Supplementary Material

Quinoline Alkaloids with Anti-inflammatory Activity from

Zanthoxylum avicennae

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Fig. S1 ECD spectrum of compound 1

Fig. S2 Chiral HPLC separation of compound 1



*Lux[®] Amylose-1 (250 mm ×10 mm, S-5 μ m) LC column (UV = 254 nm)

Mobile Phase: n-hexane/isopropanol 7:3, v/v, 3ml.min⁻¹

(+)**-1**:(−)**-1** ≈ 3:1



Fig. S3 Key ¹H-¹H COSY and HMBC correlations of **2–6**



Fig. S4 Chiral HPLC separation of compound 7

*Lux[®] Amylose-1 (250 mm ×10 mm, S-5 μ m) LC column (UV = 254 nm)

Mobile Phase: n-hexane/isopropanol 85:15, v/v, 3ml.min⁻¹

(+)**-7**:(−)**-7** ≈ 1:1



Fig. S5 ECD spectra of two enantiomers (+)-7 and (-)-7

Fig. S6 Chiral HPLC separation of compound 8



*Lux[®] Amylose-1 (250 mm ×10 mm, S-5 μ m) LC column (UV = 254 nm) Mobile Phase: n-hexane/isopropanol 7:3, v/v, 3ml.min⁻¹

(+)**-8**:(−)**-8** ≈ 1:1







Fig. S8. The effect of Compound 2–10 on the gene expression level of IL-1 β in RAW264.7 cells. The results are shown as the mean \pm s.e.m., n=3. Data were analyzed by student's T-test (two-tailed). *P < 0.05 compared with the Ctrl group. The Ctrl (Control) group means RAW264.7 macrophages were treated with LPS (100 ng/mL) but without compounds.

	5^{a}		6 ^b			
No.	$\delta_{ m H}$	$\delta_{ m C}$	$\delta_{ m H}$	$\delta_{ m C}$		
2		154.1		154.7		
3	6.20 s	111.6	6.25 s	111.5		
4		178.0		178.0		
4a		126.7		126.8		
5	8.44 dd (7.6, 1.5)	126.8	8.46 dd (7.6, 1.7)	126.9		
6	7.38 t (7.6)	123.6	7.38 t (7.6)	123.5		
7	7.67 ddd (8.6, 7.6, 1.5)	132.3	7.67 ddd (8.7, 7.6, 1.7)	132.2		
8	7.54 d (8.6)	115.6	7.51 (8.7)	115.4		
8a		142.2		142.1		
1'	2.73 m (2H)	34.4	2.72 m (2H)	34.1		
2'	1.96 m (2H)	22.4	1.76 m (2H)	28.5		
3'	2.58 t (6.3) (2H)	41.2	2.14 m (2H)	32.0		
4'		210.4	5.40 dt (14.5, 6.8)	127.5		
5'	2.39 t (7.3) (2H)	45.1	5.54 dt (14.5, 6.4)	134.1		
6'	1.59 m (2H)	17.4	2.02 m (2H)	25.7		
7'	0.91 t (7.3)	13.9	0.98 t (7.5)	14.1		
N-Me	3.84 s	34.5	3.74 s	34.3		

Table S1 ¹H and ¹³C NMR data for **5** and **6** in CDCl₃ (δ in ppm and J in Hz)

^{*a*} NMR data was recorded at 400 MHz (¹H) and 125 MHz (¹³C). ^{*b*} NMR data was recorded at 600 MHz (¹H) and 150 MHz (¹³C).

	-
Empirical formula	C ₁₆ H ₁₉ NO ₅ ^{<i>a</i>}
Formula weight	305.32
Temperature	170.02 K
Wavelength	1.34139 Å
Crystal system	Triclinic
Space group	P-1
Unit cell dimensions	a = 8.5101(3) Å $\alpha = 77.6570(10)^{\circ}$.
	b = 9.0807(3) Å β = 83.0550(10)°.
	c = 9.5903(3) Å $\gamma = 88.4020(10)^{\circ}$.
Volume	718.67(4) Å ³
Z	2
Density (calculated)	1.411 Mg/m ³
Absorption coefficient	0.561 mm ⁻¹
F(000)	324
Crystal size	0.12 x 0.1 x 0.08 mm ³
Theta range for data collection	4.134 to 55.059°.
Index ranges	-10<=h<=10, -11<=k<=11, -11<=l<=11
Reflections collected	15698
Independent reflections	2729 [R(int) = 0.0540]
Completeness to theta = 53.594°	99.5 %
Absorption correction	Semi-empirical from equivalents
Max. and min. transmission	0.7508 and 0.5779
Refinement method	Full-matrix least-squares on F ²
Data / restraints / parameters	2729 / 0 / 207
Goodness-of-fit on F ²	1.046
Final R indices [I>2sigma(I)]	R1 = 0.0391, $wR2 = 0.1024$
R indices (all data)	R1 = 0.0425, wR2 = 0.1055
Extinction coefficient	n/a
Largest diff. peak and hole	0.253 and -0.251 e.Å ⁻³
CCDC number	2160133
^{<i>a</i>} Crystals of compound 1 were obtained from	om MeOH.

Table S2 X-ray crystallographic data for compound 1

	- · ·	
Empirical formula	$C_{15}H_{17}NO_5^a$	
Formula weight	291.29	
Temperature	170.04 K	
Wavelength	1.34139 Å	
Crystal system	Monoclinic	
Space group	P 1 21 1	
Unit cell dimensions	a = 9.7252(4) Å	$\alpha = 90^{\circ}$
	b = 7.2357(3) A	$\beta = 98.737(2)^{\circ}$
	c = 10.0005(4) A	$\gamma = 90^{\circ}$
Volume	695.56(5) Å ³	
Z	2	
Density (calculated)	1.391 Mg/m ³	
Absorption coefficient	0.563 mm ⁻¹	
F(000)	308	
Crystal size	$0.12 \ x \ 0.08 \ x \ 0.06 \ mm^3$	
Theta range for data collection	5.996 to 55.068°.	
Index ranges	-11<=h<=11, -8<=k<=8,	, - 12<=l<=12
Reflections collected	9299	
Independent reflections	2585 [R(int) = 0.0640]	
Completeness to theta = 53.594°	99.0 %	
Absorption correction	Semi-empirical from equ	uivalents
Max. and min. transmission	0.7508 and 0.5615	
Refinement method	Full-matrix least-squares	s on F ²
Data / restraints / parameters	2585 / 1 / 201	
Goodness-of-fit on F ²	1.051	
Final R indices [I>2sigma(I)]	R1 = 0.0351, wR2 = 0.0351	812
R indices (all data)	R1 = 0.0401, wR2 = 0.03	869
Absolute structure parameter	0.11(16)	
Extinction coefficient	n/a	
Largest diff. peak and hole	0.159 and -0.178 e.Å ⁻³	
CCDC number	2160134	
^a Crystals of compound (-)-1 were obtained	l from MeOH.	

 Table S3 X-ray crystallographic data for compound (-)-1

Empirical formula	C ₁₅ H ₁₇ NO ₄	
Temperature	169 99 K	
Wavelength	1 34139 Å	
Crystal system	Monoclinic	
Snace group	P 1 21 1	
Unit cell dimensions	a = 8.5948(3) Å	$\alpha = 90^{\circ}$
	b = 8.5216(3) Å	$\beta = 100.0350(10)^{\circ}.$
	c = 18.7212(6) Å	$\gamma = 90^{\circ}.$
Volume	1350.19(8) Å ³	
Z	4	
Density (calculated)	1.354 Mg/m ³	
Absorption coefficient	0.522 mm ⁻¹	
F(000)	584	
Crystal size	0.12 x 0.1 x 0.08 mm	3
Theta range for data collection	4.545 to 54.944°.	
Index ranges	-10<=h<=10, -10<=k	<=10, -22<=1<=22
Reflections collected	14913	
Independent reflections	5038 [R(int) = 0.0437	7]
Completeness to theta = 53.594°	98.7 %	
Absorption correction	Semi-empirical from	equivalents
Max. and min. transmission	0.7508 and 0.6254	
Refinement method	Full-matrix least-squa	ares on F ²
Data / restraints / parameters	5038 / 1 / 371	
Goodness-of-fit on F ²	1.065	
Final R indices [I>2sigma(I)]	R1 = 0.0331, wR2 = 0.0331	0.0843
R indices (all data)	R1 = 0.0361, wR2 = 0.0361	0.0874
Absolute structure parameter	0.02(9)	
Extinction coefficient	n/a	
Largest diff. peak and hole	0.192 and -0.173 e.Å	-3
CCDC number	2160135	
^{<i>a</i>} Crystals of compound (+)-7 were obtained	l from MeOH.	

 Table S4 X-ray crystallographic data for compound (+)-7

 Table S5 Primer sequences used in RT-PCR analysis

Genes	Forward primer	Reverse primer
β -actin	TGGTACCACAGGCATTGTGAT	TGATGTCACGCACGATTTCCCT
Illb	GCAACTGTTCCTGAACTCAACT	ATCTTTTGGGGGTCCGTCAACT
<i>Il6</i>	TAGTCCTTCCTACCCCAATTTCC	TTGGTCCTTAGCCACTCCTTC

Fig. S9 ¹H NMR spectrum of 1 in CDCl₃











Fig. S12 HMBC spectrum of 1



Fig. S13 COSY spectrum of 1



Fig. S14 ROESY spectrum of 1



Fig. S15 ESI-MS spectrum of 1



Fig. S16 HRESI-MS spectrum of 1



Fig. S17 IR spectrum of 1



Fig. S18 ¹H NMR spectrum of 2 in CDCl₃



Fig. S19 ¹³C NMR spectrum of 2 in CDCl₃













Fig. S23 ESI-MS spectrum of 2



Fig. S24 HRESI-MS spectrum of 2

Elemental Composition Report

Page 1 Single Mass Analysis Tolerance = 10.0 PPM / DBE: min = -1.5, max = 50.0 Element prediction: Off Number of isotope peaks used for i-FIT = 9 Monoisotopic Mass, Even Electron Ions 232 formula(e) evaluated with 1 results within limits (up to 50 best isotopic matches for each mass) Elements Used: C: 0-100 H: 0-100 N: 0-5 O: 0-10 LCT PXE KE324 12-Sep-2018 17:10:26 JAZ-3-2-2-w 24 (0.494) Cm (24:25) 1: TOF MS ES+ 3.03e+003 310.1440 100-% 311.1483 302.1743 312.1453 313.5178 314.9419 304.2897 317.6214 319.7456.320.2288 321.7671 m/z 318.0 320.0 322.0 306.6501 308.1232 0-302.0 304.0 306.0 308.0 310.0 312.0 314.0 316.0 Minimum: -1.5 Maximum: 5.0 10.0 Mass Calc. Mass mDa PPM DBE i-FIT i-FIT (Norm) Formula 310.1440 310.1443 -0.3 -1.0 10.5 514.9 0.0 C19 H20 N O3



Fig. S25 IR spectrum of 2

Fig. S26 ¹H NMR spectrum of 3 in CDCl₃







Fig. S28 HSQC spectrum of 3









Fig. S31 ESI-MS spectrum of 3



Fig. S32 HRESI-MS spectrum of 3

Elemental Composition Report





Fig. S33 IR spectrum of 3









Fig. S36 HSQC spectrum of 4







Fig. S38 COSY spectrum of 4



Fig. S39 ESI-MS spectrum of 4



Fig. S40 HRESI-MS spectrum of 4





Fig. S41 IR spectrum of 4

Fig. S42 ¹H NMR spectrum of 5 in CDCl₃



Fig. S43 ¹³C NMR spectrum of 5 in CDCl₃



Fig. S44 HSQC spectrum of 5



Fig. S45 HMBC spectrum of 5



Fig. S46 COSY spectrum of 5



Fig. S47 ESI-MS spectrum of 5



Fig. S48 HRESI-MS spectrum of 5

Elemental Composition Report

Single Mass Analysis Tolerance = 2.0 PPM / DBE: min = -1.5, max = 50.0 Element prediction: Off Number of isotope peaks used for i-FIT = 9 Monoisotopic Mass, Even Electron lons 241 formula(e) evaluated with 1 results within limits (up to 50 closest results for each mass) Elements Used: C: 0-100 H: 0-100 N: 0-2 O: 0-20 Na: 0-1 LCT PXE KE324 06-Dec-2018 19:04:10 JAZ-18-4-2-W+ 23 (0.477) Cm (22:28) 1: TOF MS ES+ 1.53e+004 272.1653 100-% 272.0781 271.9690 272.0316 272.2685 272.3815 T m/z 0 272.100 272.300 272.400 272.500 272.600 272.200 272,000 271.700 271.800 271.900 -1.5 50.0 Minimum: 5.0 2.0 Maximum: PPM DBE i-FIT i-FIT (Norm) Formula mDa Mass Calc. Mass 0.0 C17 H22 N O2 78.6 272.1653 272.1651 0.2 0.7 7.5

Page 1



Fig. S49 IR spectrum of 5

Fig. S50 ¹H NMR spectrum of 6 in CDCl₃



Fig. S51 ¹³C NMR spectrum of 6 in CDCl₃



Fig. S52 HSQC spectrum of 6



Fig. S54 COSY spectrum of 6



Fig. S55 ROESY spectrum of 6





Fig. S56 ESI-MS spectrum of 6

Fig. S57 HRESI-MS spectrum of 6

Qualitative Analysis Report

Data Filename ESIH_20190709_YJM_JKL_03.d Sample Name JAZ-4-B1 Sample Type Sample Position P1-A2 Agilent G6520 Q-TOF 20160322_MS_ESIH_POS_1min.m **Instrument Name** Acq Method 7/9/2019 15:13:57 **IRM Calibration Status** Acquired Time Su DA Method small molecular data analysis method.m Comment ESIH by ZZY User Spectra



Diff (mDa) Diff (ppm) Calc m/z m/z 256.1697 256.1696 -0.24 C17 H22 N O (M+H)+ -0.06

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Fig. S58 IR spectrum of 6







Fig. S60 ¹³C NMR spectrum of (+)-7 in CD₃OD



Fig. S61 ¹H NMR spectrum of (-)-7 in CD₃OD





Fig. S63 HSQC spectrum of 7



Fig. S64 COSY spectrum of 7





ESIL_20190830_YJM_JKL_14.#11-12_RT: 0.17-0.19_AV: 2_SB: 9_0.02-0.06_, 0.94-1.00_NL: 3.89E6 T: ITMS + c_ESI Full ms [50.00-2000.00]



Fig. S66 HRESI-MS spectrum of 7

Qualitative Analysis Report

Data Filen Sample Ty Instrumen Acquired T DA Method User Spect	ame pe it Name l'ime d	ESIH_2019 Sample Agilent G65 8/30/2019 small moled	0830_YJM 520 Q-TOF 20:01:05 cular data	1_JKL_13.d : analysis meth	od.m	Sample Nam Position Acq Method IRM Calibra Comment	ie tion Status	JAZ-6-5B P1-B7 20160322_M Success ESIH by ZZY	S_ESIH_F	°OS_1min	i.m	
Fragm	entor Voltage 135	Co	ollision Ei 0	nergy	Ionization M ESI	1ode						
x10 ⁶	+ESI Scan (r	: 0.283 mir	n) Frag=1	35.0V ESIH_	_20190830_Y	JM_JKL_13.c	I					
1.8 1.6 1.4- 1.2- 1					276.12:	35						
0.8												
0.6 - 0.4 - 0.2 - 0 -	269 2	70 271	272	274.177 273 274	74 275 276	277.1275	279.1595 279 280	281 282	283	284 28	85 286	287
Formula C	alculator Res	ults			Count	s vs. Mass-to-	Charge (m/z)					

		ico. iteoaiteo				
m/z		Calc m/z	Diff (mDa)	Diff (ppm)	Ion Formula	Ion
	276.1235	276.123	-0.42	-1.51	C15 H18 N O4	(M+H)+
						201 - 201 - 201 - 201 - 201 - 201 - 201 - 201 - 201 - 201 - 201 - 201 - 201 - 201 - 201 - 201 - 201 - 201 - 201

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Fig. S68 ¹³C NMR spectrum of (+)-8 in CDCl₃





Fig. S69 ¹H NMR spectrum of (-)-8 in CDCl₃

Fig. S70 ESI-MS spectrum of 8

