Investigating the Mechanism of Qifu Yin in Ameliorating Memory Disorders through Pseudo-Targeted Lipidomic

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1 Materials

1.1 Herb

Panax ginseng (Lot: 210401, Jilin Baishan), Angelica sinensis (Lot: 211217002,Gansu), Rehmanniae Radix Praeparata (Lot: 210601, Henan Jiaozuo), Atractylodes macrocephala (Lot: 2020120411, Anhui Bozhou), Ziziphus jujuba (Lot: 210301, Hebei Xingtai), Polygalae Radix (Lot: 210427, Shanxi), Glycyrrhiza uralensis (Lot: 211102, Inner Mongolia Alxa Left Banner) were purchased from Beijing Tongrentang Pharmacy.

1.2 Reagent

Methanol (Lot: 216678) was purchased from Thermo Fisher Scientific. Acetonitrile (Lot: 224288) was purchased from Minobell Company. Formic acid (batch number: 3208K240) was purchased from MACKLIN Company. Ferulic acid (Lot: DSTDF008102), glycyrrhizic acid (Lot: DSTDG000603), maackiain (Lot: DST221015), adenine (Lot: DSTDP001301), liquiritin (Lot: DSTDG000902), jujuboside A (Lot: DST221102), polygalaxanthone III (Lot: DST230425), tenuifolin (Lot: DSTDX003402), ginsenoside Re (Lot: DSTDR001404), ginsenoside Rg1 (Lot: DSTDR000902), atractylon (Lot: DSTDC010703), and rehmannioside D (Lot: DSTDD230102) were purchased from Dexter Biological Co., Ltd.

1.3 Apparatus

SCIEX X500R QTOF quadrupole time-of-flight mass spectrometer was purchased from AB SCIEX (Foster City, CA, USA), Shimadzu ultra-high performance liquid phase system was purchased from Shimadzu Corporation (Kyoto, Japan). An ACQUITY UPLC BEH C18 (100 × 2.1 mm, 1.7 m) column was purchased from the Waters Corporation (Milford, MA, USA). HC-

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2518 high-speed centrifuge was bought from Anhui Zhongke Zhongjia Scientific Instrument Co., Ltd (Anhui, China). A WB-12 Termovap Sample Concentrator was purchased from Shenzhen Widen Electronics Equipment Co., Ltd (Shenzhen, China).

2 Methods

2.1QFY component analysis

2.1.1 Solution-process

Qifu Yin (Panax ginseng 6 g, Rehmanniae Radix Praeparata 9 g, Angelica sinensis 9 g, Atractylodes macrocephala 5 g, Glycyrrhiza uralensis 3 g, Ziziphus jujuba 6 g, Polygalae Radix 5 g) was weighed according to the proportion of seven herbs, and 10 times the amount of distilled water was added to ultrasonic 1 h, filtration, concentration into dry extract, mass spectrometry analysis with mass spectrometry grade methanol re-dissolution, 0.45 μm microporous membrane filtration for LC-MS analysis.

About 5 mg of each reference substance was accurately weighed and placed in a 5 mL volumetric flask, and the mass concentration of 1 mg mL^{-1} of single reference substance reserve solution was prepared by adding mass grade methanol, filtered by 0.22 μm microporous membrane, and set aside.

2.1.2 Chromatographyconditions

An Acquity UPLC BEH C18 (100×2.1mm, 1.7 μm) column was selected as the chromatographic column in this experiment. The injection volume was 5μL, and the column temperature was 40℃. A flow rate of 0.3 mL/min was used. The mobile phases were0.1% formic acid–water (A) and acetonitrile (B). The elution conditions were as follows:0-5 min，98%-90%A，5-12 min，90%-80%A；12-18 min，80%-70%A；18-23 min，70%-59%A；23-27 min，59%-52%；27-30 min，52%-39%A；30-40 min，39%-2%A，40-41 min，2%-98%A,，41-42 min，98%-98%A.

2.1.3 Mass spectrometry conditions

The data of the first and second spectra of the samples were collected by electrospray ionization source in positive ion mode and negative ion mode. The parameters of positive and negative ESI modes were set as follows : spray voltage 5500, -4500 V ; declustering potential (DP) : 60, -60 V ; ion source temperature 500 °C; the collision energies are 30, -30 eV, respectively. GAS was set as follows : atomizing gas (GAS1), 50 psi; assisted heating gas (GAS2), 55 psi. The

automatic calibration device system corrects every 5 samples to maintain quality accuracy.

2.1.4 Data processing

UPLC-Q-TOF-MS was used to analyze the samples of Qifuyin. According to the multi-level mass spectrometry information of the samples, combined with the high-resolution mass spectrometry database of natural products and related literature, the main compounds in Qifuyin were identified.

The data acquisition software was SCIEX OS, and data processing was performed using Analytics. During the identification, the mass spectrometry data were preferentially matched with the Natural Products HR-MS/MS Spectal Library 1.0 database. The compounds were preliminarily screened according to the score information of each chromatographic peak, and the compounds were further confirmed according to the primary and secondary information of each chromatographic peak. At the same time, according to literature reports, etc.for identification.

2.2 Network Pharmacology Analysis

2.2.1Obtaining the targets of active ingredients and disease targets

The Canonical SMILES structure of the above QFY components was searched by PubChem ID and imported into the Swiss Target Prediction database to obtain the component targets. The Gene Cards database library was used to search for disease-related targets with “Memory disorder” as the keyword.

2.2.2 Protein-protein interaction (PPI) network and drug-component-target-disease network were constructed.

Venny 2.1 was used to intersect the two targets. The intersection targets were uploaded to the String database, and the information on protein-protein interactions was obtained and imported into Cytoscape 3.9.2 software to draw the network diagram (PPI) of protein-protein interactions. Cytoscape 3.9.2 software was used to draw the drug-component-disease-target network.

2.2.3 KEGG pathway enrichment analysis

The intersection targets screened under “1.2” were imported into the DAVID database for KEGG pathway enrichment analysis, and the results were visualized using the micro-bioinformatics online data analysis platform.

3 Results

3.1QFY component analysis results

Fig. S1-2 shows the total ion chromatograms of water QFY extract in positive and negative ion modes. According to the multi-stage mass spectrometry information of the samples in the spectrum, combined with the high-resolution mass spectrometry database of natural products and related literature, 167 compounds were identified from the samples of QFY, as shown in Table S1.

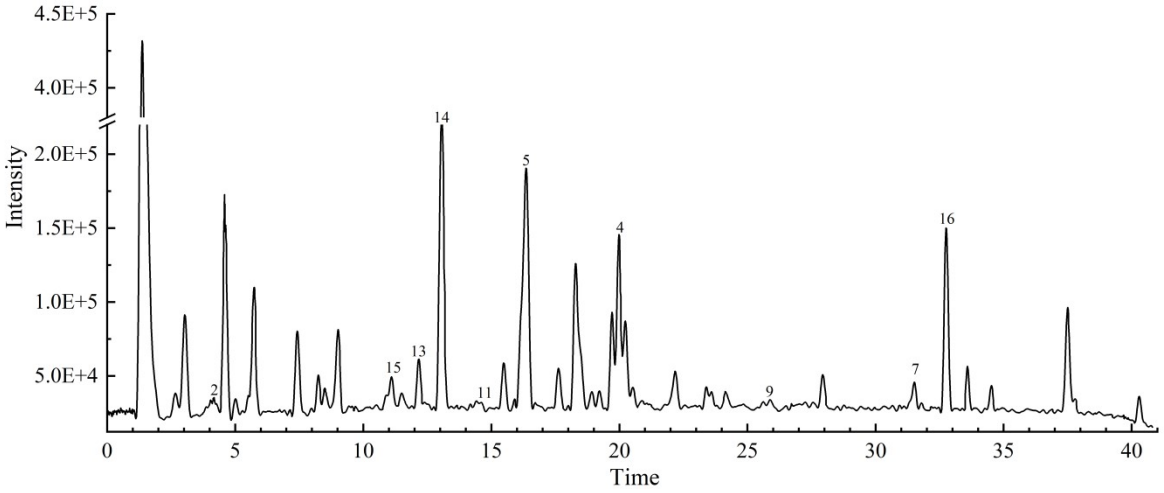


Fig. S1 Positive ion current diagram

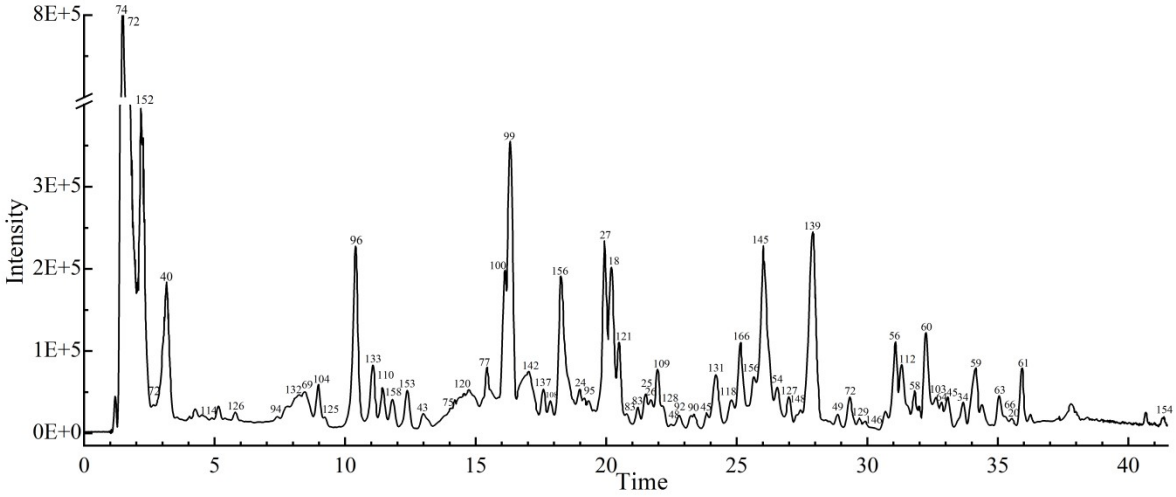


Fig. S2 Negative ion current diagram

Table S1 Analysis of chemical constituents of QFY in positive and negative ion modes

NO.	t_R	Name	Formula	Mode	Adduct	Measured value (M/Z)	Error（PPM）	Fragment ion	Source
1	19.52	Tenuifolin*	C36H56O12	+	[M+H]+	681.3488	-2.20	663.3963,484.3226,324.2365	Polygalae Radix
				-	[M-H]-	679.3989	-3.82	425.3056,179.0564,119.0342,89.0243,71.0139	
2	4.19	adenine*	C5H5N5	+	[M+H]+	136.0607	1.48	119.0352,94.0401,92.0245,82.0402,77.0135,67.02926,5.0135,55.0191,53.0137	Panax ginseng
3	28.66	Maackiain*	C16H12O5	+	[M+H]+	285.2897	1.41	123.0440,102.0472,79.0176,71.0133	Glycyrrhiza uralensis
				-	[M-H]-	283.1664	-9.50	183.0426,163.1126,107.0500	
4	20.16	ginsenoside Rg1*	C42H72O14	+	[M+Na]+	823.4821	0.62	644.4181，643.4183，203.0544	Panax ginseng
				-	[M-H]-	799.4811	3.37	179.0556,161.0452,101.0242,89.0242,71.0139,51.0135	
5	16.30	ferulic acid*	C10H10O4	+	[M+H]+	195.0555	2.06	163.0396,149.0600,145.0287,134.0366,117.0337,89.0388	Angelica sinensis
				-	[M-H]-	193.0512	6.18	178.0264,149.0604,137.0240,134.0370,133.0287,132.0214,106.0422	Angelica sinensis、Ziziphus jujuba
6	25.30	jujuboside A*	C58H94O26	+	[M+H]+	1207.5971	7.79	929.5437,865.4638,767.4057,749.4566,605.4505,587.4235,163.0626,145.0513	Ziziphus jujuba
7	32.48	atractylone*	C15H20O	+	[M+H]+	217.1927	-1.39	155.0857,129.0696,91.0544,83.0495,72.9374	Atractylodes macrocephala
					[2M-H]-	431.1809	6.94	249.9951，96.9697	
8	21.06	Ginsenoside Re*	C48H82O18	+	[M+H+Na]+	971.47937	-8.27	791.4637,421.3471,189.1648,147.0641	Panax ginseng
					[M-H]-	946.3827	-3.06	161.0456,119.0347,89.0244,59.1037,179.0561	
9	26.92	glycyrrhizic acid*	C42H62O16	+	[M+H]+	823.1275	6.69	647.3814,471.3753,453.3378,435.3273	Glycyrrhiza uralensis
					[M-H]-	821.3897	-2.92	759.4066,645.3639,627.3647,351,0570,193.0365	
10	15.72	liquiritin*	C21H22O9	+	[M+H]+	419.1614	-1.20	341.1011,257.0799,239.0703,211.0758,163.0390,147.0440,137.0232,	Glycyrrhiza uralensis
				-	[M-H]-	417.117	-4.30	255.0652,148.0166,135.0083,119.0498,91.0188	

11	14.71	Polygalaxanthone III*	C25H28O15	+	[M+H]+	569.1510	6.69	371.0788，317.0664，287.0548，313.0723，287.0548，275.0559	Polygalae Radix
					[M-H]-	567.1331	-4.58	345.0634,315.0517,272.0328	
12	6.117	Rehmannioside D*	C27H42O20	+	[M+H+NH4]+	704.2436	6.99	471.1501,327.1072,325.1160,165.0551,145.0497	Rehmanniae Radix Praeparata
				-	[M-H]-	685.216	-4.08	505.1566,323.0981,263.0765,221.0663,179.0557	
13	12.02	Coclaurine	C17H19NO3	+	[M+H]+	286.1453	4.91	237.0925，209.0952，175.0799，107.0498	Ziziphus jujuba
14	13.17	zizyphusine	C20H24NO4+	+	M+	342.1446	7.31	297.1122，282.0892，265.0861，237.0914，222.0681，219.0809，207.0808，165.0701	Ziziphus jujuba
15	11.15	magnocurarine	C19H24NO3+	+	M+	314.1756	3.82	269.1170，237.0919，178.0823，175.0749，151.0768	Ziziphus jujuba
16	1.55	(Z)-Ligustilide	C12H14O2	+	[M+H]+	191.0604	5.79	173.0444，117.5835，91.2614	Angelica sinensis
17	25.02	Ginsenoside Rb1	C54H92O23	-	[M-H]-	1107.5904	0.90	945.5367，783.4913，621.4368	Panax ginseng
18	20.20	Ginsenoside Rd	C48H82O18	-	[M-H]-	945.5369	-3.06	783.4859，765.4779，603.4207	Panax ginseng
19	32.64	Ginsenoside Rg3	C42H72O13	-	[M-H]-	783.5156	-1.53	621.4384，603.8867，459.3834，375.2944	Panax ginseng
20	35.59	Ginsenoside Rg5	C42H70O12	-	[M-H]-	765.5075	2.48	603.4221	Panax ginseng
21	18.84	Ginsenoside Re1	C48H82O19	-	[M+HCOO]-	1007.5369	-0.73	961.5333，799.4801	Panax ginseng
22	19.22	notopanaxoside-A	C36H62O10	-	[M+HCOO]-	699.425	-6.27	653.4261，491.3708，161.0451	Panax ginseng
23	20.97	6′-O-acetyl-Rg1	C44H74O15	-	[M+HCOO]-	887.49	-9.61	841.4927，781.4733，475.3733	Panax ginseng
24	18.84	Ginsenoside Re3	C48H82O19	-	[M+HCOO]-	1001.5369	-0.73	961.5333，799.4801，637.4316	Panax ginseng
25	21.55	6′-O-acetyl-Re	C50H84O19	-	[M+HCOO]-	1033.5498	6.58	987.5471，945.5455，927.5254，637.4277	Panax ginseng
26	21.71	yesanchinoside D	C44H74O15	-	[M+HCOO]-	887.497	-0.24	841.4954，781.4746，437.2831	Panax ginseng
27	20.04	Ginsenoside Rg7	C42H72O14	-	[M+HCOO]-	845.4796	4.87	799.4822，637.4299，475.3786	Panax ginseng
28	25.38	quinquenoside-R1	C56H94O24	-	[M+HCOO]-	1195.5958	-7.30	1149.6007，1107.5904，945.5340	Panax ginseng
29	20.28	gypenoside-XVII	C48H82O18	-	[M+HCOO]-	991.5437	0.53	945.5356，783.4858，621.0289	Panax ginseng
30	20.98	Ginsenoside Rg8	C42H70O13	-	[M+HCOO]-	827.4687	4.98	781.4781，619.4710	Panax ginseng

31	32.45	Isomer of Rg2	C42H72O13	-	[M+HCOO]-	829.5309	-0.76	783.4837，621.4349,475.2509	Panax ginseng
32	29.49	gypenoside-IX	C47H80O17	-	[M+HCOO]-	961.5459	-0.33	915.5283,783.4798,621.4396	Panax ginseng
33	20.98	Ginsenoside Rg9	C42H70O13	-	[M+HCOO]-	827.4598	-6.39	781.4781,619.4170	Panax ginseng
34	33.75	(I-1)-GlurA	C36H56O9	-	[M-H]-	631.3825	1.11	555.3727,455.3842	Panax ginseng
35	33.09	Ginsenoside Rs3	C44H74O14		[M+HCOO]-	871.5043	6.90	825.4936,783.4837,621.4330,459.3802	Panax ginseng
36	18.25	Ginsenoside Re3	C48H82O19		[M+HCOO]-	1007.5389	0.21	799.4820,637.4290,475.3787	Panax ginseng
37	25.31	20(S)Rg2	C42H72O13	-	[M+HCOO]-	829.4905	-7.52	637.4309,475.3782	Panax ginseng
38	28.68	Gypenoside XVII	C48H82O18	-	[M+HCOO]-	991.5437	-0.11	945.5383,783.4736,621.4387	Panax ginseng
39	8.81	N-glc-indoleaceticacid	C16H19NO8	-	[M-H] -	352.1039	9.40	188.0724,146.0602	Ziziphus jujuba
40	2.91	citric acid	C6H8O7	-	[M-H]-	191.0183	-6.84	111.0088,87.0088,85.0296	Ziziphus jujuba、Rehmanniae Radix Praeparata、Angelica sinensis
41	5.75	Guanosine	C10H13N5O5	-	[M-H]-	282.1033	0.36	1,500,416,133.02	Ziziphus jujuba
42	17.06	glutamic acid dipeptide	C9H18N25	-	[M-H]-	475.2131	-2.32	96.9694	Ziziphus jujuba
43	12.90	Vicenin-2	C27H30O15	-	[M-H -	593.1481	-6.59	575.1389,473.1067,455.0955,383.0759,353.0648	Ziziphus jujuba
44	15.48	Isospinosin	C28H32O15	-	[M-H]-	607.1440	4.95	487.1210,445.1298,427.1018,325.0710,324.0618,307.0618	Ziziphus jujuba
45	23.73	Licoricesaponin A3	C48H72O21	-	[M-2H]2-	983.4438	-2.65	863.5027,821.3955,803.3678,351.0545	Glycyrrhiza uralensis
46	25.02	Licoricesaponin G2	C42H62O17	-	[M-H]-	837.3853	-0.12	775.3862,661.3568,643.3409,485.3231,352.0573,351.0554,333.0402	Glycyrrhiza uralensis
47	26.91	yunganoside L1	C48H72O20	-	[M-H]-	967.4789	-1.35	949.4688,905.4374,498.1136,497.1125,339.0915,321.0788	Glycyrrhiza uralensis
48	22.54	yunganosideK2	C42H62O17	-	[M-H]-	837.3864	-1.55	643.3496,351.0558,289.0610	Glycyrrhiza uralensis
49	28.83	Licoricesaponin	C42H64015	-	[M-H]-	807.4115	-3.72	745.4060,352.0600,351.0542	Glycyrrhiza uralensis
50	27.75	licorice saponin H2	C42H62016	-	[M-H]-	821.3925	-1.71	645.3612,627.3595,351.0560	Glycyrrhiza uralensis
51	29.45	licorice sapo_x005f nin J2	C42H64016	-	[M-H]-	823.3597	-9.85	805.3798,762.2530,647.3687,351.0543,289.0541	Glycyrrhiza uralensis

52	18.71	5-hydroxylliquiritin	C21H22O10	-	[M-H]-	433.1439	3.47	271.0603,159.7969,119.0499	Glycyrrhiza uralensis
53	26.61	22-β-acetyl-liquiritigenin	C44H64O18	-	[M-H]-	879.3893	3.53	351.0547	Glycyrrhiza uralensis
54	26.56	Licoricesaponin E2	C42H60O16	-	[M-H]-	819.3457	6.72	351.0565,289.0564,193.0495	Glycyrrhiza uralensis
55	27.00	22-β-acetyl-glycyrrhizin C2	C44H64O16	-	[M-H]-	863.3619	-6.15	351.4019,193.0340	Glycyrrhiza uralensis
56	31.00	Glycycoumarin	C21H20O6	-	[M-H]-	367.0759	-3.82	309.0384,297.0389,256.6642	Glycyrrhiza uralensis
57	28.18	Uralsaponin B	C42H62O6	-	[M-H]-	821.3325	-0.37	351.0559,193.0352	Glycyrrhiza uralensis
58	31.80	Isolicoflavonol	C20H18O6	-	[M-H]-	353.0705	5.96	297.0387,135.0081	Glycyrrhiza uralensis
59	33.84	licobenzofuran	C21H22O5	-	[M-H]-	353.2059	-7.67	309.0705,283.9005,267.0680	Glycyrrhiza uralensis
60	32.24	Gancaonin C	C20H1805	-	[M-H]-	353.1002	-0.85	323.0922,267.1011,216.0418	Glycyrrhiza uralensis
61	35.96	Gancaonin 1	C23H22O5	-	[M-H]-	353.1381	-1.99	338.1143,267.1012,243.3070	Glycyrrhiza uralensis
62	32.97	Neoglycyrol	C33H18O16	-	[M-H]-	365.1017	4.12	307.0274,295.0235,254.0201	Glycyrrhiza uralensis
63	34.91	Glabrone	C38H16O5	-	[M-H]-	335.0910	2.10	320.0295,291.0642,135.0076	Glycyrrhiza uralensis
64	32.83	Licochalcone A	C33H22O4	-	[M-H]-	337.1062	-2.68	293.0435,281.0432,268.0328	Glycyrrhiza uralensis
65	35.89	Licorisoflavan I	C26H52O5	-	[M-H]-	423.1787	-0.24	229.0860,193.0858,174.0317	Glycyrrhiza uralensis
66	35.25	Licoricidin	C26H32O5	-	[M-H]-	423.2157	5.68	233.1165,193.0861,174.0327	Glycyrrhiza uralensis
67	14.38	Catalpol	C15H22O10	-	[M-H]-	361.1461	-7.77	199.0965,169.9769	Rehmanniae Radix Praeparata
68	0.88	Leonuride	C15H24O9	-	[M-H]-	347.7737	-3.75	329.7712,167.8910	Rehmanniae Radix Praeparata
69	8.34	8-Epi-Loganic acid	C16H24O10	-	[M-H]-	375.1300	2.14	213.0765,169.0863	Rehmanniae Radix Praeparata
70	13.37	Echinacoside	C35H46O20	-	[M-H]-	783.2491	2.56	623.2170,161.0238	Rehmanniae Radix Praeparata
71	20.67	Isomartynoside	C31H40O15	-	[M-H]-	651.2267	-0.31	475.1828,193.0497	Rehmanniae Radix

									Praeparata
72	2.58	N-(1-deoxy-D-fructos1-yl)pyroglutamic acid	C11H17NO8	-	[M-H]-	290.0893	2.77	272.0763,200.0558,128.0352	Angelica sinensis
73	9.90	coniferyl ferulate	C20H20O6	-	[M-H]-	355.1023	3.11	178.0250,149.0612	Angelica sinensis
74	1.53	2-Hydroxy-succinic acid	C4H6O5	-	[M-H]-	133.0146	3.03	115.0034	Atractylodes macrocephala
75	14.08	Lancerin	C19H18O10	-	[M-H]-	405.0796	-0.99	315.0498,307.1383,285.0394,257.0444	Polygalae Radix
76	14.42	Sibiricaxanthone A	C24H26O14	-	[M-H]-	537.1214	-1.87	417.0828,387.0742,315.0805,285.0394,267.0293	Polygalae Radix
77	15.41	7-O-Methylmangiferin	C20H20O11	-	[M-H]-	435.0920	1.84	345.0609,315.0496,272.0317,257.0065	Polygalae Radix
78	18.34	arillanin A	C33H40O18	-	[M-H]-	723.2613	1.38	547.1661,499.1441,265.0700,223.0605,205.-501	Polygalae Radix
79	19.20	Tenuifoliose G	C66H84O38	-	[M-H]-	1483.4256	-6.81	1337.3912,1295.3824,1161.3490,753.2041	Polygalae Radix
80	22.03	3,4,5-Trimethoxycinnamic acid	C12H14O5	-	[M-H]-	237.0758	-0.85	193.0874,133.0648,103.0551	Polygalae Radix
81	20.61	Tenuifoliose M	C65H82O37	-	[M-H]-	1453.4325	-5.58	1307.3794,1161.3543,1119.3408	Polygalae Radix
82	20.71	Tenuifoliose L	C67H84O38	-	[M-H]-	1495.4561	0.87	1349.3934,1307.3929,1227.3302,1203.3590,1143.3297,1081.3357,795.2122	Polygalae Radix
83	20.79	Tenuifoliside C	C35H44O19	-	[M-H]-	767.2391	-5.22	529.1523,367.1022	Polygalae Radix
84	20.63	Tenuifoliose T	C56H70O32	-	[M-H]-	1253.3710	9.82	1131.3733,1077.3246,955.2908	Polygalae Radix
85	21.87	Tenuifoliose I	C59H72O33	-	[M-H]-	1307.3815	-1.15	1265.3992,1247.3474,1161.3450,1119.3399,1101.3227,1039.3056	Polygalae Radix
86	21.33	Tenuifoliose C	C58H72O33	-	[M-H]-	1295.3819	-2.63	1173.4036,1119.3300,1101.3194	Polygalae Radix
87	23.42	Tenuifoliose O	C61H76O35	-	[M-H]-	1367.3522	-9.08	1325.4109,1191.3529,1173.3236,1149.3518	Polygalae Radix
88	21.87	Tenuifoliose J	C59H72O33	-	[M-H]-	1307.3815	-1.15	1265.3992,1247.3404,1161.3450,1143.3335,1101.3227,1039.3956	Polygalae Radix
89	22.71	Polygalasaponin	C53H84O24	-	[M-H]-	1103.5177	-1.72	1073.5119,455.3147,425.3039	Polygalae Radix

		XXVIII							
90	23.23	Tenuifoliose H	C61H74O34	-	[M-H]-	1349.3897	-0.07	1203.3555,1161.3470,1081.3292	Polygalae Radix
91	22.96	Onjisaponin TG	C64H100O32	-	[M-H]-	1379.6455	-8.20	1235.5659,455.3133	Polygalae Radix
92	22.83	Onjisaponin TF	C59H94O28	-	[M-H]-	1249.5783	-5.21	1025.5287,455.3131,425.3041	Polygalae Radix
93	23.68	Tenuifoliose N	C63H78O36	-	[M-H]-	1409.4158	1.07	1233.3633,1191.3619,1111.3222,1069.3046	Polygalae Radix
94	7.04	Sibiricose A3	C19H26O13	-	[M-H]-	461.1281	3.04	179.0349,137.0239,93.0343	Polygalae Radix
95	19.14	Arillanin C	C22H30O14	-	[M-H]-	517.1170	-0.78	175.0395,161.0472,160.0164,134.0371	Polygalae Radix
96	10.23	Sibiricose A1	C23H32O15	-	[M-H]-	547.1319	-8.24	223.0602,205.0498,190.0261,149.0232	Polygalae Radix
97	14.62	Sibiricaxanthone B	C24H26O14	-	[M-H]-	537.1209	-2.24	285.0394,267.0283	Polygalae Radix
98	14.32	Polygalaxanthone Ⅲ	C25H28O15	-	[M-H]-	567.1343	4.24	345.0603,315.0507,272.0311,259.0237	Polygalae Radix
99	16.29	Tenuifoliside B	C30H36O17	-	[M-H]-	667.2029	-1.80	281.0660,239.0547,205.0495	Polygalae Radix
100	16.07	telephiose E	C34H42O19	-	[M-H]-	753.2206	5.98	409.1267,205.0495,290.0261	Polygalae Radix
101	18.25	Sibiricose A4	C34H42O19	-	[M-H]-	753.2198	-3.06	223.0603,165.0531	Polygalae Radix
102	32.44	20(R)-Ginsenoside Rg3	C41H68O14	-	[M+HCOO]-	829.4602	1.53	783.4833,621.4340,161.0461101.0238	Panax ginseng
103	32.59	20(S)-Ginsenoside Rg3	C41H68O14	-	[M-H]-	783.4555	1.92	783.4897,621.4367,459.3834,161.0449	Panax ginseng
104	8.98	Cryptochlorogenic acid	C16H18O9	-	[M-H]-	353.0875	-1.42	353.0827,191.0556,179.0345,161.0236,135.0445	Angelica sinensis
105	27.35	Jujuboside B	C52H84O21	-	[M+HCOO]-	1089.5485	-0.48	1043.5393,911.4977,749.4831	Ziziphus jujuba
106	19.69	PolygalaxanthoneIX	C25H28O14	-	[M-H]-	551.1367	-7.82	488.6856,462.1201,243.0656,228.0419,165.0169,55.4000	Polygalae Radix
107	18.02	PolygalaxanthonVIII	C25H28O15	-	[M-H]-	567.1352	-1.41	552.1139,345.0598,315.0509,272.0397,271.0187,158.6542	Polygalae Radix
108	17.93	PolygalaxanthoneVII	C27H32O16	-	[M-H]-	611.1623	0.49	596.1373,303.0504,288.0266,287.0182,2444.0008	Polygalae Radix
109	21.90	PolygalaxanthoneIV	C27H32O15	-	[M-H]-	595.1674	0.67	497.1749,386,9544,287.0543,272.0306,229.0124,179.0403,96.9574,55.7343	Polygalae Radix
110	11.47	Sibiricose A6	C23H32O15	-	[M-H]-	547.1661	-1.65	341.1083,223.0610,205.0499,190.0266,175.0032,119.0349,89.0245,59.0137	Polygalae Radix
111	9.99	Sibiricose A5	C22H30O14	-	[M-H]-	517.1562	0.39	341.1076,193.0493,175.0395,160.0164,132.0215,89.0239,59.0142	Polygalae Radix

112	31.42	Atractylenolide III	C15H20O3	-	[M-H]-	247.1334	-2.44	203.1435,187.1113,185.0961,147.0784	Atractylodes macrocephala
113	6.85	Geniposidic acid	C19H18O8	-	[M-H]-	373.0926	-1.08	211.0614,167.0706,193.0505,123,0448,59.0136	Rehmanniae Radix Praeparata
114	4.85	Uridine	C15H16O3	-	[M-H]-	243.1053	9.50	200.0580,152.9033,110.0246,82.0299,66.0351	Rehmanniae Radix Praeparata
115	5.81	Guanosine	C10H13N5O5	-	[M-H]-	282.0842	0.71	150.0417.108.0201.80.0254.66.0098	Rehmanniae Radix Praeparata
116	6.70	Phenylalanine	C9H11NO2	-	[M-H]-	164.0716	-2.45	147.0452,119.0507,103.0552,91.0542,77.0396,72.0082,65.9976	Ziziphus jujuba
117	16.06	Liguiritigenin-7-O-D-apiosyl-4’-O-D-glucoside	C26H30O13	-	[M-H]-	549.16	-1.82	417.1167,255.0636,135.0075,119.0494	Glycyrrhiza uralensis
118	24.88	Apigenin	C15H10O5	-	[M-H]-	269.045	-3.73	239.0314,197.0573,133.0285,107.0130,79.4972,55.6688	Glycyrrhiza uralensis
119	19.74	Isoliquiritin apioside	C26H30O13	-	[M-H]-	549.1599	-2.01	417.1177,148.0163,135.0079,119.0498,91.0190	Glycyrrhiza uralensis
120	14.30	Schaftoside	C29H24O12	-	[M-H]-	563.1176	-2.49	545.1287,503.1183,443.0971,412.0879,383.0764,353.0658,325.0719,297.0764	Glycyrrhiza uralensis
121	20.43	Isoliquiritin	C21H22O9	-	[M-H]-	417.1173	-4.09	417.1161,297.0755,255.0639,180.0057,148.0158,135.0080,119.0497,91.0185	Glycyrrhiza uralensis
122	22.60	Genkwanin	C16H12O5	-	[M-H]-	283.0609	-0.35	268.0380,239.0348,211.0395,195.0444,169.7043,148.0109,135.0083,102.0373,76.6460	Glycyrrhiza uralensis
123	20.29	Ononin	C22H22O9.HCOOH	-	[M-HCOO]-	475.1242	-1.69	267.0650,052.0415,223.0391	Glycyrrhiza uralensis
124	20.29	Formononetin	C16H12O4	-	[M-H]-	267.0659	-0.38	252.0422,195.0435,167.0504,91.0186	Glycyrrhiza uralensis
125	9.19	L-Tryptophan	C11H12N2O2	-	[M-H]-	203.0826	-1.98	186.0554,159.0925,142.0653,118.5624,116.0505,95.1642,74.0244,72.0086,52.5718	Ziziphus jujuba
126	5.70	Adenosine	C10H13N5O4	-	[M-H]-	266.0893	1.13	184.5024,134.0473,107.0357,92.0245	Ziziphus jujuba

127	26.98	Isoliquiritigenin	C14H8O5	-	[M-H]-	255.0312	4.72	135.0080,119.0496,93.0339,91.0185	Rehmanniae Radix Praeparata、Ziziphus jujuba
128	22.23	Hydroxygenkwanin	C16H12O6	-	[M-H]-	299.0557	-1.01	256.0361,227.0343,158.0369,96.9697	
129	29.60	Notoginsenoside Ft1	C48H82O19	-	[M-HCOO]-	961.5377	-0.31	915.5301,783.4846,693.7268,621.4385,587.4127,418.6799,293.0947,193.5972,149.0441,	Panax ginseng
130	25.64	Ginsenoside Rb3 +HCOOH	C54H92O24	-	[M-HCOO]-	1123.5888	-1.96	1078.5819,1077.5767	Panax ginseng
131	24.20	Pseuoginsenoside F11	C42H72O14	-	[M-H]-	799.4817	-4.13	637.4291,475.3773,161.0447	Panax ginseng
132	8.12	Neochlorogenic acid	C16H18O9	-	[M-H]-	353.0877	-0.85	191.0563,179.0371,135.0444,96.9604,85.0294,53.7594	Angelica sinensis
133	12.60	Chlorogenic acid	C16H18O9	-	[M-H]-	353.0874	-1.70	191.0557,161.0251,85.0290,59.0136	Angelica sinensis
134	15.26	Glucosylvitexin	C27H30O15	-	[M-H]-	593.1507	-0.51	413.0897,311.0560,293.0455	Ziziphus jujuba
135	16.62	Verbascoside	C29H36O15	-	[M-H]-	623.1987	1.13	577.2811,461.1626,315.1085,161.0236,133.0287,113.0232	Rehmanniae Radix Praeparata
136	13.47	Purpureaside C	C35H46O20	-	[M-H]-	785.2496	-1.79	623.2170,477.1611,161.0233	Rehmanniae Radix Praeparata
137	17.42	Quercetin	C16H14O6	-	[M-H]-	301.0713	-2.33	255.2626,195.9980,179.0341,164.0116,139.0402,65.0044	Glycyrrhiza uralensis、Panax ginseng
138	1.58	Citric acid	C6H8O7	-	[M-H]-	191.0197	-1.58	129.0194,111.0086,93.0349,87.0087,85.0296,67.0188,57.0346	Ziziphus jujuba、Rehmanniae Radix Praeparata、Angelica sinensis
139	27.92	Diammonium glycyrrhizinate	C39H50O19	-	[M-H]-	821.2866	-0.49	351.0540,193.0343,113.0239	Glycyrrhiza uralensis
140	31.57	Glabrene	C20H18O4	-	[M-H]-	321.1124	-1.87	278.0575,266.0700,215.0891,199.0786,157.0786,121.0297,107.	Glycyrrhiza uralensis

								0497,79.0567	
141	15.79	Spinosin	C28H32O15	-	[M-H]-	607.166	-1.65	487.1218,445.1120,427.1005,324.0621,307.0594,292.0362,281.0441	Ziziphus jujuba
142	17.10	Isoferulic acid	C10H10O4	-	[M-H]-	193.0506	-2.08	178.0271,149.0602,137.0254,134.0363,117.0352,89.0388	Angelica sinensis
143	14.29	Isoschaftoside	C26H28O14	-	[M-H]-	563.14	-1.78	545.1287,503.1183,443.0971,383.0764,353.0658,325.0719,297.0764	Glycyrrhiza uralensis
144	16.65	Licoagroside D	C22H24O10	-	[M-H]-	447.1285	-3.36	415.5331,376.1606,285.0754,188.7080,149.0606,135.0093,91.0191	Glycyrrhiza uralensis
145	25.58	Ginsenoside Rh1	C36H62O9	-	[M + HCOO]−	683.4339	-6.42	637.4285,475.3772,161.0445,101.0240	Panax ginseng
146	30.02	Ginsenoside-Ro	C48H76O19	-	[M-H]-	955.4909	-0.10	799.5043,731.4311,678.2612,652.1297,613.3690,554.7943,343.2241,260.5904,221.0639,179.0052	Panax ginseng
147	24.20	Ginsenoside Rf	C42H72O14	-	[M-H]-	845.4864	-4.26	637.4277,475.3761,161.0444,101.0239	Panax ginseng
148	27.49	Ginsenoside Rd	C48H82O18	-	[M-H]-	991.5451	-2.93	946.5434,945.5370,783.4879	Panax ginseng
149	25.64	Ginsenoside Rc	C53H90O22	-	[M-H]-	1123.5888	-1.96	1078.5814,1077.5757	Panax ginseng
150	25.64	Ginsenoside Rb2	C53H90O22	-	[M-H]-	1123.5888	-1.96	1077.5757	Panax ginseng
151	33.38	Ginsenoside F2	C42H72O13	-	[M-H]-	829.4917	-3.98	783.4893,621.4276,572.6753,312.9850,209.3459,65.1306	Panax ginseng
152	2.12	Ginsenoside F1	C36H62O9	-	[M-H]-	683.4372	-1.17	406.9270,342.1125,161.0456,89.0244	Panax ginseng
153	12.25	Geniposide	C17H24O10	-	[M-HCOO]-	433.1348	-0.46	293.1263,225.0773,101.0239	Rehmanniae Radix Praeparata
154	41.30	Linoleic acid	C18H32O2	-	[M-H]-	279.2329	-0.36	269.1259,256.2332,209.8550,178.6480,160.1104,119.8130,110.0326,79.2623,55.8938	Ziziphus jujuba、Angelica sinensis
155	18.24	3,6’－Disinapoyl sucrose	C34H42O19	-	[M-H]-	753.223	-2.66	547.1641,529.1548,367.1028,325.0924,265.0713,223.0603,205.0491,190.0261,175.0031	Polygalae Radix
156	25.65	20(S)-Ginsenoside F1	C36H62O9	-	[M + HCOO]−	683.4339	-6.42	637.3762,161.0446,101.0235	Panax ginseng

157	15.23	p-Coumaric acid	C9H8O3	-	[M-H]-	163.0399	-0.62	145.8916,119.0499,117.0342,104.0269,93.0344,91.0547,72.2662,65.0401,51.0269	Rehmanniae Radix Praeparata
158	12.15	Caffeic acid	C6H12O6	-	[M-H]-	179.0563	1.68	135.0445,134.0362,107.0514,89.0395,79.0560	Panax ginseng
159	17.70	Naringin dihydrochalcone	C27H34O14	-	[M-H]-	581.1907	4.65	257.0088,229.0135,273.0404,258.0170,271.0247	Atractylodes macrocephala
160	24.69	Naringenin	C15H12O5	-	[M-H]-	271.0608	-0.74	253.0519,185.0611,165.0222,151.0032,145.0281,119.0501,107.0168,83.0145,65.0026	Glycyrrhiza uralensis
161	13.47	Echinacoside	C35H46O20	-	[M-H]-	785.2496	-1.79	623.2170,477.1611,161.0233,133.0286	Glycyrrhiza uralensis
162	25.31	Luteolin	C15H10O6	-	[M-H]-	285.0406	2.11	255.0366,227.0304,201.0595,184.0478,169.0681,145.0335,133.0270,119.0553,95.0140,78.9592,63.0232	Rehmanniae Radix Praeparata
163	25.31	Kaempferol	C15H10O6	-	[M-H]-	285.0406	2.11	255.0366,201.0595,184.0478,169.0681,145.0335,133.0270,119.0140,95.0140,78.9592,63.0232	Rehmanniae Radix Praeparata
164	8.88	8-Epiloganic acid	C17H20N4O6	-	[M-H]-	375.1292	-4.81	213.0769,169.0865,151.0758,125.0613,119.0352,95.0498,89.0244,59.0168	Glycyrrhiza uralensis、Panax ginseng
165	19.13	4-Hydroxybenzoic acid	C7H6O3	-	[M-H]-	137.0244	2.94	93.0399,75.0237,65.0394	Rehmanniae Radix Praeparata
166	25.14	Ginsenoside Rb1	C54H92O23	-	[M-H]-	1153.6003	-0.61	1107.5873,1108.5935	Panax ginseng
167	33.38	Ginsenoside F2	C42H72O13	-	[M-H]-	829.4917	-3.98	783.4893,621.4276,572.6753,312.9850,209.3459,65.1306	Panax ginseng

Note : * Representative compared with reference substance.

3.2 Results of network pharmacology analysis

3.2.1 Screening of components and disease targets of QFY

A total of 205 targets were obtained by screening the targets of Qifuyin components obtained by LC-MS. A total of 3971 disease targets were screened in the GeenCard database. Venny2.1 was used to obtain 112 components and disease intersection targets, as shown in Fig. S3.

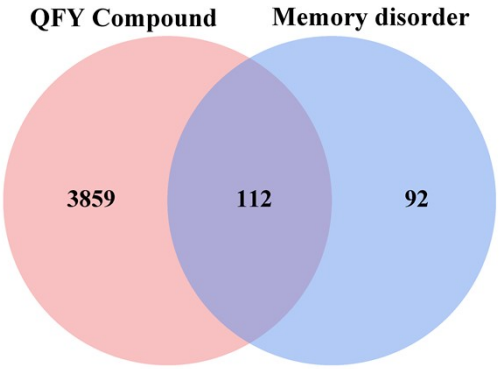


Fig. S3 Intersecting targets venn diagram

3.2.2Construction of protein-protein interaction (PPI) network and screening of key targets

The STRING online platform was used to analyze the protein-protein interaction (PPI) of the intersection targets, and the PPI data was visualized by Cytoscape 3.9.1, as shown in Fig. S4. Among them, the top 10 core targets were AKT1, GAPDH, TNF, SRC, PTGS2, EGFR, PPARG, STAT3, ESR1, and HSP90AA1.

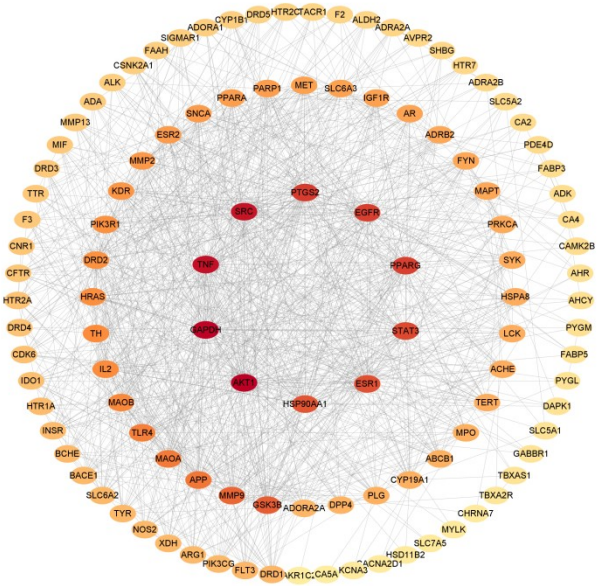


Fig. S4 PPI network diagram

3.2.3 “Drug-component-target-disease” network construction

Using the 112 intersection targets obtained above, a drug-component-target-disease net work was constructed in Cytoscape 3.9.2. As shown in Fig. S5, blue represents the QFY drug, green represents the chemical composition of QFY, orange represents the intersection target, and red represents the memory disorder disease. In terms of compounds, the active ingredients with the highest degree value are Apigenin, Luteolin, Hydroxygenkwanin, Quer cetin, Kaempferol, Isolicoflavonol, Genkwanin, Guanosine, Naringenin, Isoliquiritigenin and so on.

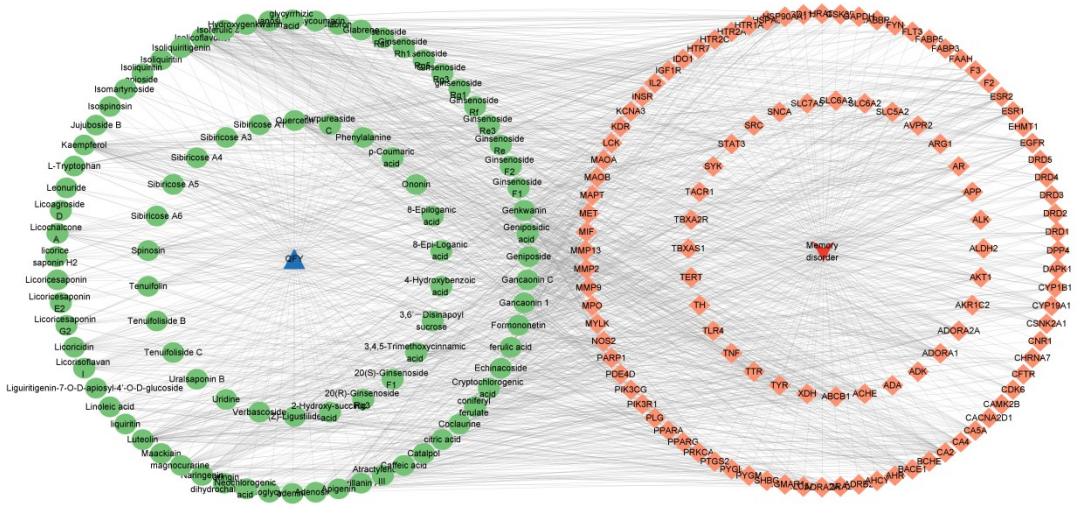
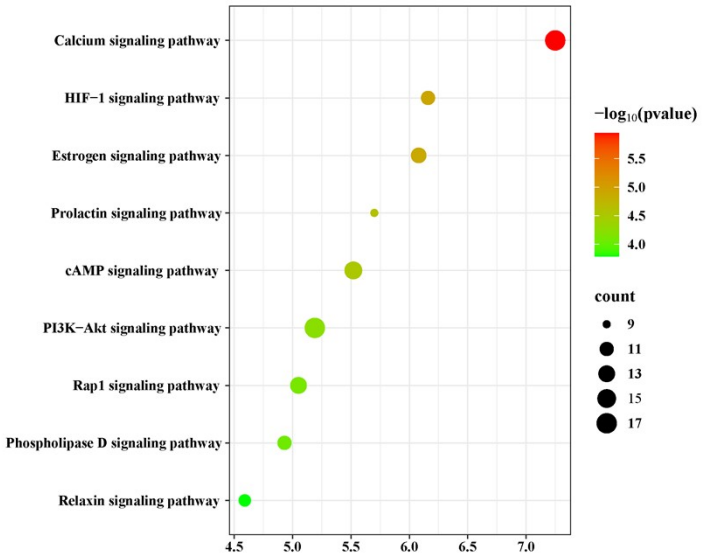


Fig. S5 Drug-component-target-disease visualization network

3.2.4 KEGG pathway enrich analysis

KEGG pathway enrichment analysis ($P < 0.05$) obtained 134 signal pathways, and the top 10 were plotted according to the P value ranking. The results are shown in Fig. S6. QFY may improve MD through the Calcium signaling pathway, HIF-1 signaling pathway, Estrogen signaling pathway, Prolactin signaling pathway, cAMP signaling pathway, and so



on.

Fig. S6 KEGG pathway enrichment analysis bubble diagram