

SUPPLEMENTARY MATERIALS

Antidepressant-Like Effect of the Eburnamine-Type Molecule Vindeburnol in Rat and Mouse Models of Ultrasound-Induced Depression

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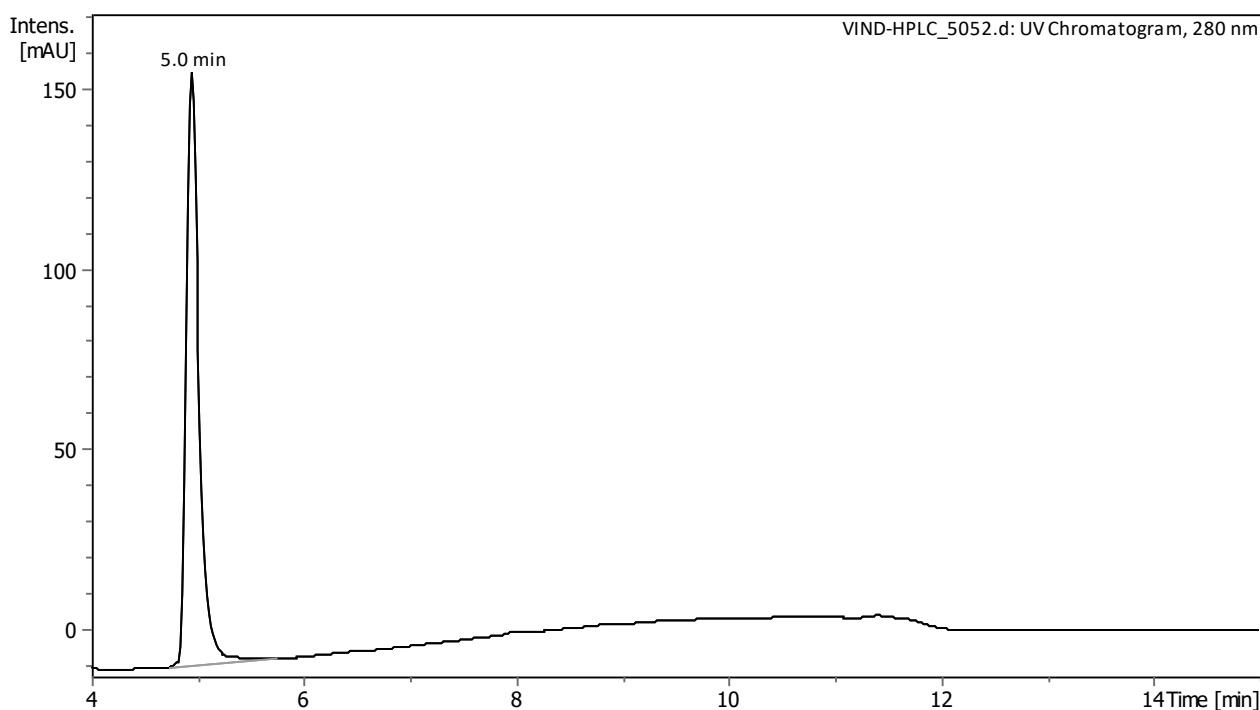
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HPLC purity of vindeburnol

Purity of vindeburnol was analyzed by analytical high-performance liquid chromatography (HPLC) on an Elute HPLC system (Bruker Daltonik, Bremen, Germany) equipped with an Azura UVD 2.1S UV detector (Knauer, Berlin, Germany) with a wavelength at 280 nm and acquisition rate at 1 Hz. Chromatographic separation was carried out on an Acquity HSS T3 column (Waters Corporation, Milford, MA USA; 2.1 × 100 mm, 1.3 μm, 100 Å) at 30 °C, sample injection volume – 1.0 μL. A mobile phase consisting of 0.1 % formic acid in water (A), and 0.1 % formic acid in acetonitrile (B) was programmed with gradient elution of 5-95% over 9 min at a flow rate of 250 μL/min. Data were processed using Compass DataAnalysis 5.1 software (Bruker Daltonik).



#	RT [min]	Area	S/N	Area Total [%]
1	5.0	1384.1	4567	100.00

Table S1. Statistical analysis of behavioural tests results using ordinary one-way ANOVA followed by Tukey's multiple comparison tests

Behavioural test	Sucrose preference test					
	F (2, 46) = 11.39; <i>p</i> value < 0.0001			F (2, 24) = 24.34; <i>p</i> < 0.0001		
Experimental groups	Mice			Rats		
	Saline	US-exposed	VIND-treated	Saline	US-exposed	VIND-treated
Saline	-	< 0.0001	0.1564	-	< 0.0001	0.3039
US-exposed	< 0.0001	-	0.0071	< 0.0001	-	< 0.0001
VIND-treated	0.1564	0.0071	-	0.3039	< 0.0001	-
Behavioural test	Social interaction test					
	F (2, 39) = 19.45; <i>p</i> < 0.0001			F (2, 24) = 17.32; <i>p</i> < 0.0001		
Experimental groups	Mice			Rats		
	Saline	US-exposed	VIND-treated	Saline	US-exposed	VIND-treated
Saline	-	0.0067	0.0221	-	0.0024	0.2219
US-exposed	0.0067	-	< 0.0001	0.0024	-	< 0.0001
VIND-treated	0.0221	< 0.0001	-	0.2219	< 0.0001	-
Behavioural test	Open field test, path					
	F (2, 45) = 6.51; <i>p</i> = 0.0033			F (2, 21) = 24.70; <i>p</i> < 0.0001		
Experimental groups	Mice			Rats		
	Saline	US-exposed	VIND-treated	Saline	US-exposed	VIND-treated
Saline	-	0.3703	0.0026	-	< 0.0001	< 0.0001
US-exposed	0.3703	-	0.1075	< 0.0001	-	0.9117
VIND-treated	0.0026	0.1075	-	< 0.0001	0.9117	-

Behavioural test	Open field test, rears					
	F (2, 45) = 2.607; <i>p</i> = 0.0849			F (2, 21) = 44.35; <i>p</i> < 0.0001		
Experimental groups	Mice			Rats		
	Saline	US-exposed	VIND-treated	Saline	US-exposed	VIND-treated
Saline	–	0.0903	0.1801	–	< 0.0001	0.0569
US-exposed	0.0903	–	0.8551	< 0.0001	–	< 0.0001
VIND-treated	0.1801	0.8551	–	0.0569	< 0.0001	–
Behavioural test	Forced swimming test					
	F (2, 45) = 34.47; <i>p</i> < 0.0001			F (2, 28) = 37.93; <i>p</i> < 0.0001		
Experimental groups	Mice			Rats		
	Saline	US-exposed	VIND-treated	Saline	US-exposed	VIND-treated
Saline	–	< 0.0001	0.9798	–	< 0.0001	0.4334
US-exposed	< 0.0001	–	< 0.0001	< 0.0001	–	< 0.0001
VIND-treated	0.9798	< 0.0001	–	0.4334	< 0.0001	–

Table S2. Concentrations of noradrenaline (NA), dopamine (DA), homovanillic acid (HVA), 3,4-dihydroxyphenylacetic acid (DOPAC), 5-hydroxytryptophan (5-HTP), serotonin (5-HT), 5-hydroxyindoleacetic acid (HIAA), and 3-*O*-methyldopa (3-OMD) in BALB/c mouse brainstem

Experimental groups	Monoamine concentration (mean±SD), pmol/mg of total protein							
	NA	DA	HVA	DOPAC	5-HTP	5-HT	HIAA	3-OMD
Saline	5.58±0.26	0.60±0.06	0.61±0.13	0.40±0.07	0.17±0.05	9.28±0.41	5.85±0.37	0.49±0.12
US-exposed	5.69±0.33	0.63±0.08	0.52±0.09	0.39±0.04	0.17±0.07	8.77±1.77	4.73±1.00	0.44±0.08
VIND-treated	6.37±0.39	0.72±0.05	0.70±0.08	0.58±0.06	0.13±0.04	8.92±0.69	5.35±0.61	0.44±0.10

Table S3. Statistical analysis of BALB/c mouse brainstem monoamine levels results using ordinary one-way ANOVA followed by Tukey's multiple comparison tests

Monoamine levels	Experimental groups	Saline	US-exposed	VIND-treated
NA F (2, 15) = 8.240 <i>p</i> = 0.0038	Saline	-	0.8620	0.0052
	US-exposed	0.8620	-	0.0147
	VIND-treated	0.0052	0.0147	-
DA F (2, 15) = 5.063 <i>p</i> = 0.0209	Saline	-	0.7877	0.0218
	US-exposed	0.7877	-	0.0772
	VIND-treated	0.0218	0.0772	-
HVA F (2, 15) = 4.118 <i>p</i> = 0.0375	Saline	-	0.3139	0.3870
	US-exposed	0.3139	-	0.0297
	VIND-treated	0,3870	0,0297	-
DOPAC F (2, 15) = 17.68 <i>p</i> = 0.0001	Saline	-	0.9584	0.0004
	US-exposed	0.9584	-	0.0003
	VIND-treated	0.0004	0.0003	-
5-HTP F (2, 15) = 0.7465 <i>p</i> = 0.4908	Saline	-	0.9987	0.5392
	US-exposed	0.9987	-	0.5684
	VIND-treated	0.5392	0.5684	-
5-HT F (2, 15) = 0.5140 <i>p</i> = 0.6083	Saline	-	0.5969	0,7673
	US-exposed	0.5969	-	0.9561
	VIND-treated	0.7673	0.9561	-
HIAA F (2, 15) = 3.106	Saline	-	0.0614	0.5217
	US-exposed	0.0614	-	0.3777

$p = 0.0743$	VIND-treated	0.5217	0.3777	-
3-OMD	Saline	-	0.6620	0.7099
F (2, 15) = 0.4711	US-exposed	0.6620	-	0.9965
$p = 0.6332$	VIND-treated	0.7099	0.9965	-

Table S4. Concentrations of dopamine (DA), 3-methoxytyramine (3-MT), 3,4-dihydroxyphenylacetic acid (DOPAC), serotonin (5-HT), and homovanillic acid (HVA) in BALB/c mouse striatum

Experimental groups	Monoamine concentration (mean±SD), pmol/mg of total protein				
	DA	3-MT	DOPAC	5-HT	HVA
Saline	258.8±41.9	12.4±3.1	24.2±3.7	11.5±2.8	34.5±5.1
US-exposed	256.1±25.9	13.1±2.8	27.2±3.0	11.4±2.3	34.1±3.5
VIND-treated	295.8±38.1	16.8±3.2	29.1±5.2	13.2±2.9	33.8±4.0

Table S5. Statistical analysis of BALB/c mouse striatum monoamine levels results using ordinary one-way ANOVA followed by Tukey's multiple comparison tests

Monoamine levels	Experimental groups	Saline	US-exposed	VIND-treated
DA F (2, 33) = 4.189 <i>p</i> = 0.0239	Saline	–	0.9835	0.0544
	US-exposed	0.9835	–	0.0369
	VIND-treated	0.0544	0.0369	–
3-MT F (2, 33) = 6.670 <i>p</i> = 0.0037	Saline	–	0.8696	0.0051
	US-exposed	0.8696	–	0.0185
	VIND-treated	0.0051	0.0185	–
DOPAC F (2, 33) = 3.941 <i>p</i> = 0.0292	Saline	–	0.2221	0.0233
	US-exposed	0.2221	–	0.5266
	VIND-treated	0.0233	0.5266	–
5-HT F (2, 33) = 1.546 <i>p</i> = 0.2281	Saline	–	0.9975	0.3093
	US-exposed	0.9975	–	0,2790
	VIND-treated	0.3093	0.2790	–
HVA F (2, 33) = 0.08488 <i>p</i> = 0.9188	Saline	–	0.9698	0.9116
	US-exposed	0.9698	–	0.9834
	VIND-treated	0.9116	0.9834	–