Supplementary material

Discovering hair biomarkers of Alzheimer's disease using high resolution mass spectrometry-based untargeted metabolomics

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Figure S1. Quality control (QC) of the analytical method. (A) RSD distribution of all the detected peak features in the QC samples. Red line represents the 85% of all detected peaks. (B) RSD distribution in the QC sample of the discriminatory peak features between AD and controls. Red line represents 90% of the discriminatory peak features.

Figure S2. Correlations between 10 metabolites and MoCA score (A) acetyl-L-carnitine, (B) propionylcarnitine, (C) butyrylcarnitine, (D) O-valeroyl-L-carnitine, (E) 6-O-methylnorlaudanosoline, (F) PC (16:0/0:0), (G) LPC 18:1, (H) pyridoxal, (I) hydroxyprolyl-leucine, and (J) piperine**.**

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Peak No.	Retention time (min)	m/z	AD / Control	P
#01	0.81	134.0762	0.49	0.0091
#02	7.89	145.0647	12.46	0.0095
#03	0.84	149.5973	0.38	0.0050
#04	1.74	150.0549	0.28	0.0077
#05	1.25	198.037	0.47	0.0039
#06	4.68	199.0865	0.48	0.0010
#07	0.98	204.1228	0.40	0.0005
#08	4.43	213.1231	0.33	0.0084
#09	1.90	218.1384	0.35	0.0035
#10	1.08	218.1385	0.30	0.0028
#11	2.52	229.1178	0.31	0.0036
#12	4.47	232.1541	0.38	0.0019
#13	4.43	235.1072	0.30	0.0089
#14	9.63	240.1955	0.24	0.0058
#15	1.04	245.1493	0.30	0.0041
#16	5.12	246.1593	0.42	0.0077
#17	5.11	246.1696	0.43	0.0019
#18	1.57	247.1284	0.46	0.0093
#19	7.89	247.1437	16.98	0.0100
#20	5.11	248.1749	0.41	0.0020
#21	8.71	261.1458	0.34	0.0031
#22	4.07	267.0953	0.40	0.0080
#23	6.39	268.1151	6.75	0.0097
#24	8.44	272.1276	0.26	0.0010
#25	1.26	272.1963	0.44	0.0069
#26	4.50	274.1756	0.41	0.0094
#27	5.07	276.1589	7.19	0.0074
#28	2.32	281.1126	4.75	0.0051
#29	8.93	286.1431	0.35	0.0020
#30	9.06	286.1431	0.37	0.0020
#31	9.14	286.1431	0.34	0.0018
#32	5.71	288.1590	10.12	0.0021
#33	5.56	290.1745	179.00	0.0030
#34	5.56	292.1807	10.67	0.0075
#35	7.15	302.1379	0.17	0.0042

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#36	7.28	302.1379	0.21	0.0044
#37	7.28	303.1415	0.10	0.0059
#38	6.24	304.1538	6.47	0.0087
#39	9.77	312.1586	0.26	0.0093
#40	6.25	327.1368	6.48	0.0099
#41	10.90	329.2467	0.39	0.0082
#42	4.98	344.1233	4.86	0.0070
#43	6.68	353.2216	0.39	0.0000
#44	7.34	364.2262	18.06	0.0097
#45	6.69	365.1974	11.61	0.0046
#46	6.21	366.2056	77.20	0.0065
#47	11.21	366.2994	0.39	0.0010
#48	9.04	367.2374	0.45	0.0011
#49	6.61	378.2056	17.77	0.0054
#50	6.69	379.9793	87.88	0.0047
#51	6.42	380.2211	24.93	0.0074
#52	6.69	380.2211	58.60	0.0030
#53	5.69	382.2005	18.49	0.0065
#54	5.95	382.2005	17.46	0.0084
#55	6.69	382.2274	69.98	0.0032
#56	6.51	382.2368	89.53	0.0040
#57	9.14	385.2473	0.35	0.0018
#58	8.94	385.2474	0.32	0.0005
#59	6.13	396.2161	135.20	0.0051
#60	6.51	396.2162	123.54	0.0064
#61	6.13	398.2222	17.78	0.0082
#62	5.56	407.2531	43.01	0.0056
#63	6.01	412.2109	68.92	0.0029
#64	6.45	414.2266	94.70	0.0069
#65	6.18	424.2583	252.94	0.0024
#66	8.80	481.3044	0.34	0.0081
#67	10.66	496.3388	0.42	0.0013
#68	6.70	497.2996	10.55	0.0028
#69	10.66	497.3419	0.38	0.0056
#70	6.13	513.2948	664.05	0.0044
#71	10.92	522.3543	0.40	0.0046
#72	6.71	795.4117	270.16	0.0032

**Table S2. Chemical structures annotated by MS-FINDER and MoNA within MS-DIAL.**

Compound name	Formula	m/z	Retention time (min)	ID method	MS-DIAL match score	MS-FINDER Formula score	MS-FINDER structure score
L-Acetylcarnitine	C9H17NO4	204.1228	0.98	MoNA/MS-Finder	71.8	3.66	6.21
Hydroxyprolyl-Leucine	C11H20N2O4	245.1493	1.04	MS-Finder		3.60	
Pyridoxal	C8H9NO3	150.0549	1.74	MoNA	79.3		
Propionylcarnitine	C8H17NO4	218.1384	1.90	MS-Finder		3.93	6.7
Butyryl-carnitine	C11H21NO4	232.1541	4.47	MoNA/MS-Finder	95.6	3.55	5.91
Valeryl-carnitine	C12H23NO4	246.1696	5.11	MoNA/MS-Finder	90.8	3.55	5.99
Hyoscyamine	C17H23NO3	290.1745	5.56	MoNA/MS-Finder	81.8	3.61	6.98
Cyrmenin A	C22H34N2O5	407.2531	5.56	MS-Finder		3.42	
Galantamine	C17H21NO3	288.1590	5.71	MS-Finder		3.54	
Sacubitril	C24H29NO5	412.2109	6.01	MS-Finder		3.85	
	C24H29NO4	396.2161	6.13	MS-Finder		3.70	
	C25H33N3O3	424.2583	6.18	MS-Finder		3.02	
6-O-Desmethyldonepezil	C23H27NO3	366.2056	6.21	MS-Finder		4.05	7.18
Scopolamine	C17H21NO4	304.1538	6.24	MS-Finder		3.74	7.19
	C24H31NO5	414.2266	6.45	MS-Finder		3.77	
2-benzoyl-1-(2,5-dimethoxyphenyl)-decahydroisoquinolin-4a-ol	C24H29NO4	396.2162	6.51	MS-Finder		3.69	
	C24H27NO3	378.2056	6.61	MS-Finder		3.58	
	C21H28NO3	365.1974	6.69	MS-Finder		3.23	

Donepezil	C24H29NO3	380.2211	6.69	MoNA/MS-Finder	92.1	3.83	7.25
6-O-methylnorlaudanosoline	C17H19NO4	302.1379	7.15	MS-Finder		3.78	
	C24H29NO2	364.2262	7.34	MS-Finder		3.26	
Lenticin	C14H18N2O2	247.1437	7.89	MoNA/MS-Finder	95.8	3.36	5.71
Piperine	C17H19NO3	286.1431	9.14	MoNA/MS-Finder	94.8	3.69	6.59
PC(16:0/0:0)	C24H50NO7P	496.3388	10.66	MoNA	88.6		
LPC 18:1	C26H52NO7P	522.3543	10.92	MoNA	75.2		
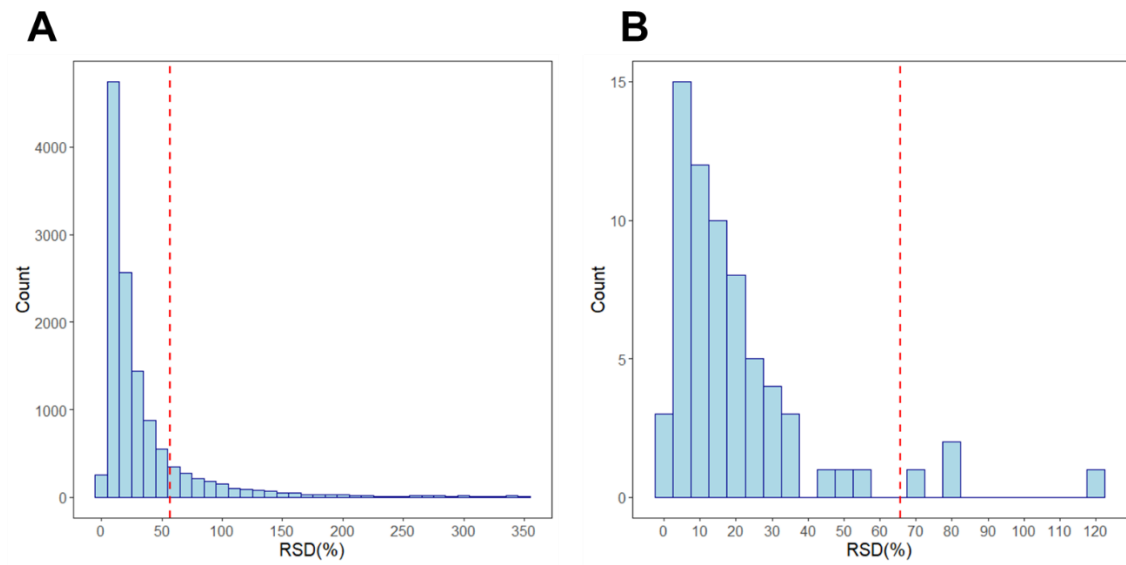
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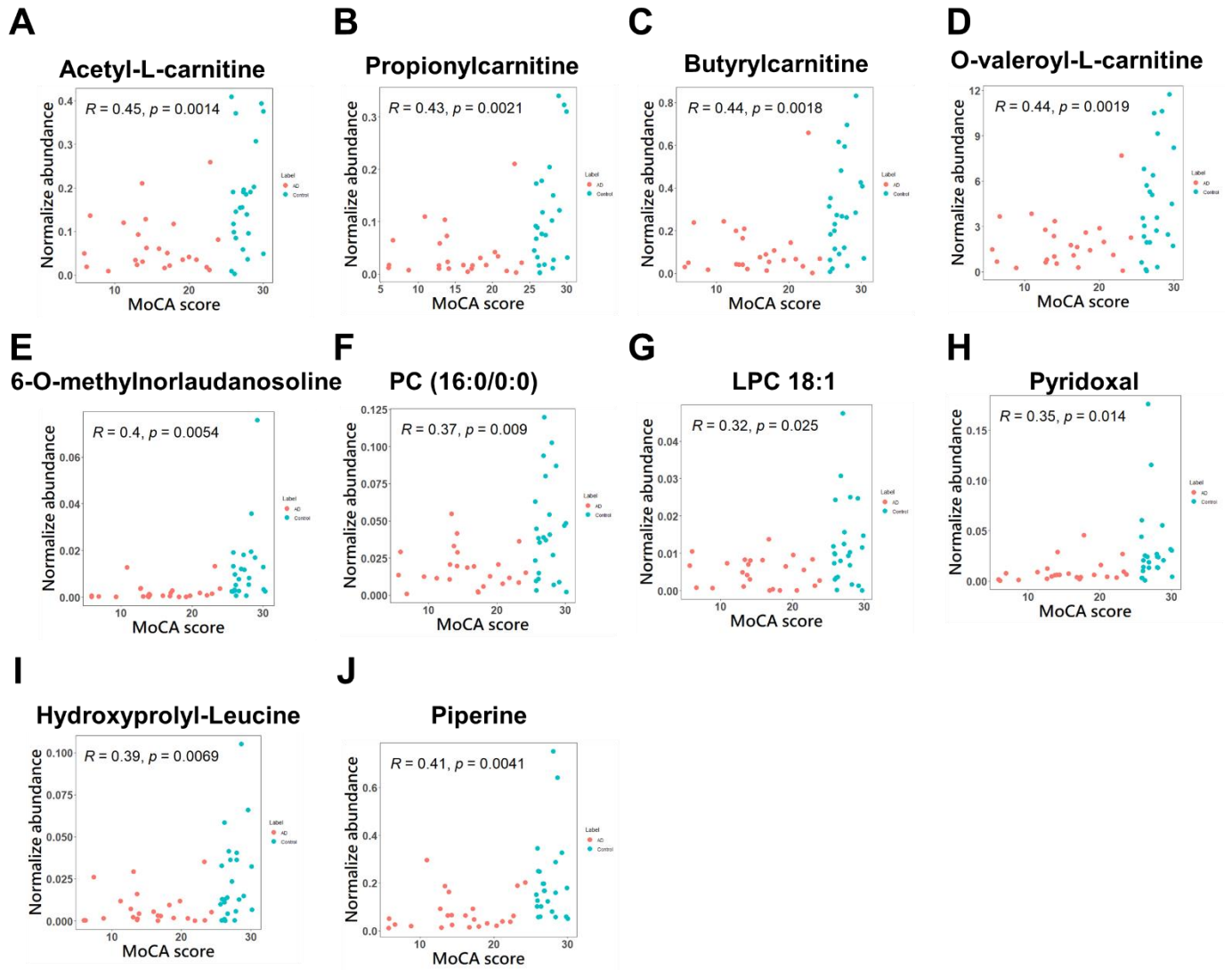
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