Supporting Information

***Rec. Nat. Prod*. 7:3 (2013) 250-253**

Phytochemical Study of *Myrtopsis corymbosa*, Perspectives for Anti-dengue Natural Compound Research

Paul Coulerie[1], Alexandre Maciuk[2], E. Hnawia[1], N. Lebouvier[1], Jean-Claude Guillemot[3], Bruno Canard[3], Bruno Figadère[2] and Mohammed Nour[1*]

[1] Laboratoire Insulaire du Vivant et de l'Environnement, Université de la Nouvelle-Calédonie, BP R4, 98851 Nouméa Cedex, Nouvelle-Calédonie

[2] Laboratoire BioCIS, UMR CNRS 8076 Université Paris-Sud XI, 5 rue J.-B. Clément, 92296 Châtenay-Malabry, France

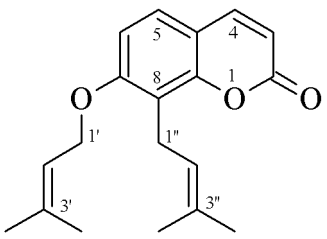
[3] Laboratoire AFMB, UMR 6098, Université Aix-Marseille, Campus de Luminy, 13288 Marseille cedex 09, France

Table of Contents **Page**

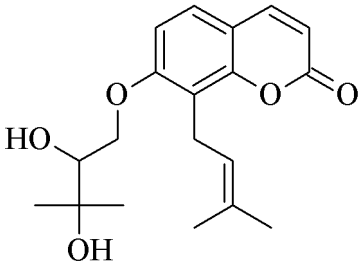
S2: Procedure for the purification of coumarins from the barks of *M. corymbosa*

500 g of dry bark powder was extracted with CH_2Cl_2 under agitation and sonication at room temperature (3 × 3 L, 6 h each). Once vacuum-dried, CH_2Cl_2 crude extract (9.5 g) was subjected to silica gel chromatography (63-200 µm; 4×30 cm, Merck) eluted with a step gradient of CH_2Cl_2/MeOH 100:0, 95:5, 70:30 and 0:100 (1 L each) to obtain 10 fractions (F1-F10, 250 mL each). Compounds **1**, **2** and **3** were respectively purified from F4 (240 mg), F5 (4.3 g) and F6 (260 mg) using a preparative HPLC on a sunfire RP-18 column (19x250 mm, 10 µm, Waters) eluted with an isocratic mix of TFA 0.5%/MeOH 45:55 at flow rate 17 mL/min.

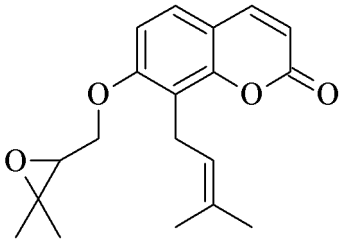
Position		**1**		**1**		**3**	
		^{1}H	^{13}C	^{1}H	^{13}C	^{1}H	^{13}C
1	O	-		-	-	-	-
2	C=O		161.6		161.5	-	161.5
3	CH	6.20 d (9.0)	112.9	6.22 d (9.0)	113.3	6.24 d (9.0)	113.4
4	CH	7.59 d (9.0)	143.9	7.58 d (9.0)	143.8	7.62 d (9.0)	143.9
5	CH	7.24 d (9.0)	126.2	7.28 d (9.0)	126.5	7.29 d (9.0)	126.5
6	CH	6.80 d (9.0)	108.7	6.83 d (9.0)	108.5	6.84 d (9.0)	108.5
7	C^{IV}	-	159.7	-	159.5	-	159.4
8	C^{IV}	-	118.4	-	118.2	-	118.2
9	C^{IV}	-	153.0	-	153.0	-	152.9
10	C^{IV}	-	112.9	-	113.0	-	113.1
1'	CH_2	4.60 d (8.0)	65.8	4.24 d (8.0)	68.6	4.24 d (2.0 ; 8.0); 3.85 d (2.0 ; 8.0)	70.1
2'	CH	5.48 t (8.0)	121.3	3.22 m	73.3	4.08 t (8.0)	75.9
3'	C^{IV}	-	138.3	-	71.3	-	71.5
4'	CH_3	1.80 s	25.8	1.38 s	26.5	1.33 s	26.7
5'	CH_3	1.75 s	18.1	1.36 s	24.8	1.28 s	25.1
1"	CH_2	3.53 d (8.0)	22. 2	3.63 dd (3.0 ; 8.0)	22.2	3.55 m	22.2
2"	CH	5.23 t (8.0)	121.3	5.24 t (8.0)	121.6	5.14 t (8.0)	121.6
3"	C^{IV}	-	131.6	-	133.1	-	133.3
4"	CH_3	1.83 s	25.8	1.83 s	25.8	1.82 s	25.7
5"	CH_3	1.67 s	17.1	1.68 s	17.9	1.68 s	18.1



1



2



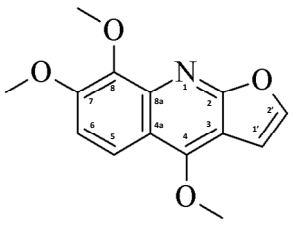
3

S3: ^{1}H and ^{13}C NMR data obtained with compounds 1, 2 and 3 in $CDCl_3$ (δ ppm, *J* (Hz)).

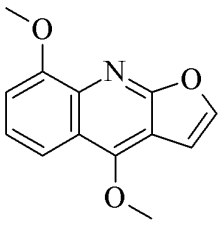
S4: Protocol for isolation of alkaloids from leaves of *M. corymbosa*

200 g of dry leave powder, spread for 24 h with ammonia 20%, was extracted with CH_2Cl_2 under agitation and sonication at room temperature (3 L, 3 h). The alkaloidal fraction (AF) was obtained by an acid–base extraction. CH_2Cl_2 was partitioned between 2% acetic acid CH_2Cl_2. The aqueous layer was adjusted to pH 10 with ammonia and alkaloids were extracted with chloroform and dried (0.035% dry weight/weight). Compound **4**, **5** and **6** were then separated on a preparative HPLC on a sunfire RP-18 column (19x250 mm, 10 µm, Waters) eluted with an isocratic mix of TFA 0.1%/MeOH 50:50.

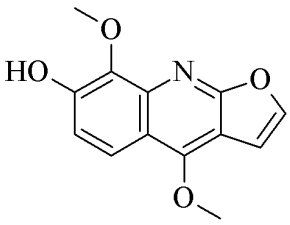
		4		5		6	
Position		**^{1}H**	**^{13}C**	**^{1}H**	**^{13}C**	**^{1}H**	**^{13}C**
1	N	-	-	-	-	-	-
2	C^{IV}	-	160.7	-	160.7	-	160.7
3	C^{IV}	-	103.2	-	104.4	-	103.3
4	C^{IV}	-	163.6	-	159.8	-	163.6
4a	C^{IV}	-	113.3	-	117.8	-	113.6
4b	O-CH_3	4.49 (s)	60.8	4.42 (s)	59.2	4.47 (s)	60.6
5	CH	8.04 (d. *J=9.2Hz*)	119.7	7.73 (d. *J=8.0Hz*)	114.7	7.82 (d. *J=9.2 Hz*)	119.9
6	CH	7.33 (d. *J=9.2Hz*)	114.0	7.33 (t. *J=8.0Hz*)	124.2	7.26 (d. *J=9.2 Hz*)	115.7
7	Cx	-	155.0	7.05 (d. *J=8.0Hz*)	109.7	-	151.2
7b	O-CH_3	4.04 (s)	56.9	-	-	-	-
8	C^{IV}	-	137.8	-	152.3	-	139.1
8a	C^{IV}	-	133.3	-	133.6	-	133.5
8b	O-CH_3	4.01 (s)	61.5	3.36 (s)	56.7	3.88 (s)	61.6
1'	CH	7.28 (d. *J=2.5Hz*)	106.2	7.09 (d. *J=2.5Hz*)	105.8	7.13 (d. *J=2.5Hz*)	106.1
2'	CH	7.65 (d. *J=2.5Hz*)	144.0	7.59 (d. *J=2.5Hz*)	143.4	7.60 (d. *J=2.5Hz*)	143.9



4



5



6

S5: ^{1}H and ^{13}C NMR data obtained with compounds **4**, **5** and **6** in CDCl3 (δ ppm, *J* (Hz)).

Isolated compounds	Inhibition of the DENV-NS5 RdRp (%)		
	at 50 µM	at 10 µM	at 1 µM
1	49 ±6	44 ±6	23 ±3
2	75 ±2	56 ±2	17 ±9
3	26 ±4	19 ±5	0 ±0
4	46 ± 2	12 ± 4	1 ± 1
5	37 ± 6	29 ± 4	16 ± 5
6	33 ± 5	28 ± 6	21 ± 4
dGTP	-	100	100

S6: Inhibition of the DENV-NS5 RdRp by coumarins (1-3) and alkaloids (4-6) isolated from *M. corymbosa* (dGTP was used as positive control).