Supporting Information for

Short Communication

A systematic strategy for screening therapeutic constituents of *Schisandra chinensis* (Turcz.) Baill infiltrated blood−brain barrier oriented in lesions using ethanol and water extracts: a novel perspective for exploring chemical material basis of herb medicines

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Table S1 Chronic unpredictable stress procedure.

Day	Stressor		
	Morning	Afternoon	Night
1	Cage tilting (45°, 6 h)	Sawdust-free cage (12 h)	Overnight illumination
2	Electric shock (0.8 mA, 5 s duration, 60 s inter-shock interval)	–	Water deprivation (overnight)
3	Soiled cage (200 mL water sprinkled on sawdust bedding for 12 h)	–	Food deprivation (overnight)
4	Restraint stress	Horizontal shaking cage (30min)	White noise (30 min)
5	Cold swimming (10 °C)	–	Sawdust-free cage (12 h)
6	Sawdust-free cage (12 h)	Electric shock (0.8 mA, 5 s duration, 60 s inter-shock interval)	–
7	Horizontal shaking cage (30 min)	Cage tilting (45°, 6 h)	–
8	White noise (30 min)	Electric shock (0.8 mA, 5 s duration, 60 s inter-shock interval)	–
9	Soiled cage (200 mL water sprinkled on sawdust bedding for 12 h)	Horizontal shaking cage (30 min)	Overnight illumination
10	Water deprivation (12 h)	–	Eage tilting (45°, 6 h)
11	Restraint stress	Cold swimming (10 °C)	–
12	Electric shock (0.8 mA, 5 s duration, 60 s inter-shock interval)	Horizontal shaking cage (30 min)	Food deprivation (overnight)
13	Sawdust-free cage (12 h)	White noise (30 min)	–
14	Horizontal shaking cage (30 min)	Restraint stress	Soiled cage (200 mL water sprinkled on sawdust bedding for 12 h)

–Not applicable.

Table S2 Optimized multiple-reaction-monitoring (MRM) parameters for all samples.

Analyte	Mode (±)	Q1(amu)	Q3 (amu)	DP (V)	CE (eV)	t_R (min)
NE	+	170.1	152.0	38	21	2.01
5-HT	+	177.2	160.2	43	12	2.23
DA	+	154.1	137.1	42	14	2.58
Isoprenaline (IS)	+	212.0	194.0	46	12	2.82

Table S3 The calibration parameters of analytes.

Analyte	Liner range (μg/mL)	Slope	Intercept	Regression coefficient (*R*)
DA	1.00–200	7.340×10^{-2}	2.375×10^{-2}	0.9962
NE	5.00–1000	1.795×10^{-2}	4.833×10^{-1}	0.9953
5-HT	5.00–1000	6.060×10^{-2}	6.775×10^{-2}	0.9977

Table S4 Summary of precision of analytes in rat brain (n = 6).

Analyte	Intra-day (RSD, %)				Inter-day (RSD, %)			
	LLOQ	Low	Medium	High	LLOQ	Low	Medium	High
DA	6.0	3.4	4.6	7.6	13.8	4.4	10.5	8.7
NE	12.6	5.4	9.8	6.5	10.3	5.6	7.5	1.9
5-HT	15.8	8.7	4.3	5.0	12.5	4.9	6.4	8.5

Table S5 Summary of accuracy of analytes in rat brain (n = 6).

Analyte	Intra-day (RE, %)				Inter-day (RE, %)			
	LLOQ	Low	Medium	High	LLOQ	Low	Medium	High
DA	8.0	12.3	–8.9	7.3	14.3	14.5	4.5	–6.2
NE	–5.7	10.2	12.9	–8.8	–6.1	3.9	9.9	5.1
5-HT	–1.2	7.7	4.3	–3.4	–9.3	4.6	–6.2	–6.6

Table S6 Summary of matrix effect of analytes in rat brain (n = 6).

Analyte	IS-normalized MF (%)			RSD (%)		
	Low	Medium	High	Low	Medium	High
DA	110.4	91.8	98.1	12.5	12.1	7.1
NE	96.7	91.4	102.6	8.9	9.3	7.3
5-HT	90.8	101.3	82.6	6.5	8.7	5.9

Table S7 Stability of analytes in brain at different conditions (n = 3).

Analytes	−80 °C for a month (RE, %)		Three freeze-thaw cycles (RE, %)		4 °C in autosampler for 24 h (RE, %)		1 week at r.t. for derivatized analytes (RE, %)	
	Low	High	Low	High	Low	High	Low	High
DA	12.1	−5.7	13.2	8.9	12.8	6.8	7.2	3.3
NE	12.3	11.3	6.9	9.8	−2.3	−10.3	1.1	2.9
5-HT	10.6	12.5	−13.7	−10.4	3.8	4.8	2.5	1.9

Table S8 Precision, repeatability and stability in the method validation of contents *in vitro*.

RT (min)	Fragment ions (*m*/*z*)	Precision (RSD, %)		Repeatability (RSD, %)		Stability (RSD, %)	
		RT	Intensity	RT	RT	Intensity	RT
5.05	221.0583	0.6	3.3	0.2	2.6	0.3	2.9
16.82	419.1992	0.5	5.1	0.4	2.6	0.8	4.3
24.49	433.2148	0.8	4.4	0.0	3.4	0.1	2.5
31.90	417.1835	0.0	1.8	1.2	0.9	0.8	2.4
50.60	537.2046	0.3	2.2	1.5	3.4	0.6	4.5
62.02	403.2042	0.7	2.8	1.1	1.9	0.9	3.3
77.81	417.2199	1.2	4.8	0.6	4.8	0.2	4.6
85.09	385.1573	0.5	4.9	0.2	1.9	0.0	5.0

Table S9 Precision, repeatability and stability in the method validation of contents *in vivo*.

RT (min)	Fragment ions (*m*/*z*)	Precision (RSD, %)		Repeatability (RSD, %)		Stability (RSD, %)	
		RT	Intensity	RT	Intensity	RT	Intensity
5.05	221.0583	0.5	6.9	1.2	7.1	1.0	9.5
16.82	419.1992	1.4	10.1	0.7	5.9	0.9	4.8
24.49	433.2148	0.9	6.8	1.4	7.8	0.8	9.9
31.90	417.1835	1.3	8.2	0.7	10.5	0.2	9.1
50.60	537.2046	0.6	9.2	0.3	6.2	0.6	6.7
62.02	403.2042	0.7	5.9	1.5	8.0	1.3	7.9
77.81	417.2199	1.0	7.9	0.8	9.1	0.7	5.1
85.09	385.1573	0.4	10.8	0.3	7.7	0.3	10.2

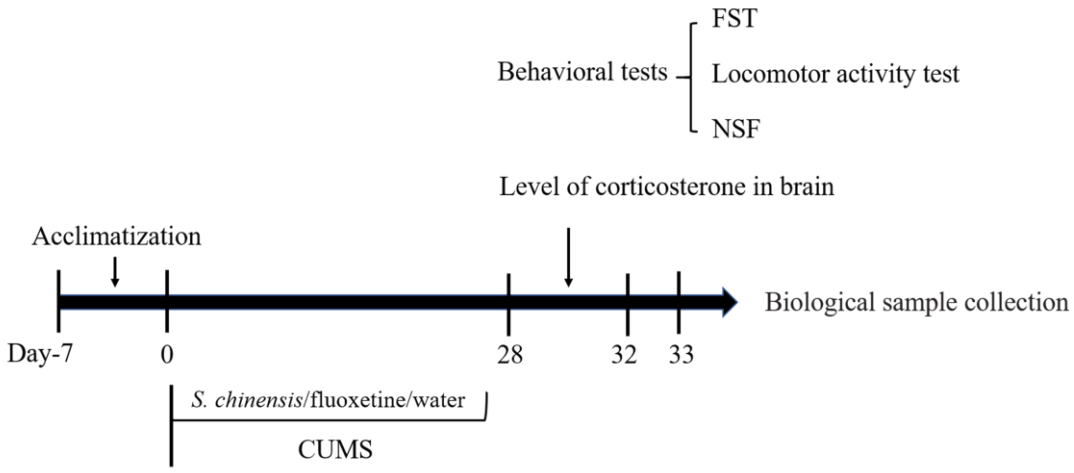


Figure S1 The experimental design of our study.

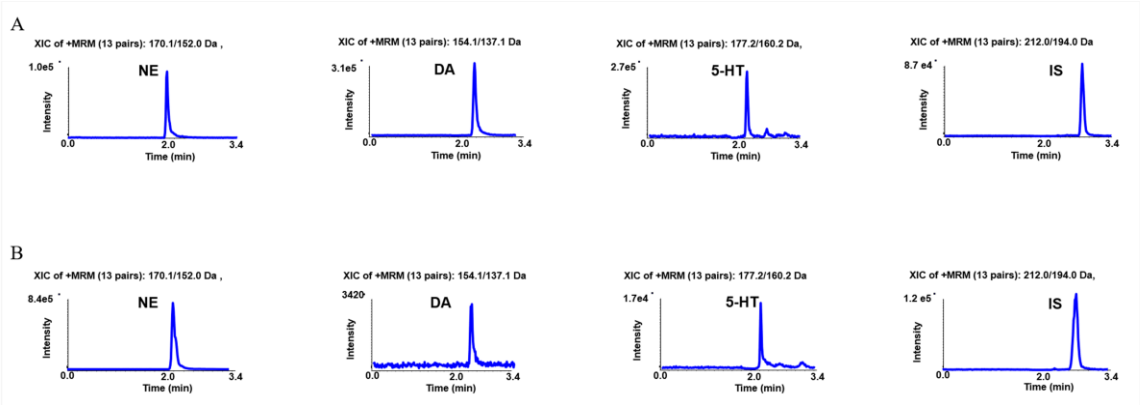
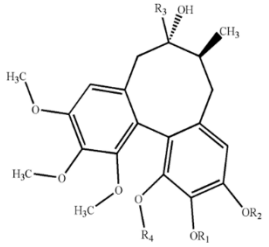
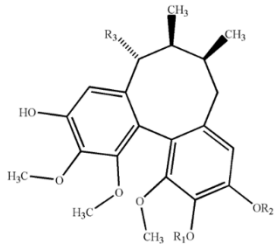


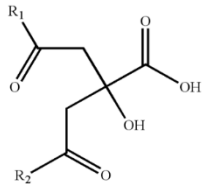
Figure S2 Typical LC–MS MRM chromatograms of (A) reference standard solution of analytes and IS. (B) 5-HT, DA and NE in rat brain sample.



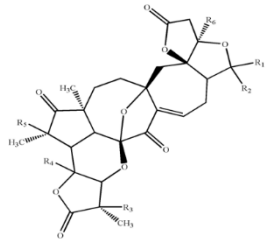
Componund	R_1	R_2	R_3	R_4
Schisandrol B	——CH2——		Me	Me
Schisandrin A	Me	Me	H	Me
Angeloygomisin H	Me	Me	OH	Ang
Tigloylgomisin H	Me	Me	OH	Tig
Benzoylgomisin H	Me	Me	Me	Ben



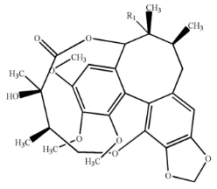
Componund	R_1	R_2	R_3
wilsonilignan C	——CH2——		OMe
Gomisin O	——CH2——		OH
Gomisin N	——CH2——		H
Schisanhenol	Me	Me	H
Gomisin S	Me	Me	OH



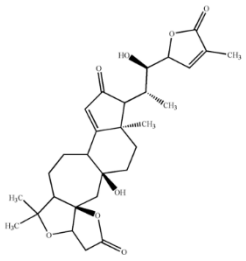
Componund	R_1	R_2
Citric acid	OH	OH
1,5 – Dimethyl citrate	Me	Me



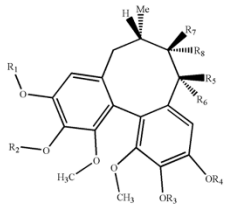
Componund	R_1	R_2	R_3	R_4	R_5	R_6
Schindilactone A	Me	Me	H	H	H	OH
Lancifodilactone C	Me	Me	H	H	H	H



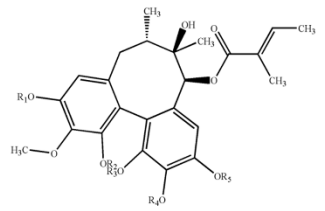
Componund	R_1
Gomisin D	OH
Gomisin E	H



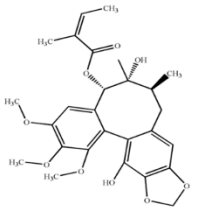
Propindilactone G



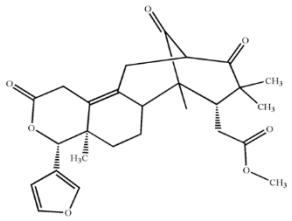
Componund	R_1	R_2	R_3	R_4	R_5	R_6	R_7	R_8
Schisandrin B	Me	Me	——CH2——		H	H	H	Me
Schisandrin C	——CH2——		——CH2——		H	H	H	Me
Gomisin J	Me	H	Me	H	H	H	H	Me
Epigomisin O	Me	Me	——CH2——		H	OH	H	Me
Tigloylgomisin Q	Me	Me	Me	Me	Me	OTig	Me	OH
Angeloylgomisin Q	Me	Me	Me	Me	Me	OAng	Me	OH
Gomisin C	Me	Me	——CH2——		OBen	H	Me	OH
Schisantherin B	Me	Me	——CH2——		OAng	H	Me	OH
Schisantherin C	Me	Me	——CH2——		OTig	H	Me	OH
Schisantherin D	——CH2——		——CH2——		OBen	H	Me	OH
Gomisin G	——CH2——		Me	Me	OBen	H	Me	OH
Schisantherin E	OH	Me	Me	Me	OBen	H	Me	OH
Gomisin F	——CH2——		Me	Me	OTig	H	Me	OH
Benzoylgomisin Q	Me	Me	Me	Me	OBen	H	Me	OH
Benzoylgomisin P	——CH2——		Me	Me	OBen	H	Me	OH
Benzoylgomisin O	Me	Me	——CH2——		OBen	H	H	Me
Rubrisandrin A	OH	Me	Me	OH	H	H	H	Me
Schisphenlignan A	OH	Me	——CH2——		OBen	H	OH	Me



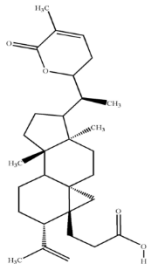
Componund	R_1	R_2	R_3	R_4	R_5
Schisanwilsonin D	H	H	Me	Me	Me
Schisanwilsonin B	Me	Me	OH	——CH2——	



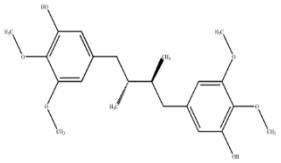
Schisanwilsonin A



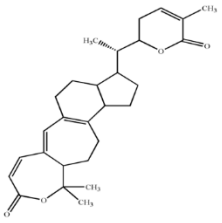
Mexicanolide



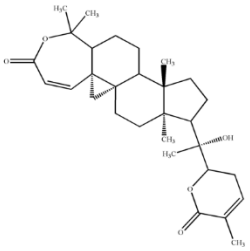
Schisanlactone E



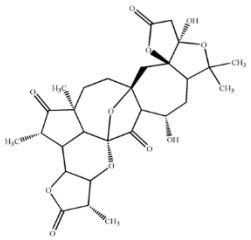
Pregomisin



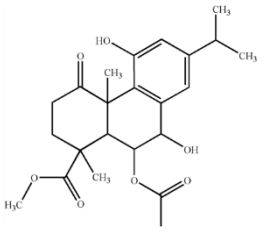
Schisanlactone B



Kadsuphilactone B



Schisanlactone D



PRXFPPAVWQDTHT-UHFFFAOYSA-N

Figure S3 Structures of the components of *S. chinensis.*

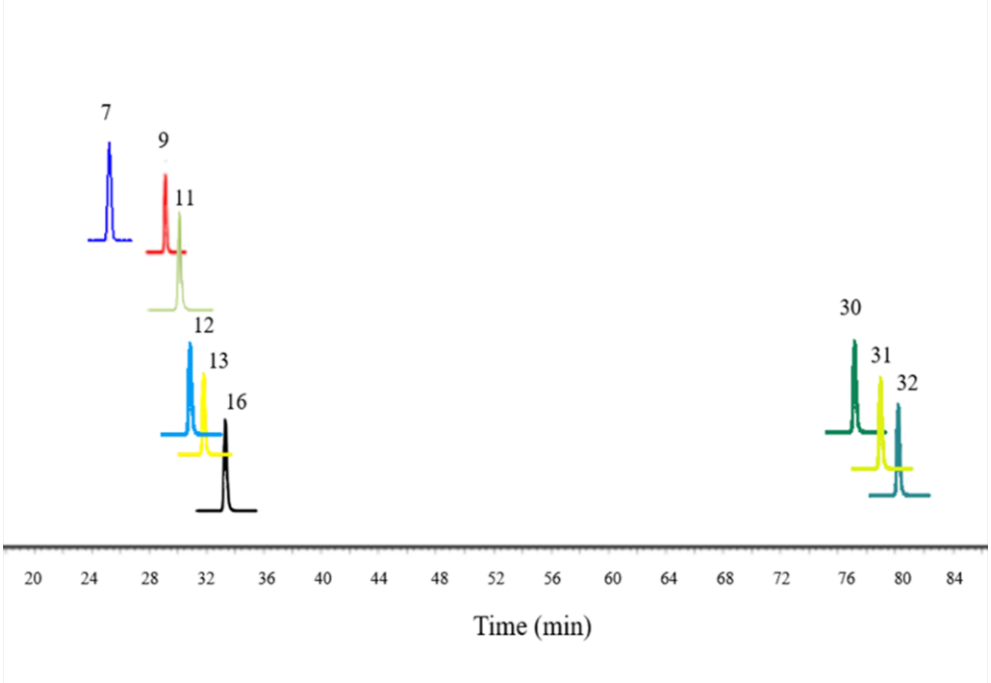


Figure S4 Results of 9 reference standards:7, schisandrol A; 9, gomisin D; 11, gomisin J; 12, gomisin O; 13, epigomisin O; 16, schisandrol B; 30, schisandrin A; 31, gomisin N; 32, schisandrin B (the numbers represent the peak No. in Table 2).

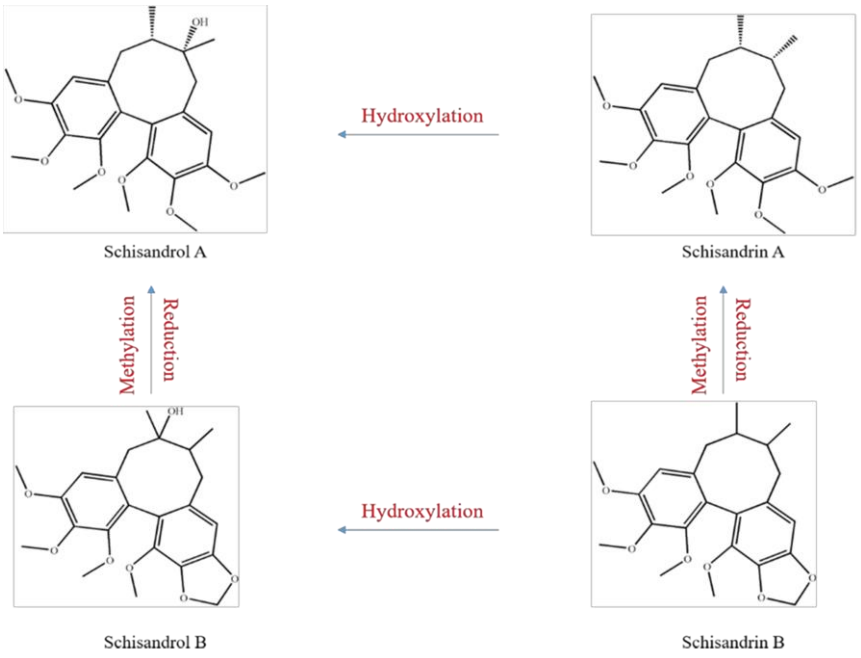


Figure S5 Proposed metabolic pathways of transform process between schisandrol A, schisandrol B, schisandrirn A, schisandrin B.

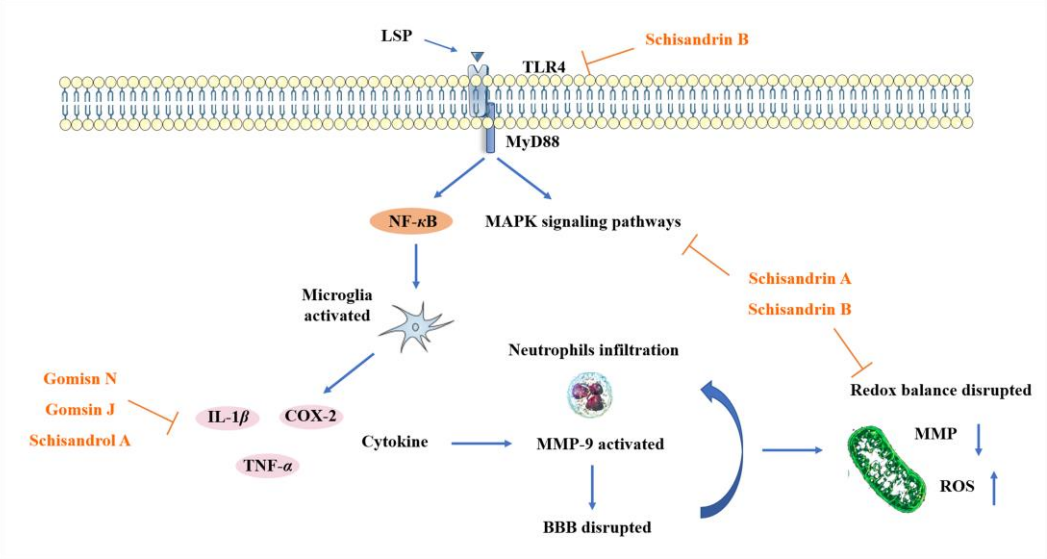


Figure S6 The active ingredients of *S. chinensis* protect against oxidative stress and inflammation in CNS diseases.