	11.75	57, 117 , 130, 219	2	400.02 ± 108.63 ^a	29.47 ± 3.49 ^b	42.02 ± 12.02 ^b
		14.08	101, 117, 218 , 291	3			
19	Tryptophan	30.92	100, 202 , 218, 291	3	ND	2.96 ± 0.59 ^b	4.44 ± 2.75 ^b
20	Tyrosine	26.37	100, 179, 218 , 280	3	1.03 ± 0.15 ^a	22.60 ± 3.00 ^b	36.46 ± 14.88 ^c
21	Valine	6.72	55, 72 , 130, 146	1	69.38 ± 17.30 ^a	50.55 ± 7.69 ^a	91.39 ± 24.40 ^b
		9.74	100, 133, 144 , 218	2			
Organic acid							
22	Aminomalononic acid	16.16	133, 174, 218 , 320	3	1.23 ± 0.44 ^a	4.65 ± 0.67 ^b	10.64 ± 3.68 ^c
23	Fumaric acid	13.27	133, 155, 217, 245	2	1.68 ± 0.45 ^a	5.33 ± 0.50 ^b	3.87 ± 0.80 ^c
24	Lactic acid	6.05	117 , 133, 191, 219	2	106.90 ± 31.72 ^a	326.20 ± 59.15 ^b	752.36 ± 103.85 ^c
25	Malic acid	16.67	133, 233 , 245, 335	3	3.96 ± 1.06 ^a	12.59 ± 1.06 ^b	10.42 ± 2.19 ^c
26	Succinic acid	12.31	55, 129, 172, 247	2	ND	0.28 ± 0.12 ^b	0.54 ± 0.23 ^c
Purine							
27	Hypoxanthine	23.74	193, 206, 265 , 280	2	9.16 ± 3.18 ^a	277.38 ± 15.69 ^b	103.39 ± 29.76 ^c
28	Inosine	36.11	217 , 230, 245, 281	4	2.10 ± 1.64 ^a	2.59 ± 0.20 ^a	11.70 ± 2.62 ^b
29	Guanine	29.58	264, 280, 352 , 367	3	0.34 ± 0.20 ^a	0.63 ± 0.06 ^a	13.01 ± 4.54 ^b
30	Xanthine	27.81	279, 294, 353 , 368	3	ND	14.63 ± 1.28 ^b	ND
Pyrimidine							
31	Uracil	12.86	99, 113, 241 , 256	2	0.43 ± 0.14 ^a	21.81 ± 2.88 ^b	20.43 ± 5.47 ^b
32	Uridine	34.33	103, 169, 217 , 259	3	2.44 ± 0.65 ^a	39.27 ± 4.34 ^b	17.18 ± 4.81 ^c
Sugar							
33	Glucose	27.13	129, 191, 204 , 217	5	34.32 ± 3.38 ^a	47.94 ± 3.83 ^b	51.33 ± 19.24 ^b

34	Glyceric acid	12.68	133, 189 , 292, 307	3	0.34 ± 0.17^a	3.53 ± 0.84^b	0.95 ± 0.22^c
35	Glucose-6-phosphate	33.49	204, 299, 357, 387	6	6.36 ± 2.55^a	25.50 ± 2.10^b	40.98 ± 11.97^c
36	Mannose-6-phosphate	32.48	299, 357, 387 , 471	6(1MEOX)	1.40 ± 0.57^a	5.35 ± 0.78^b	5.89 ± 1.29^b
37	Ribose	21.02	103 , 189, 217, 307	4(1MEOX)	ND	3.27 ± 0.29^b	12.05 ± 4.36^c
Other							
38	Creatinine	18.10	100, 115 , 143, 329	3	3.92 ± 1.07^a	10.96 ± 1.49^b	15.52 ± 3.70^c
39	Phosphoric acid	22.88	103, 299, 315, 357	4	68.59 ± 12.93^a	19.67 ± 6.06^b	15.94 ± 7.56^b

Supplementary Table S2. Lipid species identified from extracts of human epidermal melanocytes (HEMn-LP) and melanoma cells (A375, A2058) using nanoESI-MS/MS.

Lipid molecular species	Ion species	m/z	MS/MS fragment ion (m/z)
Positive ion mode			
Phosphatidylcholine (PC)			
PC (14:0/16:0)	[M + H] ⁺	706	450 [lyso-PC(14:0) – H ₂ O + H] ⁺ ; 468 [lyso-PC(14:0) + H] ⁺ ; 478 [lyso-PC(16:0) – H ₂ O + H] ⁺ ; 496 [lyso-PC(16:0) + H] ⁺ ; 523 [M – C ₅ H ₁₄ NO ₄ P + H] ⁺ ; 647 [M – C ₃ H ₉ N + H] ⁺ ; 688 [M – H ₂ O + H] ⁺
PC (16:0/16:1)	[M + H] ⁺	732	476 [lyso-PC(16:1) – H ₂ O + H] ⁺ ; 478 [lyso-PC(16:0) – H ₂ O + H] ⁺ ; 494 [lyso-PC(16:1) + H] ⁺ ; 496 [lyso-PC(16:0) + H] ⁺ ; 549 [M – C ₅ H ₁₄ NO ₄ P + H] ⁺ ; 673 [M – C ₃ H ₉ N + H] ⁺ ; 714 [M – H ₂ O + H] ⁺
PC (16:0/16:0)	[M + H] ⁺	734	478 [lyso-PC(16:0) – H ₂ O + H] ⁺ ; 496 [lyso-PC(16:0) + H] ⁺ ; 551 [M – C ₅ H ₁₄ NO ₄ P + H] ⁺ ; 675 [M – C ₃ H ₉ N + H] ⁺ ; 716 [M – H ₂ O + H] ⁺
PC (16:1/18:1)	[M + H] ⁺	758	476 [lyso-PC(16:1) – H ₂ O + H] ⁺ ; 494 [lyso-PC(16:1) + H] ⁺ ; 504 [lyso-PC(18:1) – H ₂ O + H] ⁺ ; 522 [lyso-PC(18:1) + H] ⁺ ; 575 [M – C ₅ H ₁₄ NO ₄ P + H] ⁺ ; 699 [M – C ₃ H ₉ N + H] ⁺ ; 740 [M – H ₂ O + H] ⁺
PC (16:0/18:1)	[M + H] ⁺	760	478 [lyso-PC(16:0) – H ₂ O + H] ⁺ ; 496 [lyso-PC(16:0) + H] ⁺ ; 504 [lyso-PC(18:1) – H ₂ O + H] ⁺ ; 522 [lyso-PC(18:1) + H] ⁺ ; 577 [M – C ₅ H ₁₄ NO ₄ P + H] ⁺ ; 701 [M – C ₃ H ₉ N + H] ⁺ ; 742 [M – H ₂ O + H] ⁺
PC (16:0/18:0)	[M + H] ⁺	762	478 [lyso-PC(16:0) – H ₂ O + H] ⁺ ; 496 [lyso-PC(16:0) + H] ⁺ ; 506 [lyso-PC(18:0) – H ₂ O + H] ⁺ ; 524 [lyso-PC(18:0) + H] ⁺ ; 579 [M – C ₅ H ₁₄ NO ₄ P + H] ⁺ ; 703 [M – C ₃ H ₉ N + H] ⁺ ; 744 [M – H ₂ O + H] ⁺
PC (16:0/19:0)	[M + H] ⁺	776	478 [lyso-PC(16:0) – H ₂ O + H] ⁺ ; 496 [lyso-PC(16:0) + H] ⁺ ; 520 [lyso-PC(19:0) – H ₂ O + H] ⁺ ; 538 [lyso-PC(19:0) + H] ⁺ ; 593 [M – C ₅ H ₁₄ NO ₄ P + H] ⁺ ; 717 [M – C ₃ H ₉ N + H] ⁺ ; 758 [M – H ₂ O + H] ⁺
PC (18:1/18:1)	[M + H] ⁺	786	504 [lyso-PC(18:1) – H ₂ O + H] ⁺ ; 522 [lyso-PC(18:1) + H] ⁺ ; 603 [M – C ₅ H ₁₄ NO ₄ P

			+ H] ⁺ ; 727 [M – C ₃ H ₉ N + H] ⁺ ; 768 [M – H ₂ O + H] ⁺
PC (18:0/18:1)	[M + H] ⁺	788	504 [lyso-PC(18:1) – H ₂ O + H] ⁺ ; 506 [lyso-PC(18:0) – H ₂ O + H] ⁺ ; 522 [lyso-PC(18:1) + H] ⁺ ; 524 [lyso-PC(18:0) + H] ⁺ ; 605 [M – C ₅ H ₁₄ NO ₄ P + H] ⁺ ; 729 [M – C ₃ H ₉ N + H] ⁺ ; 770 [M – H ₂ O + H] ⁺
Plasmenylphosphatidylcholine (plasmenyl-PC)			
plasmenyl-PC (16:0/18:0)	[M + Na] ⁺	768	279 [(M + Na) – C ₅ H ₁₄ NO ₄ P – Na – C18:0] ⁺ ; 563 [(M + Na) – C ₅ H ₁₄ NO ₄ P – Na + H] ⁺ ; 585 [(M + Na) – C ₅ H ₁₄ NO ₄ P] ⁺ ; 709 [(M + Na) – C ₃ H ₉ N] ⁺
Plasmenylphosphatidylethanolamine (plasmenyl-PE)			
plasmenyl-PE (16:0/22:6)	[M + H] ⁺	748	266 [C16:0 ether + C ₂ H ₈ NO ₃ P – H ₃ PO ₄] ⁺ ; 364 [C16:0 ether + C ₂ H ₈ NO ₃ P] ⁺ ; 385 [lyso(p-16:0) – C ₂ H ₈ NO ₃ P + H] ⁺ ; 607 [M – H ₂ O + H] ⁺
plasmenyl-PE (16:0/22:5)	[M + Na] ⁺	772	386 [C16:0 ether + C ₂ H ₈ NO ₃ P + Na] ⁺ ; 631 [M – C ₂ H ₈ NO ₃ P + Na] ⁺ ; 649 [M – C ₂ H ₈ NO ₃ P + H ₂ O + Na] ⁺ ; 729 [M – C ₂ H ₅ N + Na] ⁺ ; 754 [M – H ₂ O + Na] ⁺
Sphingomyelin (SM)			
SM (d18:1/22:0)	[M + Na] ⁺	809	626 [M – C ₅ H ₁₄ NO ₄ P] ⁺ ; 732 [M – C ₃ H ₉ N – H ₂ O] ⁺ ; 750 [M – C ₃ H ₉ N] ⁺ ; 791 [M – H ₂ O] ⁺
SM (d18:0/22:0)	[M + Na] ⁺	811	628 [M – C ₅ H ₁₄ NO ₄ P] ⁺ ; 734 [M – C ₃ H ₉ N – H ₂ O] ⁺ ; 752 [M – C ₃ H ₉ N] ⁺ ; 793 [M – H ₂ O] ⁺
Negative ion mode			
Ceramide (Cer)			
Cer (d18:1/16:0)	[M – H] [–]	536	237 [M – H – 299] [–] ; 254 [M – H – 282] [–] ; 255 [M – H – 281] [–] ; 280 [M – H – 256] [–] ; 296 [M – H – 240] [–] ; 488 [M – H – CH ₂ O – H ₂ O] [–] ; 504 [M – H – CH ₃ OH] [–] ; 506 [M – H – CH ₂ O] [–] ; 518 [M – H – H ₂ O] [–]
Cer (d18:1/18:1)	[M – H] [–]	562	263 [M – H – 299] [–] ; 280 [M – H – 282] [–] ; 281 [M – H – 281] [–] ; 306 [M – H – 256] [–] ; 322 [M – H – 240] [–] ; 514 [M – H – CH ₂ O – H ₂ O] [–] ; 530 [M – H – CH ₃ OH] [–] ; 532

			$[M - H - CH_2O]^-$; 544 $[M - H - H_2O]^-$
Cer (d18:1/18:0)	$[M - H]^-$	564	265 $[M - H - 299]^-$; 282 $[M - H - 282]^-$; 283 $[M - H - 281]^-$; 308 $[M - H - 256]^-$; 324 $[M - H - 240]^-$; 516 $[M - H - CH_2O - H_2O]^-$; 532 $[M - H - CH_3OH]^-$; 534 $[M - H - CH_2O]^-$; 546 $[M - H - H_2O]^-$
Cer (d18:1/20:0)	$[M - H]^-$	592	293 $[M - H - 299]^-$; 310 $[M - H - 282]^-$; 311 $[M - H - 281]^-$; 336 $[M - H - 256]^-$; 352 $[M - H - 240]^-$; 544 $[M - H - CH_2O - H_2O]^-$; 560 $[M - H - CH_3OH]^-$; 562 $[M - H - CH_2O]^-$; 574 $[M - H - H_2O]^-$
Cer (d18:1/22:0)	$[M - H]^-$	620	321 $[M - H - 299]^-$; 338 $[M - H - 282]^-$; 339 $[M - H - 281]^-$; 364 $[M - H - 256]^-$; 380 $[M - H - 240]^-$; 572 $[M - H - CH_2O - H_2O]^-$; 588 $[M - H - CH_3OH]^-$; 590 $[M - H - CH_2O]^-$; 602 $[M - H - H_2O]^-$
Plasmenylphosphatidylethanolamine (plasmenyl-PE)			
plasmenyl-PE (16:0/18:1)	$[M - H]^-$	700	281 $[C18:1 - H]^-$; 418 $[lyso-PE(P-16:0) - H_2O - H]^-$; 436 $[lyso-PE(P-16:0) - H]^-$
plasmenyl-PE (16:0/20:4)	$[M - H]^-$	722	303 $[C20:4 - H]^-$; 418 $[lyso-PE(P-16:0) - H_2O - H]^-$; 436 $[lyso-PE(P-16:0) - H]^-$
plasmenyl-PE (18:0/20:4)	$[M - H]^-$	750	303 $[C20:4 - H]^-$; 446 $[lyso-PE(P-18:0) - H_2O - H]^-$; 464 $[lyso-PE(P-18:0) - H]^-$
plasmenyl-PE (18:0/22:5)	$[M - H]^-$	776	329 $[C22:5 - H]^-$; 446 $[lyso-PE(P-18:0) - H_2O - H]^-$; 464 $[lyso-PE(P-18:0) - H]^-$
Phosphatidylethanolamine (PE)			
PE (16:1/18:1)	$[M - H]^-$	714	253 $[C16:1 - H]^-$; 281 $[C18:1 - H]^-$; 432 $[lyso-PE(16:1) - H_2O - H]^-$; 450 $[lyso-PE(16:1) - H]^-$; 460 $[lyso-PE(18:1) - H_2O - H]^-$; 478 $[lyso-PE(18:1) - H]^-$
PE (16:0/18:1)	$[M - H]^-$	716	255 $[C16:0 - H]^-$; 281 $[C18:1 - H]^-$; 434 $[lyso-PE(16:0) - H_2O - H]^-$; 452 $[lyso-PE(16:0) - H]^-$; 460 $[lyso-PE(18:1) - H_2O - H]^-$; 478 $[lyso-PE(18:1) - H]^-$
PE (18:1/18:1)	$[M - H]^-$	742	281 $[C18:1 - H]^-$; 460 $[lyso-PE(18:1) - H_2O - H]^-$; 478 $[lyso-PE(18:1) - H]^-$
PE (18:0/18:1)	$[M - H]^-$	744	281 $[C18:1 - H]^-$; 283 $[C18:0 - H]^-$; 460 $[lyso-PE(18:1) - H_2O - H]^-$; 462 $[lyso-PE(18:0) - H_2O - H]^-$; 478 $[lyso-PE(18:1) - H]^-$; 480 $[lyso-PE(18:0) - H]^-$
PE (18:1/20:4)	$[M - H]^-$	764	281 $[C18:1 - H]^-$; 303 $[C20:4 - H]^-$; 460 $[lyso-PE(18:1) - H_2O - H]^-$; 478 $[lyso-PE(18:1) - H]^-$; 482 $[lyso-PE(20:4) - H_2O - H]^-$; 500 $[lyso-PE(20:4) - H]^-$

PE (18:0/20:4)	[M – H] [–]	766	283 [C18:0 – H] [–] ; 303 [C20:4 – H] [–] ; 462 [lyso-PE(18:0) – H ₂ O – H] [–] ; 480 [lyso-PE(18:0) – H] [–] ; 482 [lyso-PE(20:4) – H ₂ O – H] [–] ; 500 [lyso-PE(20:4) – H] [–]
PE (17:0/22:5)	[M – H] [–]	778	269 [C17:0 – H] [–] ; 329 [C22:5 – H] [–] ; 448 [lyso-PE(17:0) – H ₂ O – H] [–] ; 466 [lyso-PE(17:0) – H] [–] ; 508 [lyso-PE(22:5) – H ₂ O – H] [–] ; 526 [lyso-PE(22:5) – H] [–]
PE (18:0/22:5)	[M – H] [–]	792	283 [C18:0 – H] [–] ; 329 [C22:5 – H] [–] ; 462 [lyso-PE(18:0) – H ₂ O – H] [–] ; 480 [lyso-PE(18:0) – H] [–] ; 508 [lyso-PE(22:5) – H ₂ O – H] [–] ; 526 [lyso-PE(22:5) – H] [–]
Cardiolipin (CL)			
CL (18:1/18:1/18:1/18:1)	[M – 2H] ^{2–}	727	281 [C18:1 – H] [–] ; 417 [C18:1 + C ₃ H ₆ PO ₄ – H] [–] ; 463 [lyso-CL(FA1/FA2) – 2H] ^{2–} [lyso-CL(FA3/FA4) – 2H] ^{2–} ; 595 [lyso-CL(FA2/FA3/FA4) – 2H] ^{2–} [lyso-CL(FA1/FA2/FA4) – 2H] ^{2–}
Phosphatidylglycerol (PG)			
PG (16:1/18:1)	[M – H] [–]	745	253 [C16:1 – H] [–] ; 281 [C18:1 – H] [–] ; 389 [lyso-PG(16:1) – C ₃ H ₆ O ₂ – H] [–] ; 417 [lyso-PG(18:1) – C ₃ H ₆ O ₂ – H] [–] ; 463 [lyso-PG(16:1) – H ₂ O – H] [–] ; 481 [lyso-PG(16:1) – H] [–] ; 491 [lyso-PG(18:1) – H ₂ O – H] [–] ; 509 [lyso-PG(18:1) – H] [–]
PG (16:0/18:1)	[M – H] [–]	747	255 [C16:0 – H] [–] ; 281 [C18:1 – H] [–] ; 391 [lyso-PG(16:0) – C ₃ H ₆ O ₂ – H] [–] ; 417 [lyso-PG(18:1) – C ₃ H ₆ O ₂ – H] [–] ; 465 [lyso-PG(16:0) – H ₂ O – H] [–] ; 483 [lyso-PG(16:0) – H] [–] ; 491 [lyso-PG(18:1) – H ₂ O – H] [–] ; 509 [lyso-PG(18:1) – H] [–]
PG (18:1/18:2)	[M – H] [–]	771	279 [C18:2 – H] [–] ; 281 [C18:1 – H] [–] ; 415 [lyso-PG(18:2) – C ₃ H ₆ O ₂ – H] [–] ; 417 [lyso-PG(18:1) – C ₃ H ₆ O ₂ – H] [–] ; 489 [lyso-PG(18:2) – H ₂ O – H] [–] ; 491 [lyso-PG(18:1) – H ₂ O – H] [–] ; 507 [lyso-PG(18:2) – H] [–] ; 509 [lyso-PG(18:1) – H] [–]
PG (18:1/18:1)	[M – H] [–]	773	281 [C18:1 – H] [–] ; 417 [lyso-PG(18:1) – C ₃ H ₆ O ₂ – H] [–] ; 491 [lyso-PG(18:1) – H ₂ O – H] [–] ; 509 [lyso-PG(18:1) – H] [–]
PG (18:0/18:1)	[M – H] [–]	775	281 [C18:1 – H] [–] ; 283 [C18:0 – H] [–] ; 417 [lyso-PG(18:1) – C ₃ H ₆ O ₂ – H] [–] ; 419 [lyso-PG(18:0) – C ₃ H ₆ O ₂ – H] [–] ; 491 [lyso-PG(18:1) – H ₂ O – H] [–] ; 493 [lyso-PG(18:0) – H ₂ O – H] [–] ; 509 [lyso-PG(18:1) – H] [–] ; 511 [lyso-PG(18:0) – H] [–]
PG (18:1/20:1)	[M – H] [–]	801	281 [C18:1 – H] [–] ; 309 [C20:1 – H] [–] ; 417 [lyso-PG(18:1) – C ₃ H ₆ O ₂ – H] [–] ; 445 [lyso-PG(20:1) – C ₃ H ₆ O ₂ – H] [–] ; 491 [lyso-PG(18:1) – H ₂ O – H] [–] ; 509 [lyso-PG(18:1) – H] [–] ; 519 [lyso-PG(20:1) – H ₂ O – H] [–] ; 537 [lyso-PG(20:1) – H] [–]

PG (18:0/22:6)	[M – H] [–]	821	283 [C18:0 – H] [–] ; 327 [C22:6 – H] [–] ; 419 [lyso-PG(18:0) – C ₃ H ₆ O ₂ – H] [–] ; 463 [lyso-PG(22:6) – C ₃ H ₆ O ₂ – H] [–] ; 493 [lyso-PG(18:0) – H ₂ O – H] [–] ; 511 [lyso-PG(18:0) – H] [–] ; 537 [lyso-PG(22:6) – H ₂ O – H] [–] ; 555 [lyso-PG(22:6) – H] [–]
Phosphatidylserine (PS)			
PS (16:1/18:1)	[M – H] [–]	758	253 [C16:1 – H] [–] ; 281 [C18:1 – H] [–] ; 389 [lyso-PS(16:1) – C ₃ H ₅ NO ₂ – H] [–] ; 407 [lyso-PS(16:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 417 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ – H] [–] ; 435 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 671 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (16:0/18:1)	[M – H] [–]	760	225 [C16:0 – H] [–] ; 281 [C18:1 – H] [–] ; 391 [lyso-PS(16:0) – C ₃ H ₅ NO ₂ – H] [–] ; 409 [lyso-PS(16:0) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 417 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ – H] [–] ; 435 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 673 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (16:0/18:0)	[M – H] [–]	762	255 [C16:0 – H] [–] ; 283 [C18:0 – H] [–] ; 391 [lyso-PS(16:0) – C ₃ H ₅ NO ₂ – H] [–] ; 409 [lyso-PS(16:0) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 419 [lyso-PS(18:0) – C ₃ H ₅ NO ₂ – H] [–] ; 437 [lyso-PS(18:0) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 675 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (18:1/18:2)	[M – H] [–]	784	279 [C18:2 – H] [–] ; 281 [C18:1 – H] [–] ; 415 [lyso-PS(18:2) – C ₃ H ₅ NO ₂ – H] [–] ; 417 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ – H] [–] ; 433 [lyso-PS(18:2) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 435 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 697 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (18:1/18:1)	[M – H] [–]	786	281 [C18:1 – H] [–] ; 417 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ – H] [–] ; 435 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 699 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (18:0/18:1)	[M – H] [–]	788	281 [C18:1 – H] [–] ; 283 [C18:0 – H] [–] ; 417 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ – H] [–] ; 419 [lyso-PS(18:0) – C ₃ H ₅ NO ₂ – H] [–] ; 435 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 437 [lyso-PS(18:0) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 701 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (18:0/18:0)	[M – H] [–]	790	283 [C18:0 – H] [–] ; 419 [lyso-PS(18:0) – C ₃ H ₅ NO ₂ – H] [–] ; 437 [lyso-PS(18:0) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 703 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (18:1/20:4)	[M – H] [–]	808	281 [C18:1 – H] [–] ; 303 [C20:4 – H] [–] ; 417 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ – H] [–] ; 435 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 439 [lyso-PS(20:4) – C ₃ H ₅ NO ₂ – H] [–] ; 457 [lyso-PS(20:4) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 721 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (18:0/20:4)	[M – H] [–]	810	283 [C18:0 – H] [–] ; 303 [C20:4 – H] [–] ; 419 [lyso-PS(18:0) – C ₃ H ₅ NO ₂ – H] [–] ; 437 [lyso-PS(18:0) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 439 [lyso-PS(20:4) – C ₃ H ₅ NO ₂ – H] [–] ; 457 [lyso-PS(20:4) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 723 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (18:0/20:3)	[M – H] [–]	812	283 [C18:0 – H] [–] ; 305 [C20:3 – H] [–] ; 419 [lyso-PS(18:0) – C ₃ H ₅ NO ₂ – H] [–] ; 437 [lyso-PS(18:0) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 441 [lyso-PS(20:3) – C ₃ H ₅ NO ₂ – H] [–] ; 459

			[lyso-PS(20:3) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 725 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (18:1/20:1)	[M – H] [–]	814	281 [C18:1 – H] [–] ; 309 [C20:1 – H] [–] ; 417 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ – H] [–] ; 435 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 445 [lyso-PS(20:1) – C ₃ H ₅ NO ₂ – H] [–] ; 463 [lyso-PS(20:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 727 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (18:1/20:0)	[M – H] [–]	816	281 [C18:1 – H] [–] ; 311 [C20:0 – H] [–] ; 417 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ – H] [–] ; 435 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 447 [lyso-PS(20:0) – C ₃ H ₅ NO ₂ – H] [–] ; 465 [lyso-PS(20:0) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 729 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (18:1/22:1)	[M – H] [–]	842	281 [C18:1 – H] [–] ; 337 [C22:1 – H] [–] ; 417 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ – H] [–] ; 435 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 473 [lyso-PS(22:1) – C ₃ H ₅ NO ₂ – H] [–] ; 491 [lyso-PS(22:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 755 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (18:1/22:0)	[M – H] [–]	844	281 [C18:1 – H] [–] ; 339 [C22:0 – H] [–] ; 417 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ – H] [–] ; 435 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 475 [lyso-PS(22:0) – C ₃ H ₅ NO ₂ – H] [–] ; 493 [lyso-PS(22:0) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 757 [M – C ₃ H ₅ NO ₂ – H] [–]
PS (18:1/24:1)	[M – H] [–]	870	281 [C18:1 – H] [–] ; 365 [C24:1 – H] [–] ; 417 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ – H] [–] ; 435 [lyso-PS(18:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 501 [lyso-PS(24:1) – C ₃ H ₅ NO ₂ – H] [–] ; 519 [lyso-PS(24:1) – C ₃ H ₅ NO ₂ + H ₂ O – H] [–] ; 783 [M – C ₃ H ₅ NO ₂ – H] [–]
Phosphatidylinositol (PI)			
PI (16:1/18:1)	[M – H] [–]	833	241 [C ₆ H ₁₀ O ₈ P] [–] ; 253 [C16:1 – H] [–] ; 281 [C18:1 – H] [–] ; 297 [C ₉ H ₁₄ O ₉ P] [–] ; 315 [C ₉ H ₁₆ O ₁₀ P] [–] ; 389 [lyso-PI(16:1) – C ₆ H ₁₂ O ₆ – H] [–] ; 417 [lyso-PI(18:1) – C ₆ H ₁₂ O ₆ – H] [–] ; 551 [lyso-PI(16:1) – H ₂ O – H] [–] ; 569 [lyso-PI(16:1) – H] [–] ; 579 [lyso-PI(18:1) – H ₂ O – H] [–] ; 597 [lyso-PI(18:1) – H] [–]
PI (16:0/18:1)	[M – H] [–]	835	241 [C ₆ H ₁₀ O ₈ P] [–] ; 255 [C16:0 – H] [–] ; 281 [C18:1 – H] [–] ; 297 [C ₉ H ₁₄ O ₉ P] [–] ; 315 [C ₉ H ₁₆ O ₁₀ P] [–] ; 391 [lyso-PI(16:0) – C ₆ H ₁₂ O ₆ – H] [–] ; 417 [lyso-PI(18:1) – C ₆ H ₁₂ O ₆ – H] [–] ; 553 [lyso-PI(16:0) – H ₂ O – H] [–] ; 571 [lyso-PI(16:0) – H] [–] ; 579 [lyso-PI(18:1) – H ₂ O – H] [–] ; 597 [lyso-PI(18:1) – H] [–]
PI (16:0/18:0)	[M – H] [–]	837	241 [C ₆ H ₁₀ O ₈ P] [–] ; 255 [C16:0 – H] [–] ; 283 [C18:0 – H] [–] ; 297 [C ₉ H ₁₄ O ₉ P] [–] ; 315 [C ₉ H ₁₆ O ₁₀ P] [–] ; 391 [lyso-PI(16:0) – C ₆ H ₁₂ O ₆ – H] [–] ; 419 [lyso-PI(18:0) – C ₆ H ₁₂ O ₆ – H] [–] ; 553 [lyso-PI(16:0) – H ₂ O – H] [–] ; 571 [lyso-PI(16:0) – H] [–] ; 581 [lyso-PI(18:0) – H ₂ O – H] [–] ; 599 [lyso-PI(18:0) – H] [–]
PI (18:1/18:2)	[M – H] [–]	859	241 [C ₆ H ₁₀ O ₈ P] [–] ; 279 [C18:2 – H] [–] ; 281 [C18:1 – H] [–] ; 297 [C ₉ H ₁₄ O ₉ P] [–] ; 315 [C ₉ H ₁₆ O ₁₀ P] [–] ; 415 [lyso-PI(18:2) – C ₆ H ₁₂ O ₆ – H] [–] ; 417 [lyso-PI(18:1) – C ₆ H ₁₂ O ₆ –

			H] ⁻ ; 577 [lyso-PI(18:2) – H ₂ O – H] ⁻ ; 579 [lyso-PI(18:1) – H ₂ O – H] ⁻ ; 595 [lyso-PI(18:2) – H] ⁻ ; 597 [lyso-PI(18:1) – H] ⁻
PI (18:1/18:1)	[M – H] ⁻	861	241 [C ₆ H ₁₀ O ₈ P] ⁻ ; 281 [C18:1 – H] ⁻ ; 297 [C ₉ H ₁₄ O ₉ P] ⁻ ; 315 [C ₉ H ₁₆ O ₁₀ P] ⁻ ; 417 [lyso-PI(18:1) – C ₆ H ₁₂ O ₆ – H] ⁻ ; 579 [lyso-PI(18:1) – H ₂ O – H] ⁻ ; 597 [lyso-PI(18:1) – H] ⁻
PI (18:0/18:1)	[M – H] ⁻	863	241 [C ₆ H ₁₀ O ₈ P] ⁻ ; 281 [C18:1 – H] ⁻ ; 283 [C18:0 – H] ⁻ ; 297 [C ₉ H ₁₄ O ₉ P] ⁻ ; 315 [C ₉ H ₁₆ O ₁₀ P] ⁻ ; 417 [lyso-PI(18:1) – C ₆ H ₁₂ O ₆ – H] ⁻ ; 419 [lyso-PI(18:0) – C ₆ H ₁₂ O ₆ – H] ⁻ ; 579 [lyso-PI(18:1) – H ₂ O – H] ⁻ ; 581 [lyso-PI(18:0) – H ₂ O – H] ⁻ ; 597 [lyso-PI(18:1) – H] ⁻ ; 599 [lyso-PI(18:0) – H] ⁻
PI (18:0/18:0)	[M – H] ⁻	865	241 [C ₆ H ₁₀ O ₈ P] ⁻ ; 283 [C18:0 – H] ⁻ ; 297 [C ₉ H ₁₄ O ₉ P] ⁻ ; 315 [C ₉ H ₁₆ O ₁₀ P] ⁻ ; 419 [lyso-PI(18:0) – C ₆ H ₁₂ O ₆ – H] ⁻ ; 581 [lyso-PI(18:0) – H ₂ O – H] ⁻ ; 599 [lyso-PI(18:0) – H] ⁻
PI (18:1/20:4)	[M – H] ⁻	883	241 [C ₆ H ₁₀ O ₈ P] ⁻ ; 281 [C18:1 – H] ⁻ ; 297 [C ₉ H ₁₄ O ₉ P] ⁻ ; 303 [C20:4 – H] ⁻ ; 315 [C ₉ H ₁₆ O ₁₀ P] ⁻ ; 417 [lyso-PI(18:1) – C ₆ H ₁₂ O ₆ – H] ⁻ ; 439 [lyso-PI(20:4) – C ₆ H ₁₂ O ₆ – H] ⁻ ; 579 [lyso-PI(18:1) – H ₂ O – H] ⁻ ; 597 [lyso-PI(18:1) – H] ⁻ ; 601 [lyso-PI(20:4) – H ₂ O – H] ⁻ ; 619 [lyso-PI(20:4) – H] ⁻
PI (18:0/20:4)	[M – H] ⁻	885	241 [C ₆ H ₁₀ O ₈ P] ⁻ ; 283 [C18:0 – H] ⁻ ; 297 [C ₉ H ₁₄ O ₉ P] ⁻ ; 303 [C20:4 – H] ⁻ ; 315 [C ₉ H ₁₆ O ₁₀ P] ⁻ ; 419 [lyso-PI(18:0) – C ₆ H ₁₂ O ₆ – H] ⁻ ; 439 [lyso-PI(20:4) – C ₆ H ₁₂ O ₆ – H] ⁻ ; 581 [lyso-PI(18:0) – H ₂ O – H] ⁻ ; 599 [lyso-PI(18:0) – H] ⁻ ; 601 [lyso-PI(20:4) – H ₂ O – H] ⁻ ; 619 [lyso-PI(20:4) – H] ⁻
PI (18:0/20:3)	[M – H] ⁻	887	241 [C ₆ H ₁₀ O ₈ P] ⁻ ; 283 [C18:0 – H] ⁻ ; 297 [C ₉ H ₁₄ O ₉ P] ⁻ ; 305 [C20:3 – H] ⁻ ; 315 [C ₉ H ₁₆ O ₁₀ P] ⁻ ; 419 [lyso-PI(18:0) – C ₆ H ₁₂ O ₆ – H] ⁻ ; 441 [lyso-PI(20:3) – C ₆ H ₁₂ O ₆ – H] ⁻ ; 581 [lyso-PI(18:0) – H ₂ O – H] ⁻ ; 599 [lyso-PI(18:0) – H] ⁻ ; 603 [lyso-PI(20:3) – H ₂ O – H] ⁻ ; 621 [lyso-PI(20:3) – H] ⁻
PI (18:0/20:2)	[M – H] ⁻	889	241 [C ₆ H ₁₀ O ₈ P] ⁻ ; 283 [C18:0 – H] ⁻ ; 297 [C ₉ H ₁₄ O ₉ P] ⁻ ; 307 [C20:2 – H] ⁻ ; 315 [C ₉ H ₁₆ O ₁₀ P] ⁻ ; 419 [lyso-PI(18:0) – C ₆ H ₁₂ O ₆ – H] ⁻ ; 443 [lyso-PI(20:2) – C ₆ H ₁₂ O ₆ – H] ⁻ ; 581 [lyso-PI(18:0) – H ₂ O – H] ⁻ ; 599 [lyso-PI(18:0) – H] ⁻ ; 605 [lyso-PI(20:2) – H ₂ O – H] ⁻ ; 623 [lyso-PI(20:2) – H] ⁻

Supplementary Table S3. Normalized ion intensities of the identified lipids in human epidermal melanocytes (HEMn-LP) and melanoma cell lines (A375, A2058) using DI-MS. Each value represents the mean $\pm$ standard deviation (SD) (n = 10). For simplicity, ‘ $\times 10^4$ ’ and ‘ $\times 10^5$ ’ was omitted from the values for lipids identified in positive mode and negative modes, respectively.

No.	Lipid species	Ion species	m/z	HEMn-LP	A375	A2058
Positive ion mode						
Phosphatidylcholine (PC)						
1	PC (14:0/16:0)	[M + H] ⁺	706	7.60 $\pm$ 2.55 ^a	2.45 $\pm$ 0.21 ^b	5.04 $\pm$ 0.68 ^c
2	PC (16:0/16:1)	[M + H] ⁺	732	19.20 $\pm$ 6.45 ^a	8.98 $\pm$ 0.61 ^b	21.12 $\pm$ 3.07 ^a
3	PC (16:0/16:0)	[M + H] ⁺	734	14.85 $\pm$ 5.25 ^a	3.95 $\pm$ 0.28 ^b	8.58 $\pm$ 1.19 ^c
4	PC (16:1/18:1)	[M + H] ⁺	758	25.96 $\pm$ 8.46 ^a	7.05 $\pm$ 0.46 ^b	18.09 $\pm$ 2.75 ^c
5	PC (16:0/18:1)	[M + H] ⁺	760	64.05 $\pm$ 23.62 ^a	25.38 $\pm$ 1.78 ^b	52.23 $\pm$ 7.57 ^a
6	PC (16:0/18:0)	[M + H] ⁺	762	15.92 $\pm$ 4.64 ^a	4.56 $\pm$ 0.30 ^b	8.38 $\pm$ 1.01 ^c
7	PC (16:0/19:0)	[M + H] ⁺	776	3.83 $\pm$ 1.21 ^a	0.68 $\pm$ 0.03 ^b	1.12 $\pm$ 0.18 ^b
8	PC (18:1/18:1)	[M + H] ⁺	786	172.49 $\pm$ 48.97 ^a	26.18 $\pm$ 2.10 ^b	55.60 $\pm$ 7.39 ^b
9	PC (18:0/18:1)	[M + H] ⁺	788	113.71 $\pm$ 32.12 ^a	16.07 $\pm$ 1.34 ^b	32.52 $\pm$ 4.67 ^b
Plasmenylphosphatidylcholine (plasmenyl-PC)						
10	plasmenyl-PC (16:0/18:0)	[M + Na] ⁺	768	4.26 $\pm$ 1.36 ^a	2.31 $\pm$ 0.09 ^b	3.90 $\pm$ 0.86 ^a
Plasmenylphosphatidylethanolamine (plasmenyl-PE)						
11	plasmenyl-PE (16:0/22:6)	[M + H] ⁺	748	1.72 $\pm$ 0.65 ^a	1.74 $\pm$ 0.11 ^a	3.12 $\pm$ 0.36 ^b
12	plasmenyl-PE (16:0/22:5)	[M + Na] ⁺	772	3.33 $\pm$ 1.54 ^a	1.86 $\pm$ 0.15 ^b	5.59 $\pm$ 0.86 ^c
Sphingomyelin (SM)						
13	SM (d18:1/22:0)	[M + Na] ⁺	809	10.62 $\pm$ 4.33 ^a	2.21 $\pm$ 0.18 ^b	4.79 $\pm$ 1.26 ^b

Sample	Compound	Ion	Abundance	Mass (Da)	Intensity (a.u.)	Intensity (a.u.)
14	SM (d18:0/22:0)	[M + Na] ⁺	811	6.05 ± 1.94 ^a	0.68 ± 0.04 ^b	0.91 ± 0.14 ^b
	Negative ion mode					
	Ceramide (Cer)					
15	Cer (d18:1/16:0)	[M − H] [−]	536	46.62 ± 11.13 ^a	187.48 ± 34.27 ^b	66.73 ± 27.90 ^a
16	Cer (d18:1/18:1)	[M − H] [−]	562	89.30 ± 24.71 ^a	72.53 ± 14.24 ^{ab}	55.09 ± 18.35 ^b
17	Cer (d18:1/18:0)	[M − H] [−]	564	73.29 ± 18.12 ^a	169.20 ± 31.00 ^b	63.56 ± 18.43 ^a
18	Cer (d18:1/20:0)	[M − H] [−]	592	5.37 ± 1.58 ^a	8.58 ± 1.64 ^b	5.67 ± 1.08 ^a
19	Cer (d18:1/22:0)	[M − H] [−]	620	2.63 ± 0.85 ^a	4.84 ± 0.85 ^b	2.70 ± 0.69 ^a
	Plasmenylphosphatidylethanolamine (plasmenyl-PE)					
20	plasmenyl-PE (16:0/18:1)	[M − H] [−]	700	29.22 ± 5.12 ^a	18.76 ± 1.06 ^b	31.35 ± 8.31 ^a
21	plasmenyl-PE (16:0/20:4)	[M − H] [−]	722	33.17 ± 5.87 ^a	23.47 ± 1.79 ^b	25.29 ± 6.14 ^b
22	plasmenyl-PE (18:0/20:4)	[M − H] [−]	750	38.54 ± 6.58 ^a	20.36 ± 1.38 ^b	22.64 ± 5.76 ^b
23	plasmenyl-PE (18:0/22:5)	[M − H] [−]	776	22.20 ± 6.27 ^a	14.04 ± 1.53 ^b	29.18 ± 8.76 ^c
	Phosphatidylethanolamine (PE)					
24	PE (16:1/18:1)	[M − H] [−]	714	11.85 ± 2.77 ^a	10.08 ± 0.80 ^a	20.00 ± 5.64 ^b
25	PE (16:0/18:1)	[M − H] [−]	716	21.28 ± 3.87 ^{ab}	20.46 ± 1.44 ^a	26.05 ± 7.64 ^b
26	PE (18:1/18:1)	[M − H] [−]	742	84.67 ± 16.34 ^a	32.35 ± 2.12 ^b	63.00 ± 19.25 ^c
27	PE (18:0/18:1)	[M − H] [−]	744	46.23 ± 10.09 ^a	32.21 ± 2.17 ^b	44.46 ± 12.51 ^a
28	PE (18:1/20:4)	[M − H] [−]	764	25.28 ± 3.84 ^a	14.96 ± 1.28 ^b	24.47 ± 7.09 ^a
29	PE (18:0/20:4)	[M − H] [−]	766	16.79 ± 2.49 ^a	17.90 ± 1.02 ^a	16.27 ± 4.95 ^a
30	PE (17:0/22:5)	[M − H] [−]	778	13.56 ± 2.66 ^a	10.58 ± 0.85 ^a	19.31 ± 6.22 ^b
31	PE (18:0/22:5)	[M − H] [−]	792	27.79 ± 4.56 ^a	22.57 ± 1.80 ^a	36.34 ± 10.76 ^b
	Cardiolipin (CL)					

32	CL (18:1/18:1/18:1/18:1)	$[M - 2H]^{2-}$	727	23.56 ± 4.09^a	4.92 ± 0.33^b	12.44 ± 3.44^c
Phosphatidylglycerol (PG)						
33	PG (16:1/18:1)	$[M - H]^-$	745	21.00 ± 4.42^a	26.33 ± 2.96^a	55.81 ± 13.95^b
34	PG (16:0/18:1)	$[M - H]^-$	747	20.30 ± 3.89^a	49.68 ± 6.88^b	76.11 ± 16.82^c
35	PG (18:1/18:2)	$[M - H]^-$	771	10.81 ± 2.20^a	8.31 ± 1.29^a	18.14 ± 4.86^b
36	PG (18:1/18:1)	$[M - H]^-$	773	89.05 ± 15.80^a	36.78 ± 5.66^b	136.90 ± 36.70^c
37	PG (18:0/18:1)	$[M - H]^-$	775	26.71 ± 4.53^a	25.12 ± 3.06^a	38.11 ± 9.02^b
38	PG (18:1/20:1)	$[M - H]^-$	801	9.60 ± 2.89^a	3.90 ± 0.75^b	10.35 ± 2.41^a
39	PG (18:0/22:6)	$[M - H]^-$	821	19.95 ± 3.22^a	9.12 ± 0.83^b	20.07 ± 4.31^a
Phosphatidylserine (PS)						
40	PS (16:1/18:1)	$[M - H]^-$	758	48.32 ± 9.45^a	29.13 ± 4.65^b	49.88 ± 12.17^a
41	PS (16:0/18:1)	$[M - H]^-$	760	73.91 ± 13.53^a	92.10 ± 12.54^a	130.21 ± 23.80^b
42	PS (16:0/18:0)	$[M - H]^-$	762	28.09 ± 4.47^a	19.48 ± 2.20^b	30.21 ± 5.74^a
43	PS (18:1/18:2)	$[M - H]^-$	784	26.13 ± 6.04^a	18.16 ± 3.41^b	17.22 ± 4.42^b
44	PS (18:1/18:1)	$[M - H]^-$	786	240.79 ± 40.62^a	115.99 ± 18.54^b	156.37 ± 36.01^c
45	PS (18:0/18:1)	$[M - H]^-$	788	487.58 ± 69.41^a	154.23 ± 13.02^b	230.89 ± 41.24^c
46	PS (18:0/18:0)	$[M - H]^-$	790	93.60 ± 15.11^a	52.11 ± 4.46^b	85.32 ± 22.88^a
47	PS (18:1/20:4)	$[M - H]^-$	808	8.63 ± 2.61^a	7.02 ± 1.30^a	12.73 ± 3.19^b
48	PS (18:0/20:4)	$[M - H]^-$	810	13.85 ± 2.08^a	4.09 ± 0.39^b	4.37 ± 0.87^b
49	PS (18:0/20:3)	$[M - H]^-$	812	21.55 ± 2.71^a	5.46 ± 0.38^b	4.91 ± 1.26^b
50	PS (18:1/20:1)	$[M - H]^-$	814	51.79 ± 7.40^a	6.77 ± 0.66^b	16.20 ± 4.07^c
51	PS (18:1/20:0)	$[M - H]^-$	816	24.40 ± 4.33^a	21.77 ± 2.06^a	42.63 ± 12.27^b
52	PS (18:1/22:1)	$[M - H]^-$	842	37.11 ± 5.59^a	7.48 ± 0.67^b	14.39 ± 2.94^c
53	PS (18:1/22:0)	$[M - H]^-$	844	52.20 ± 10.07^a	36.41 ± 4.10^b	62.17 ± 18.72^a
54	PS (18:1/24:1)	$[M - H]^-$	870	23.28 ± 4.81^a	4.52 ± 0.35^b	9.80 ± 1.66^c

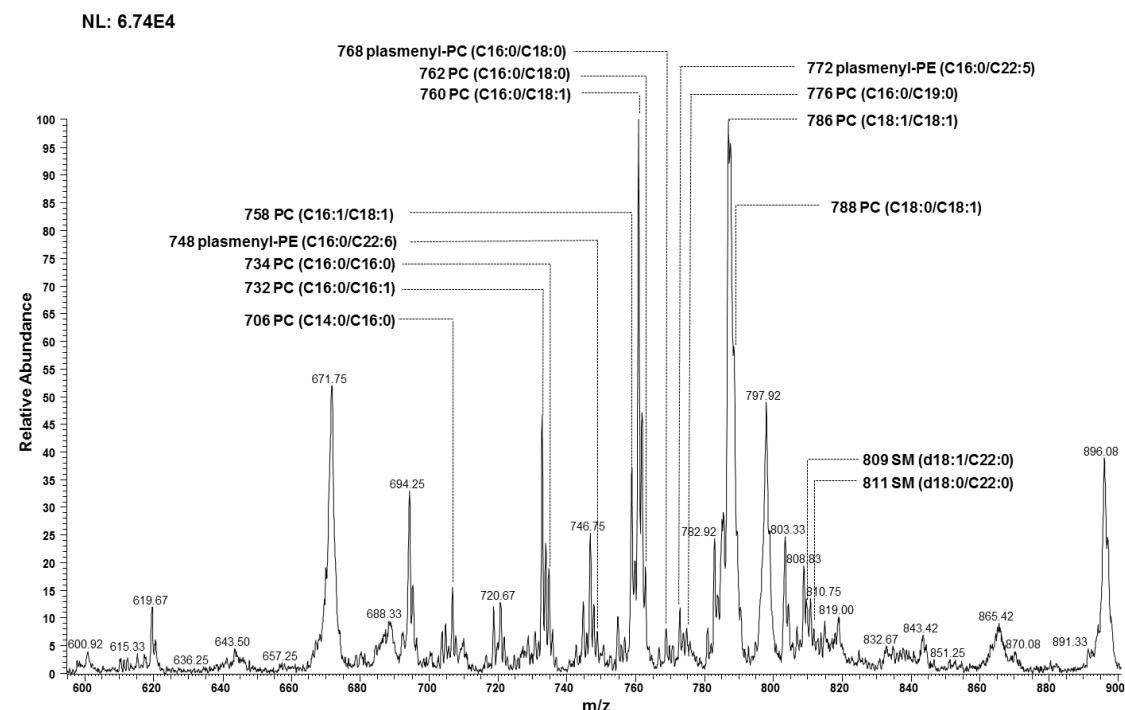
Phosphatidylinositol (PI)						
55	PI (16:1/18:1)	[M – H] [–]	833	26.95 ± 4.18 ^a	18.12 ± 2.06 ^b	54.82 ± 11.17 ^c
56	PI (16:0/18:1)	[M – H] [–]	835	23.90 ± 4.09 ^a	48.38 ± 4.23 ^b	122.77 ± 21.75 ^c
57	PI (16:0/18:0)	[M – H] [–]	837	5.54 ± 1.05 ^a	14.62 ± 1.18 ^b	24.48 ± 3.56 ^c
58	PI (18:1/18:2)	[M – H] [–]	859	12.69 ± 1.89 ^a	11.22 ± 1.14 ^a	16.33 ± 3.61 ^b
59	PI (18:1/18:1)	[M – H] [–]	861	88.25 ± 15.73 ^a	86.92 ± 8.28 ^a	178.04 ± 35.89 ^b
60	PI (18:0/18:1)	[M – H] [–]	863	42.62 ± 7.69 ^a	111.70 ± 9.50 ^b	139.72 ± 25.15 ^c
61	PI (18:0/18:0)	[M – H] [–]	865	15.87 ± 3.15 ^a	54.07 ± 4.56 ^b	65.71 ± 11.60 ^c
62	PI (18:1/20:4)	[M – H] [–]	883	48.38 ± 8.30 ^a	28.56 ± 3.09 ^b	46.99 ± 8.53 ^a
63	PI (18:0/20:4)	[M – H] [–]	885	183.29 ± 32.05 ^a	102.89 ± 9.41 ^b	62.21 ± 10.45 ^c
64	PI (18:0/20:3)	[M – H] [–]	887	188.29 ± 33.37 ^a	51.51 ± 4.67 ^b	33.92 ± 5.98 ^b
65	PI (18:0/20:2)	[M – H] [–]	889	39.43 ± 6.99 ^a	26.92 ± 2.38 ^b	33.52 ± 6.87 ^a

Supplementary Table S4. Changes in the relative levels of metabolites in fresh and spent media. The *p*-values were obtained by *t*-test. Each fresh and spent medium sample was taken after 2 days of melanoma cell culture. *, significant difference (*p* < 0.05); **, significant difference (*p* < 0.01); ***, significant difference (*p* < 0.001); ↑, increase in metabolites in spent medium compared to those in fresh medium; ↓, decrease in metabolites in spent medium compared to those in fresh medium. RT, retention time; TMS, trimethylsilylation.

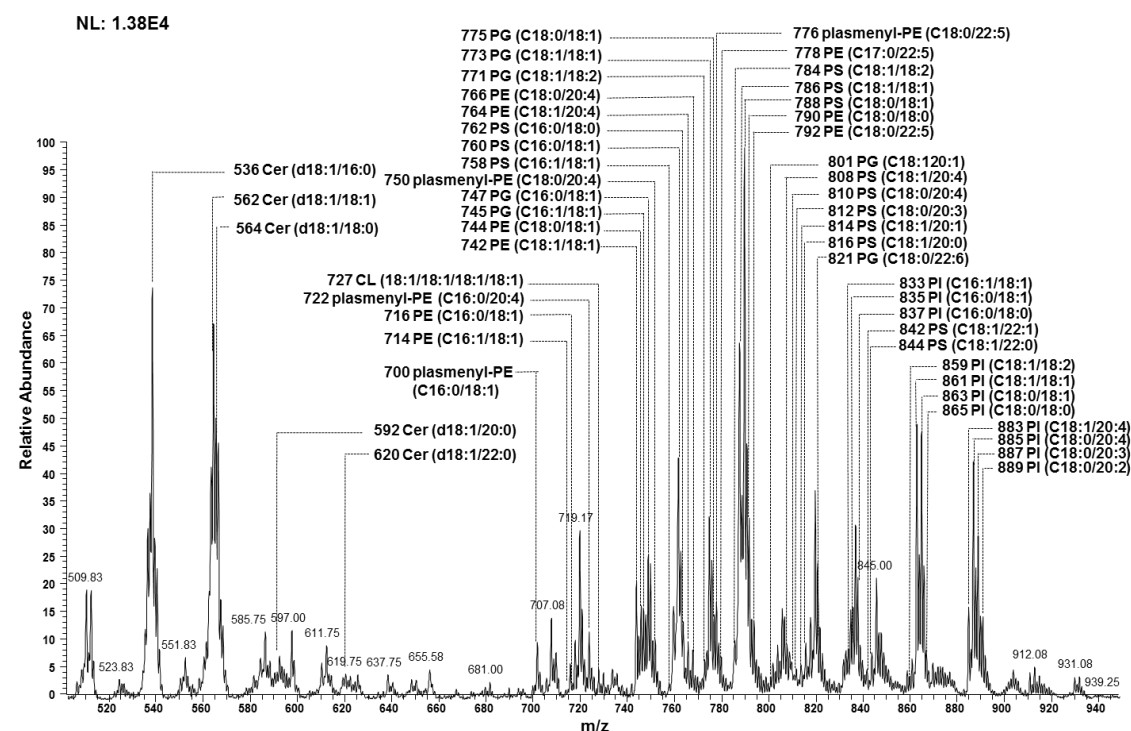
Compound	RT (min)	Mass fragment (m/z)	TMS	A375	A2058
Alcohol					
Myo-inositol***	36.33	191, 217 , 305, 318	6	23.27 ± 1.83 (↓)	7.47 ± 2.52 (↓)
Amino acid					
Alanine**	8.57	45, 116 , 190, 218	2	25.29 ± 3.77 (↑)	44.35 ± 16.60 (↑)
Glutamic acid***	24.57	218, 246 , 348, 363	3	22.96 ± 3.74 (↑)	37.14 ± 6.12 (↑)
Glutamine***	28.74	156 , 245, 347, 362	3	32.37 ± 0.91 (↓)	44.05 ± 4.32 (↓)
Isoleucine	14.53	45, 158 , 218, 260	2	35.78 ± 5.90 (↓)	38.19 ± 14.59 (↓)
Leucine	10.2	44, 75, 86 , 188	1	31.17 ± 5.70 (↓)	40.06 ± 23.95 (↓)
	13.84	158 , 218, 232, 260	2		
Ornithine***	29.8	142 , 174, 200, 258	4	1.30 ± 0.29 (↑)	2.55 ± 0.56 (↑)
Proline***	14.62	100, 133, 142 , 216	2	79.45 ± 15.91 (↑)	47.29 ± 15.86 (↑)
Pyroglutamic acid**	21.55	45, 156 , 230, 258	2	84.49 ± 18.62 (↑)	119.52 ± 27.79 (↑)
Serine**	16.71	100, 204 , 218, 306	3	29.06 ± 0.87 (↓)	23.52 ± 3.77 (↓)
Tyrosine***	32.86	218 , 280, 354, 382	3	21.74 ± 13.41 (↑)	52.56 ± 21.49 (↑)
Valine***	8.19	55, 72 , 146, 174	1	24.13 ± 4.70 (↑)	54.45 ± 13.90 (↑)
	12.02	45, 100, 144 , 218	2		
Organic acid					

Lactic acid ^{***}	7.41	117 , 133, 191, 219	2	2437.85 ± 207.04 (↑)	3074.88 ± 333.52 (↑)
Malic acid	20.72	133, 233 , 245, 335	3	0.26 ± 0.05 (↑)	0.30 ± 0.16 (↑)
Pyruvic acid ^{**}	7.11	89, 100, 115, 174	1(1MEOX)	18.59 ± 0.38 (↓)	11.51 ± 5.13 (↓)
Succinic acid	15.27	45, 129, 172, 247	2	0.35 ± 0.17 (↑)	0.44 ± 0.22 (↑)
Sugar					
Glucose ^{**}	34.01	129, 191, 204 , 217	5	3019.56 ± 333.00 (↓)	2206.99 ± 864.69 (↓)
	31.83	160, 205, 217, 319	5(1MEOX)		
Fatty acid					
Palmitic acid	35.54	117, 132, 313 , 328	1	1.72 ± 0.21 (↓)	1.98 ± 0.48 (↓)

A



B



Supplementary Figure S1. Representative spectra of lipid profiles from melanocytes and melanoma cell lines from nanoESI-MS in (A) positive ion mode and (B) negative ion mode

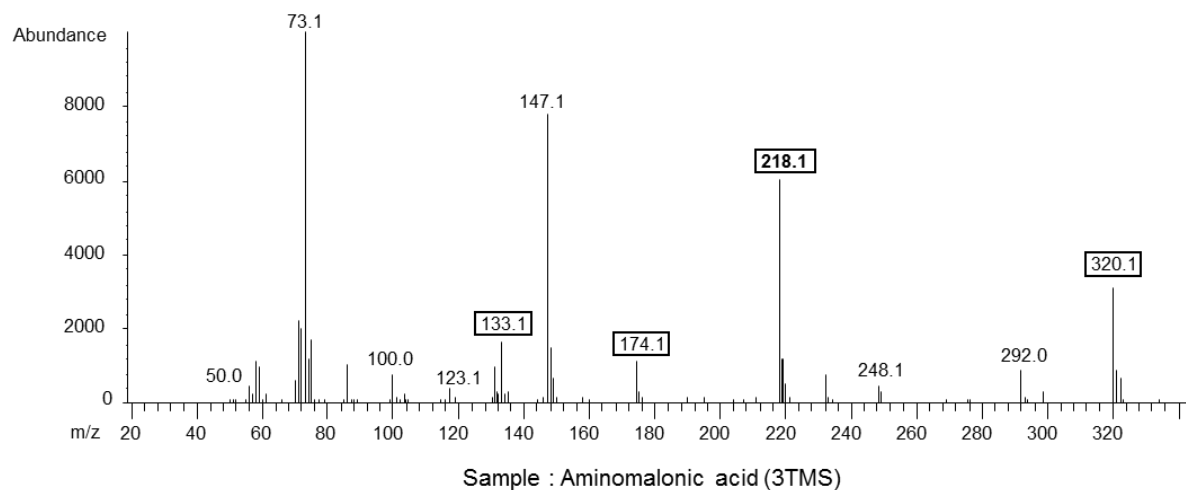
Compound #1

Aminomalonic acid

Retention time (min) : 16.16

Fragmentation ion (m/z) : 133, 174, 218, 320

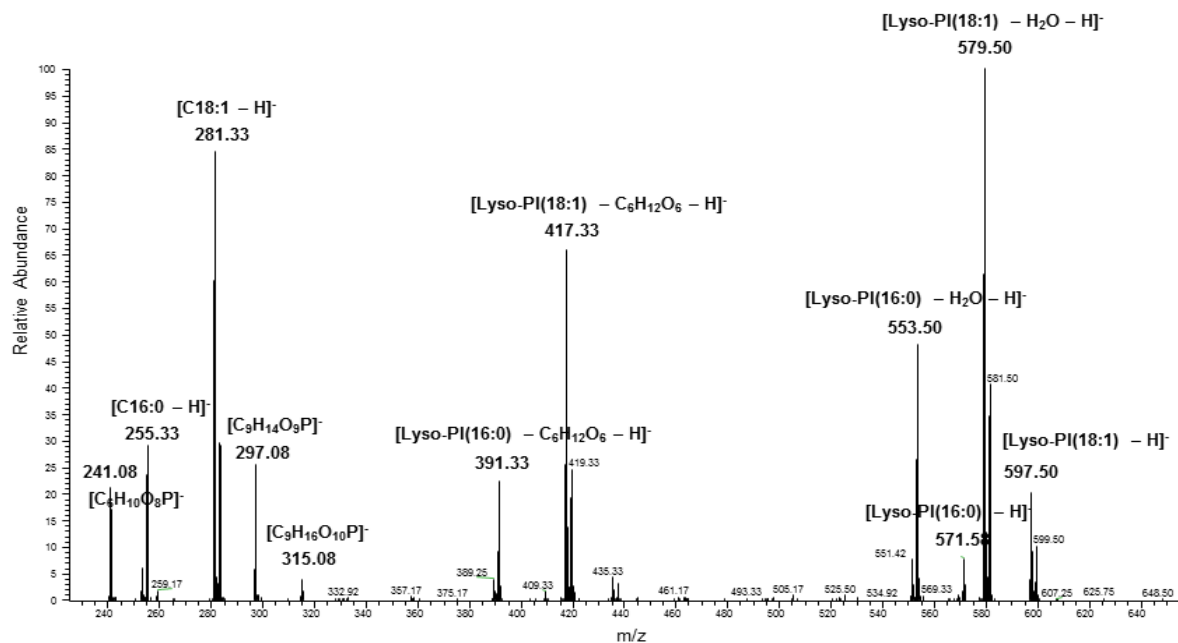
Formula : $C_3H_5NO_4$



Compound #2

[PI(16:0/18:1) - H]⁻, m/z 835

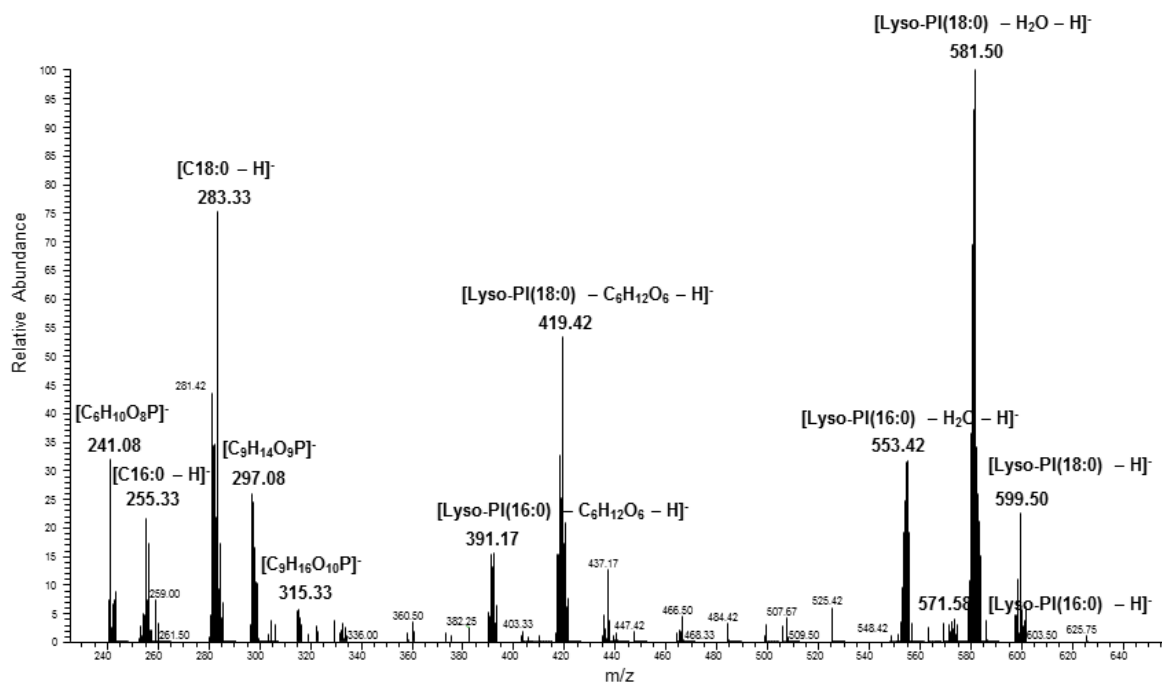
• $C_8H_{12}O_8$: inositol group



Compound #3

[PI(16:0/18:0) - H]⁻, *m/z* 837

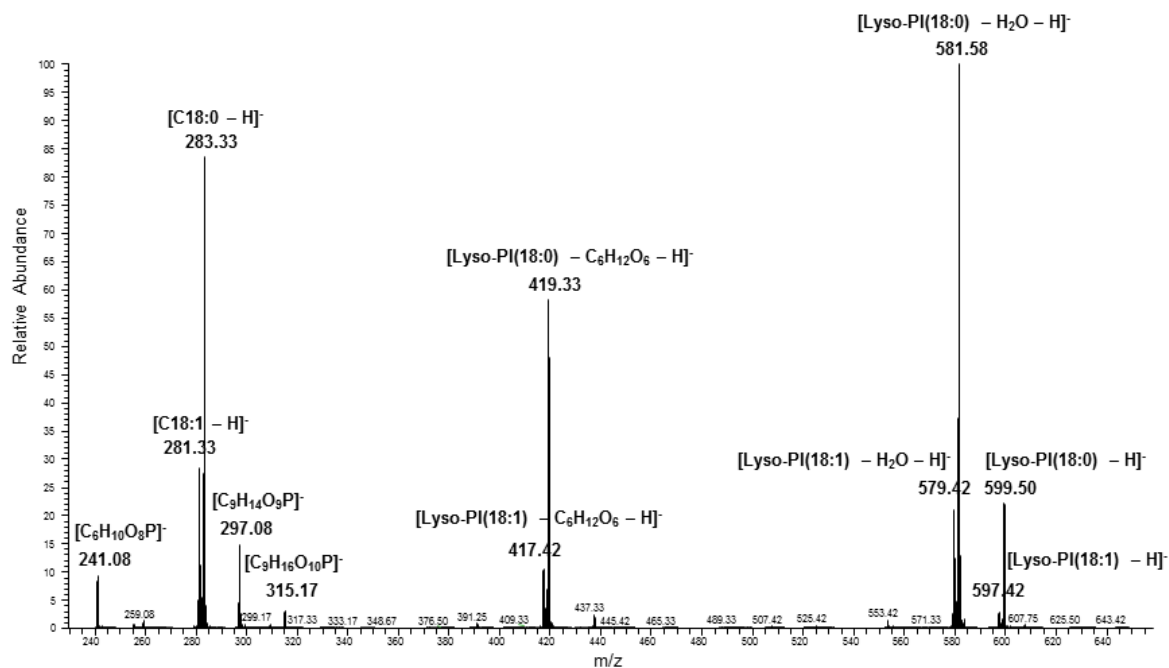
• C₆H₁₂O₆ : inositol group



Compound #4

[PI(18:0/18:1) - H]⁻, *m/z* 863

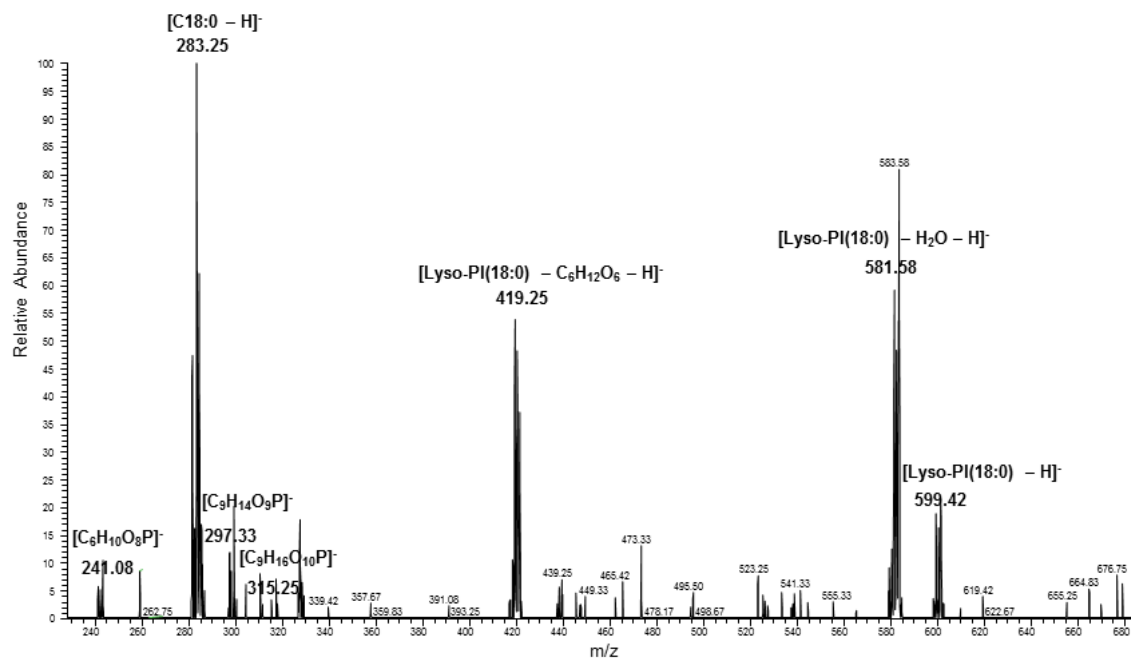
• C₆H₁₂O₆ : inositol group



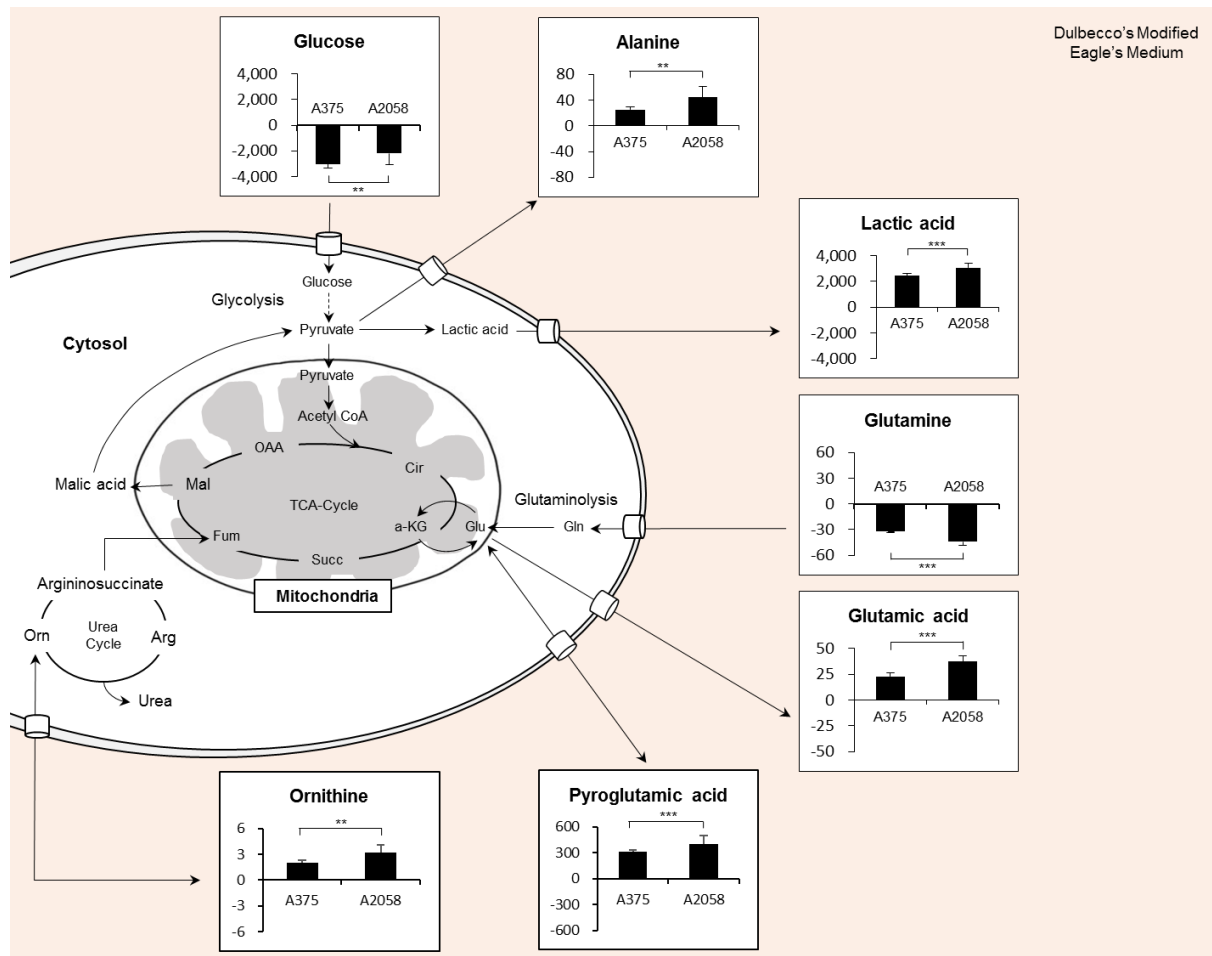
Compound #5

[PI(18:0/18:0) - H]⁻, m/z 865

• C₆H₁₂O₆ : inositol group



Supplementary Figure S2. Fragmentation mass spectra of the novel metabolite and lipid biomarkers of melanoma with different metastatic potential



Supplementary Figure S3. Changes in the relative levels of glucose, alanine, lactic acid, glutamine, glutamic acid, ornithine, and pyroglutamic acid in fresh and spent media. Samples were taken fresh and spent media after 2 days of melanoma cell culture.