

Article title: Plasma microRNAs as a potential biomarker for identification of progressive supranuclear palsy

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Table S1a Clinical characteristics of patients with PSP and healthy controls for initial miRNA screening

Variable	Control (n=9)	Case (n=12)	P-value
Age, year (mean±SE)	60.9±1.7	61.6±1.6	0.77
Gender, male/female	6/3	8/4	0.61
Disease duration, year (mean±SE)	-	2.4±0.37	
PSPRS (mean±SD)	-	40.8±1.06	
Hypertension (%)	0/9	0/12	
Diabetes (%)	0/9	0/12	
Smoking (%)	0/9	0/12	
Alcohol (%)	0/9	0/12	

Table S1b Demographic and clinical variables of patients with PSP and control subjects for validation

Variable	Controls (n=17)	Cases (n=18)	P-value
Age, year (mean±SE)	57.8±1.4	60.11±1.6	0.29
Gender, male/female	10/7	14/4	0.12
Disease duration, year (mean±SE)	-	2.37±1.17	
Hypertension (%)	4/17	3/18	
Diabetes (%)	2/17	3/18	
Smoking (%)	1/17	1/18	
Alcohol (%)	0/17	1/18	

N=number; S.E= standard error; SD=standard deviation; PSP= progressive supranuclear palsy; PSPRS= PSP rating scale.

Table S2 Plasma qPCR miRNAs profiling results with >1.5 or <1.5 fold change

miRNAs	Average Delta cq Case	Average Delta cq Control	Delta delta cq (case– control)	Fold change
hsa-miR-33a-5p	-1.49	-0.46	-1.03	2.04
hsa-miR-99a-5p	1.48	2.52	-1.04	2.06
hsa-miR-19b-3p	-4.50	-3.48	-1.02	2.03
hsa-miR-154-5p	2.46	3.18	-0.72	1.66
hsa-miR-92b-3p	4.01	5.22	-1.21	2.32
hsa-miR-32-5p	2.00	3.19	-1.19	2.28
hsa-miR-133a-3p	2.00	2.72	-0.72	1.65
hsa-miR-130a-3p	-3.50	-2.78	-0.72	1.65
hsa-miR-29b-3p	0.02	1.23	-1.21	2.31
hsa-miR-136-5p	1.50	2.20	-0.7	1.63
hsa-miR-142-5p	-2.51	-1.79	-0.72	1.65
hsa-miR-100-5p	4.51	5.15	-0.64	1.56
hsa-miR-326	0.00	0.72	-0.72	1.64
hsa-miR-193a-5p	4.48	5.13	-0.65	1.56
hsa-miR-127-3p	1.98	2.69	-0.71	1.64
hsa-miR-130b-3p	0.08	1.79	-1.71	3.29
hsa-miR-423-3p	-2.53	-1.81	-0.72	1.65
hsa-miR-210-3p	3.11	5.66	-2.55	5.85
hsa-miR-374a-5p	-0.50	0.21	-0.71	1.64
hsa-miR-877-5p	2.99	4.18	-1.19	2.28
hsa-miR-136-3p	2.51	3.73	-1.22	2.34
hsa-miR-29c-3p	-2.49	-1.78	-0.71	1.64
hsa-miR-543	3.72	2.51	1.21	2.31
hsa-miR-106a-5p	-2.50	-3.48	0.98	-1.96
hsa-miR-145-5p	-0.51	-1.47	0.96	-1.96
hsa-miR-424-5p	-0.49	-1.46	0.97	-1.96
hsa-miR-18b-5p	0.15	-1.32	1.47	-2.78
hsa-miR-223-5p	3.50	2.71	0.79	-1.73
has-miR-16-5p	-6.52	-6.48	-0.04	1.03

Table S3 Locked Nucleic Acid (LNA) microRNAs primers

miRNA	Mature miRNA sequence	Qiagen product number
hsa-miR-23a-3p	5'AUCACAUUGCCAGGGAUUUCC	YP00204772
hsa-miR-451a	5'AAACCGUUACCAUACUGAGUU	YP02119305
hsa- miR-19b-3p	5'UGUGCAAAUCCAUGCAAAACUGA	YP00204450
hsa-miR-33a-5p	5'GUGCAUUGUAGUUGCAUUGCA	YP00205690
hsa-miR-130b-3p	5'CAGUGCAAUGAUGAAAGGGCAU	YP00204317
hsa-miR-136-3p	5'CAUCAUCGUCUCAAUGAGUCU	YP00205503
hsa-miR-210-3p	5'CUGUGCGUGUGACAGCGGCUGA	YP00204333
hsa-miR-16-5p	5'UAGCAGCACGUAAAUAUUGGCG	YP00205702
RNA spike-in kit	UniSp2, UniSp6 & cel-miR-39-3p	339390

Table S4: Multiple KEGG pathways regulated by candidate microRNAs.

SNo.	KEGG pathway	p-value	#genes	#miRNAs
1	Fatty acid biosynthesis	1.89E-19	1	1
2	Endocytosis	7.82E-16	51	4
3	Hippo signaling pathway	3.60E-14	44	4
4	FoxO signaling pathway	6.76E-14	42	4
5	Protein processing in endoplasmic reticulum	9.87E-12	40	4
6	Estrogen signaling pathway	2.47E-11	24	4
7	Viral carcinogenesis	2.97E-11	29	3
8	p53 signaling pathway	4.23E-11	25	3
9	TGF-beta signaling pathway	9.09E-11	24	4
10	Proteoglycans in cancer	9.04E-09	35	4
11	Adrenergic signaling in cardiomyocytes	1.13E-08	26	4
12	Signaling pathways regulating pluripotency of stem cells	1.30E-08	26	3
13	AMPK signaling pathway	2.42E-08	34	3
14	Glioma	1.32E-07	17	3
15	Prostate cancer	1.92E-07	25	3
16	Chronic myeloid leukemia	2.30E-07	21	4
17	Colorectal cancer	5.94E-07	17	4
18	Hepatitis B	8.80E-07	29	4
19	Sphingolipid signaling pathway	9.84E-06	23	3
20	Endometrial cancer	1.44E-05	16	3
21	Pancreatic cancer	2.34E-05	17	4
22	Melanoma	2.47E-05	16	3
23	Pathways in cancer	7.34E-05	50	4

24	Cocaine addiction	7.81E-05	8	2
25	Non-small cell lung cancer	7.81E-05	14	3
26	PI3K-Akt signaling pathway	0.000225	46	3
27	Progesterone-mediated oocyte maturation	0.000246	20	3
28	Thyroid cancer	0.000302	7	3
29	Long-term depression	0.000406	12	3
30	Prolactin signaling pathway	0.000406	15	3
31	Acute myeloid leukemia	0.000867	14	3
32	Oocyte meiosis	0.001016	22	4
33	Chagas disease (American trypanosomiasis)	0.002782	20	4
34	Central carbon metabolism in cancer	0.005534	13	3
35	Bladder cancer	0.006369	10	3
36	Insulin signaling pathway	0.006369	23	4
37	Dopaminergic synapse	0.006829	23	4
38	Platelet activation	0.008042	19	4
39	Melanogenesis	0.008474	17	3
40	Fatty acid metabolism	0.008727	3	3
41	Hepatitis C	0.009513	22	3
42	Cholinergic synapse	0.011449	16	4
43	HTLV-I infection	0.012462	33	4
44	Oxytocin signaling pathway	0.028471	23	4
45	Toxoplasmosis	0.033598	17	4
46	Gap junction	0.037395	13	3
47	Cell cycle	0.038744	22	4
48	Renal cell carcinoma	0.044706	12	4

SI Table S5. Sorted KEGG pathways regulated by candidate microRNAs and associated target genes identified using three databases.

	FoxO signal ing path way	TGF- beta signal ing path way	Signalin g pathwa ys regulati ng pluripot ency of stem cells	p53 signal ing path way	ErbB signal ing path way	Prola ctin signal ing path way	Protein processi ng in endoplas mic reticulu m	Cell cycle	Insuli n signal ing path way	PI3K- Akt signal ing path way	Hipp o signal ing path way	Wnt signal ing path way
hsa - miR - 19b -3p	IRS2 CCNB 1 STK4 PIK3C B SETD 7 MAP K14 CCND 2 RAF1 CDKN 1B SMA D4 S1PR 1 MAP K8 PRKA A1 CDKN 1A PTEN TGFB R2 BCL2 L11	INHBA ACVR 1 SMA D4 ZFVY E9 SMA D5 BAM BI TGFB R2	GSK3B FZD5 PIK3CB MAPK1 4 RAF1 INHBA ACVR1 SMAD4 SMAD5	ZMAT3 CCNB 1 CCND 2 CHEK 1 CASP 3 CDKN 1A PTEN	GSK3B PIK3CB RAF1 CDKN 1B MAP K8 CDKN 1A	GSK3B PIK3CB MAP K14 CCND 2 RAF1 MAP K8	DNAJA1 SAR1B UBQLN2 SEC24C MAPK8 HSP90B 1 UBE2D3	GSK3B CCNB 1 CCN A2 CCN D2 CDK N1B CHEK 1 SMA D4 CDK N1A	IRS2 GSK3B PPP2 R5E PIK3CB RAF1 CAL M2 SREB F1 MAP K8 PRKAA1	GSK3B FZD5 CCND 2 SMA D4 FRM D6 PARD 6B TGFB R2	GSK3B FZD5 CCND 2 SMA D4 MAP K8 BAM BI	

hsa - miR - 33a -5p	IRS2 CCNB 1 SETD 7 MAP K14 RAF1 CDKN 1B MAP K8 SOS1 BCL2 L11	MYC SMA D5 SP1	FZD5 MAPK1 4 RAF1 MYC SMAD5	CCNB 1 CASP 3	RAF1 CDKN 1B MAP K8 MYC SOS1	MAP K14 RAF1 MAP K8 SOS1	SAR1B HERPUD 1 UBQLN2 SEC24C MAPK8 VCP HSP90B 1	CCNB 1 CDK N1B MYC	IRS2 RAF1 SREB F1 MAP K8 SOS1	PPP2 R5E RAF1 CDKN 1B MYC SOS1 HSP9 OB1 BCL2L 11	FZD5 MYC PARD 6B	GSK3 B CCND 2 SMA D4 MYC BAM BI
hsa - miR - 130 b- 3p	CCNB 1 STK4 PIK3C B CCND 2 SMA D4 S1PR 1 SOS1 PRKA A1 CDKN 1A PTEN TGFB R2 BCL2 L11	INHBA ACVR 1 SMA D4 MYC ZFYV E9 SMA D5 SP1 BAM BI TGFB R2	GSK3B PIK3CB INHBA ACVR1 SMAD4 MYC SMAD5	ZMA T3 CCNB 1 CCND 2 CHEK 1 CDKN 1A PTEN	GSK3 B PIK3C B MYC SOS1 CDKN 1A	GSK3 B PIK3C B CCND 2 SOS1	DNAJA1 SAR1B HERPUD 1 CANX VCP UBE2D3	GSK3 B CCNB 1 CCN A2 CCN D2 CHEK 1 SMA D4 MYC CDK N1A	GSK3 B PIK3C B CAL M2 SOS1 PRKA A1	GSK3 B PIK3C B CCND 2 MYC SOS1 PRKA A1 CDKN 1A PTEN BCL2L 11	GSK3 B CCND 2 SMA D4 MYC FRM D6 TGFB R2	FZD5 MAP K8 MYC
hsa - miR - 136 -3p	TGFB 2	TGFB 2					CANX		PPP1 CB		TGFB 2 PPP1 CB	