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# Cytotoxic, Anti-hemolytic, and Antioxidant Activities of *Ruta chalepensis* L. (Rutaceae) Extract, Fractions, and Isolated Compounds

## Spectroscopic Data

### Summary

This research reported the previous isolation of chalepensin (CHL; Figure 1.1) in DOI: 10.3390/molecules191221044 [1], rutamarin (RTM; Figure 2.1) in DOI: 10.3390/molecules26123684 [2], and graveoline (GRV; Figure 3.1) in DOI: 10.1055/s-0036-1596528 [3], whose identification was based on spectroscopic/spectrometric analysis obtained by a Bruker Spectrometer (Model Advance DPX400, 9.4 Teslas; Bruker Corporation, Billerica, MA, USA) and in comparison with bibliographic data; all structures were matched on the PubChem website (https://pubchem.ncbi.nlm.nih.gov/).

### Spectroscopic Data

***Chalepensin (CHL)***

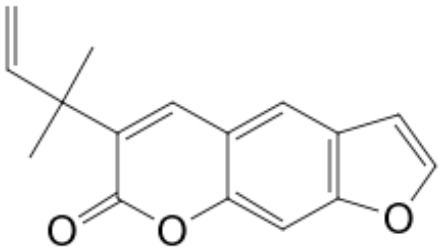

**Figure S1.1.** Structure of chalepensin (CHL). PubChem CID = 128834 (https://pubchem.ncbi.nlm.nih.gov/compound/Chalepensin).

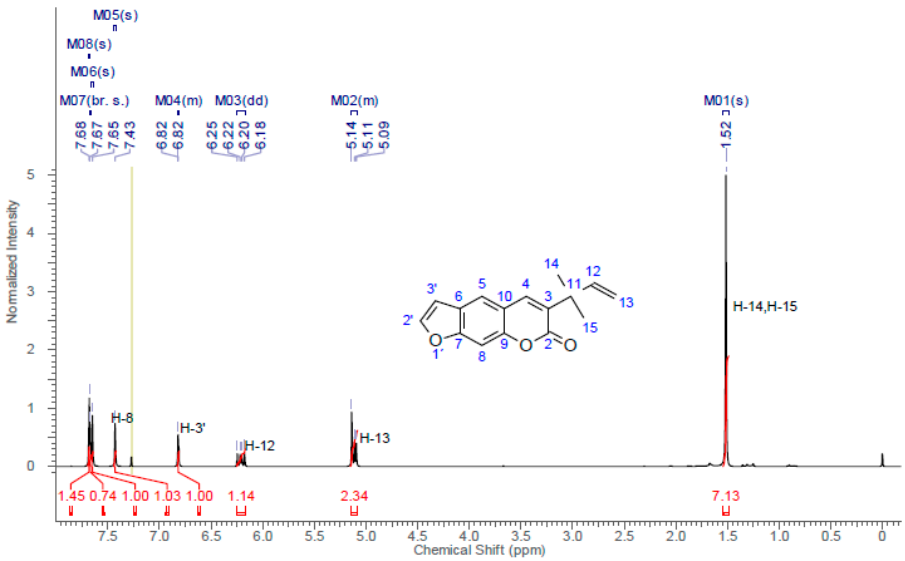


**Figure S1.2.1.** $^{1}$H-NMR spectrum of CHL (Deuterochloroform ($CDC_{13}$), 400 MHz).

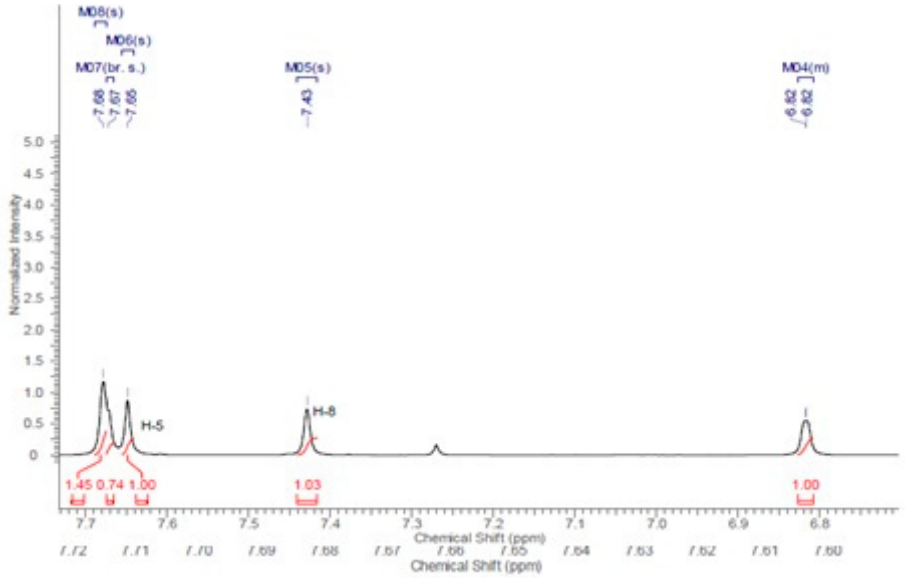


**Figure S1.2.2.** $^{1}$H-NMR spectrum of CHL ($CDC_{13}$, 400 MHz). Expansion in the area from 7.7 - 6.7 ppm.

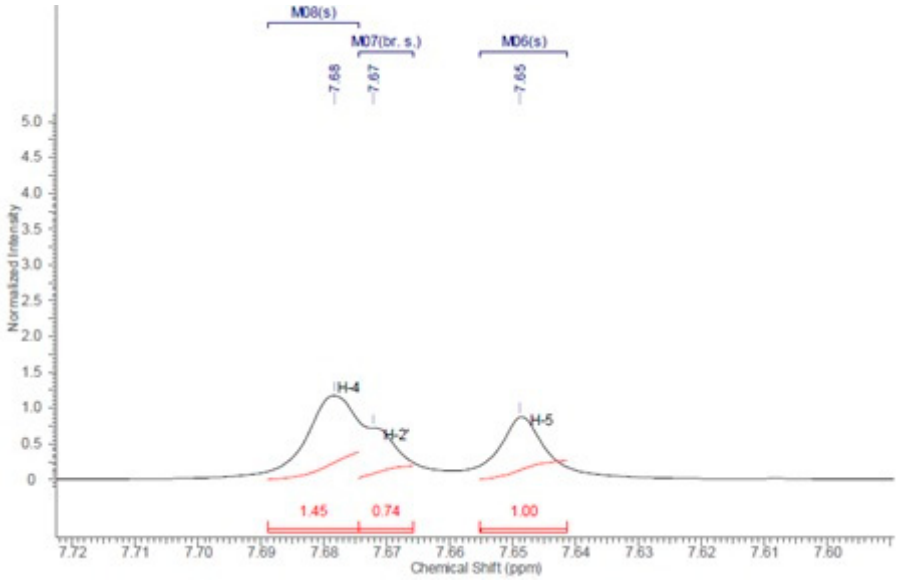


**Figure S1.2.3.** $^{1}$H-NMR spectrum of CHL ($CDC_{13}$, 400 MHz). Expansion in the area from 7.72 – 7.60 ppm.

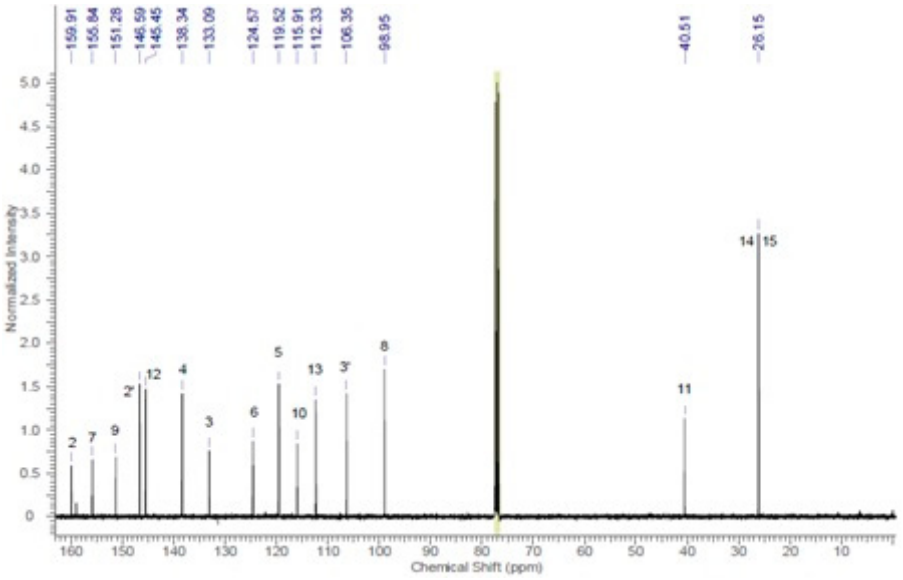


**Figure S1.3.** $^{13}$C-NMR spectrum of CHL ($CDC_{l3}$, 400 MHz).

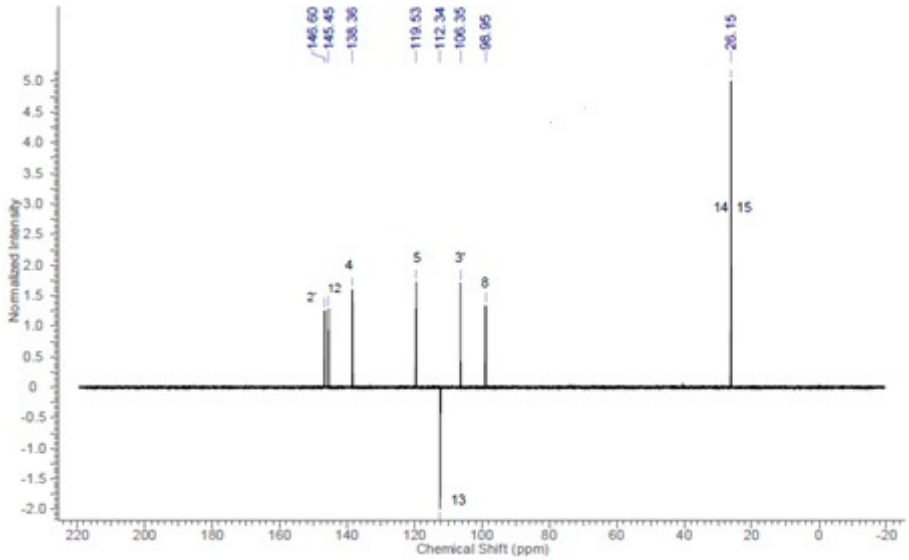


**Figure S1.4.** DEPT-135 spectrum of CHL ($CDC_{l3}$, 100 MHz).

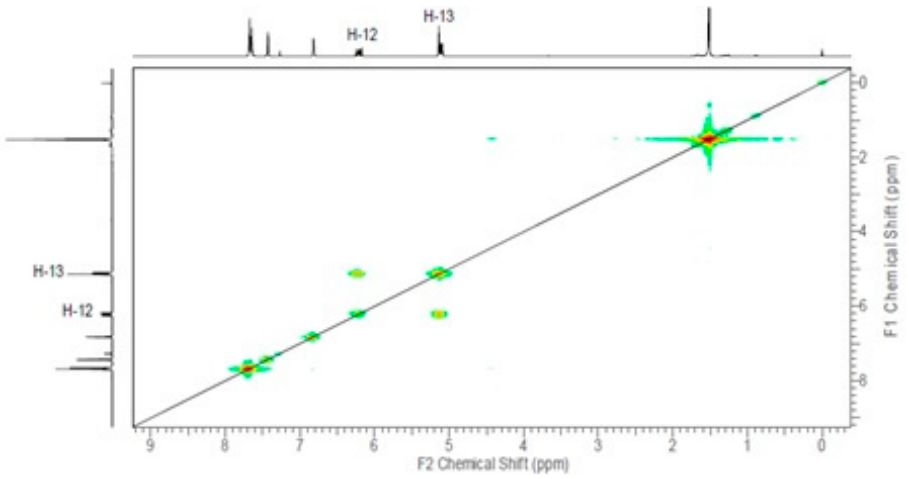


**Figure S1.5.** $^{1}$H-$^{1}$H COSY spectrum of CHL ($CDC_{13}$, 400 MHz).

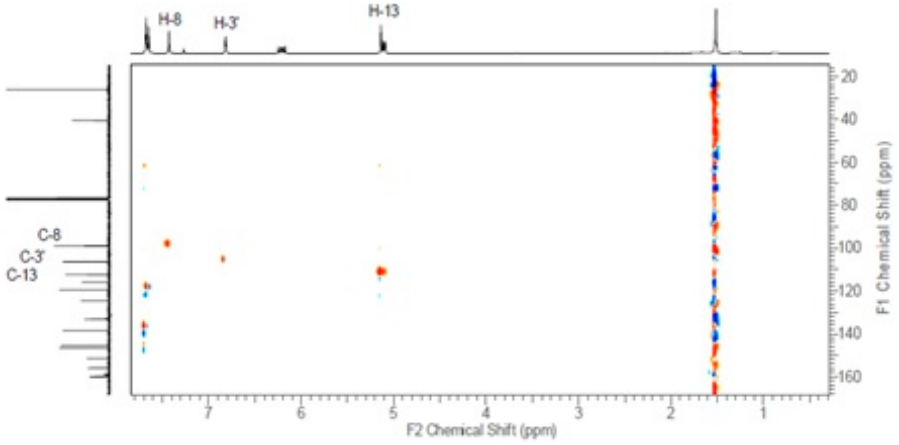


**Figure S1.6.1.** $^{1}$H-$^{13}$C-HSQC spectrum of CHL ($CDC_{13}$, 100 MHz).

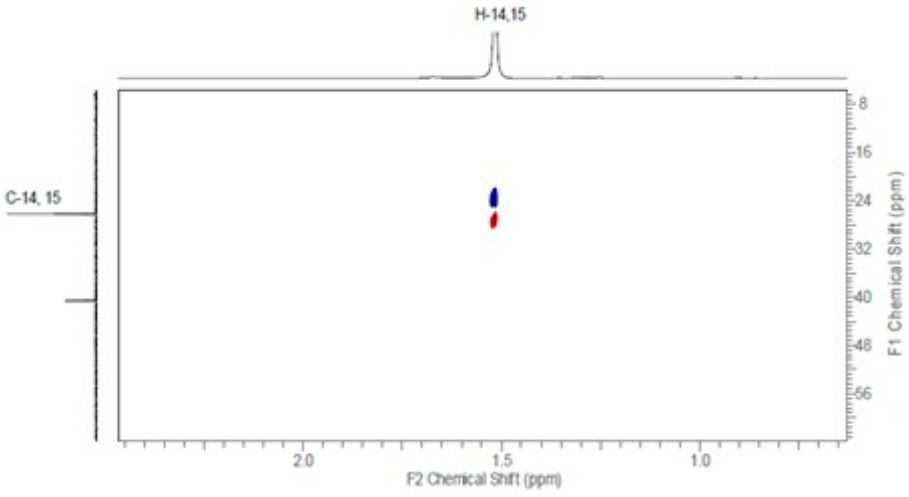


**Figure S1.6.2.** $^{1}$H-$^{13}$C-HSQC spectrum of CHL ($CDC_{13}$, 100 MHz). Expansion of the area$^{1}$H: 0.5 - 2.7 ppm, $^{13}$C: 6.0 - 64 ppm.

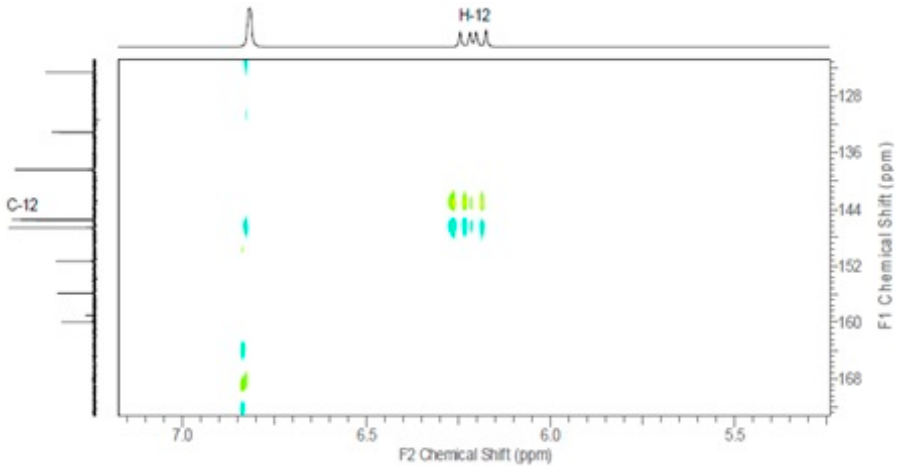


**Figure S1.6.3.** $^{1}$H-$^{13}$C-HSQC spectrum of CHL ($CDC_{l3}$, 100 MHz). Expansion of the area $^{1}$H: 5.3 - 7.1 ppm, $^{13}$C: 123 - 172 ppm.

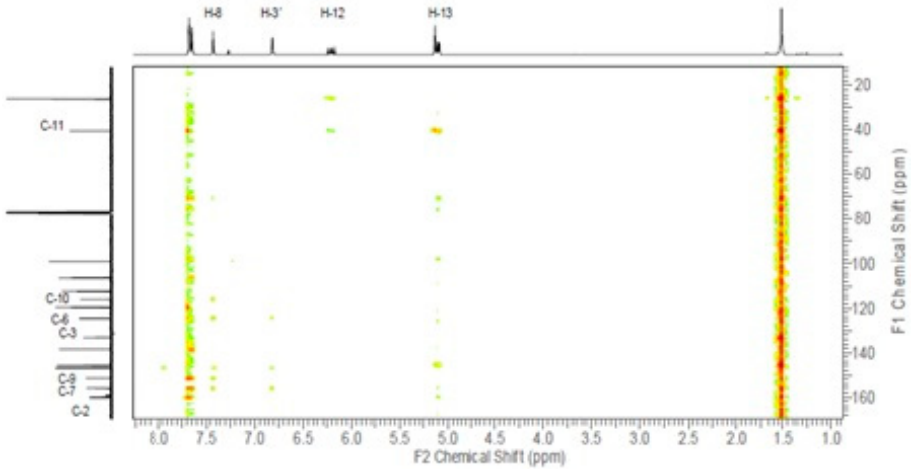


**Figure S1.7.** $^{1}$H-$^{13}$C-HMBC spectrum of CHL ($CDC_{l3}$, 100 MHz).

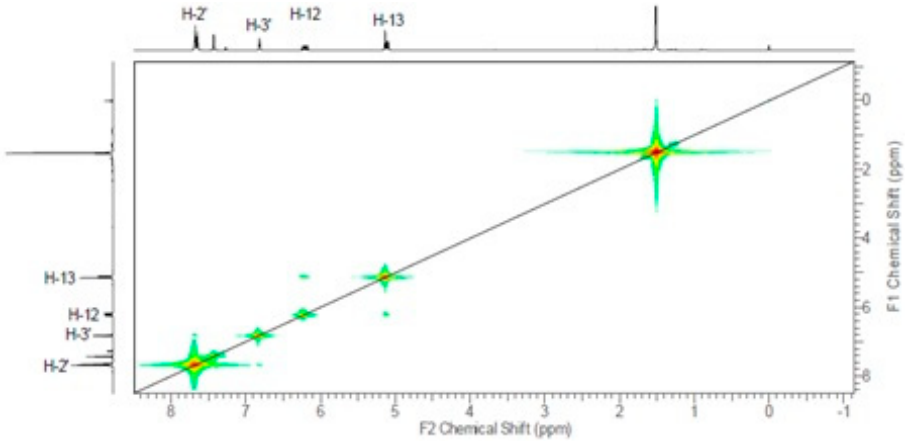


**Figure S1.8.** $^{1}$H-$^{1}$H-NOESY spectrum of CHL ($CDC_{l3}$, 400 MHz).

***Rutamarin (RTM)***

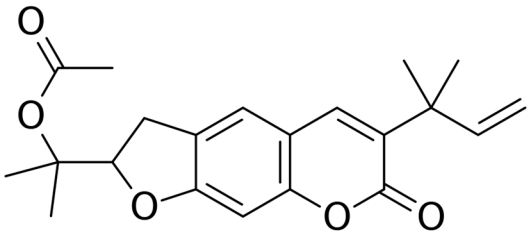

**Figure S2.1.** Structure of rutamarin (RTM). PubChem CID = 26948 (https://pubchem.ncbi.nlm.nih.gov/compound/26948).

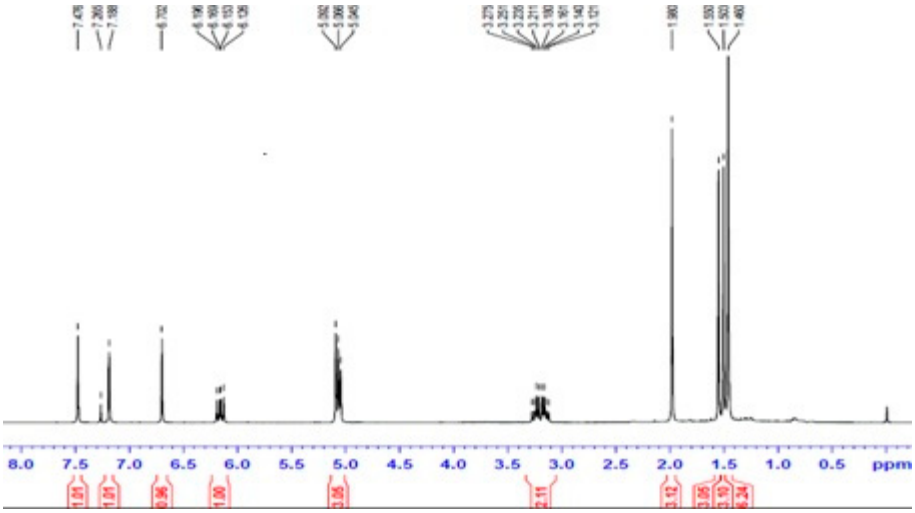


**Figure S2.2.1.** $^{1}$H-NMR spectrum of RTM ($CDCl_3$, 400 MHz).

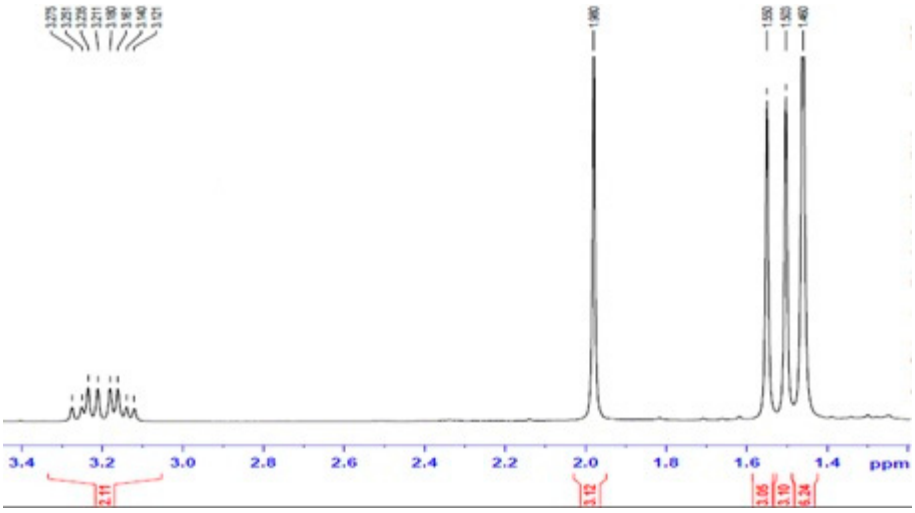


**Figure S2.2.2.** $^{1}$H-NMR spectrum of RTM ($CDCl_3$, 400 MHz). Expansion in the area from 1.2 - 3.4 ppm.

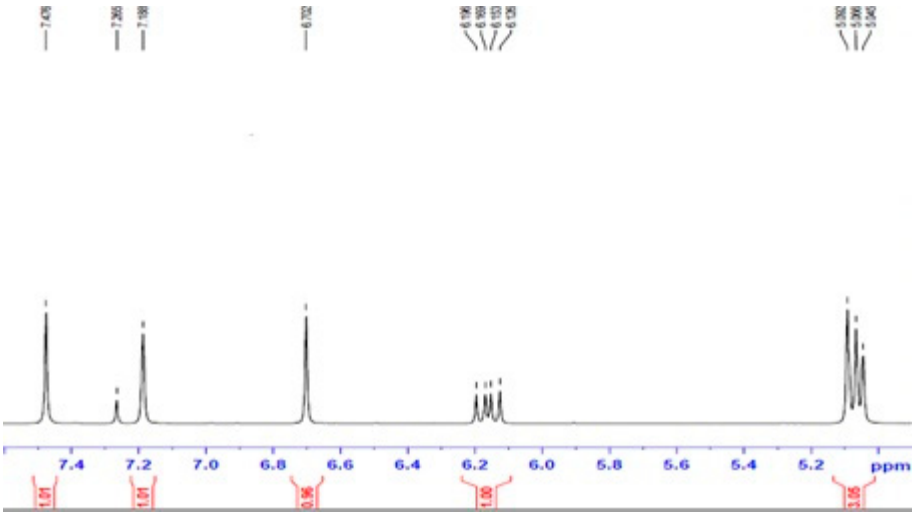


**Figure S2.2.3.** $^{1}$H-NMR spectrum of RTM ($CDCl_3$, 400 MHz). Expansion in the area from 4.8 - 7.6 ppm.

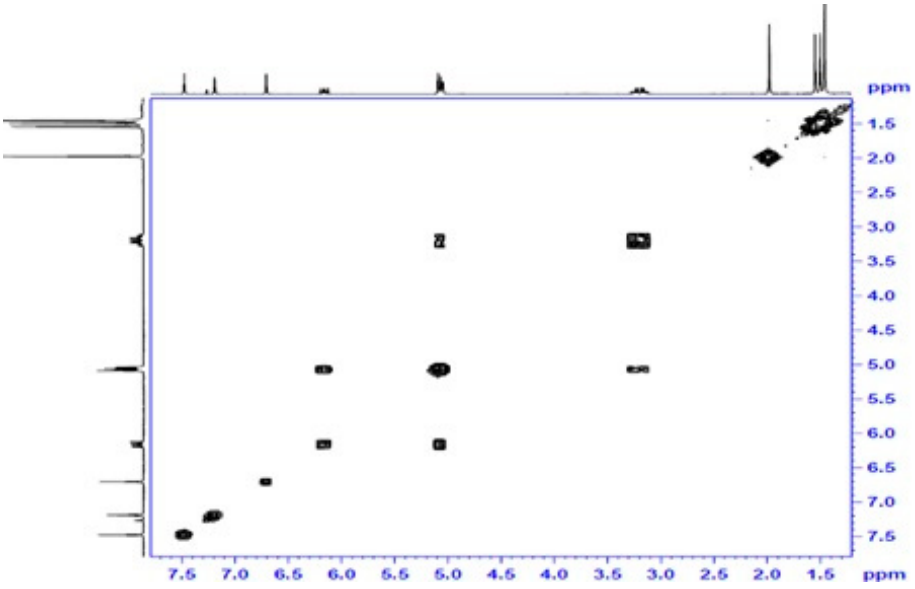


**Figure S2.3.** $^{1}$H-$^{1}$H COSY spectrum of RTM ($CDCl_3$, 400 MHz).

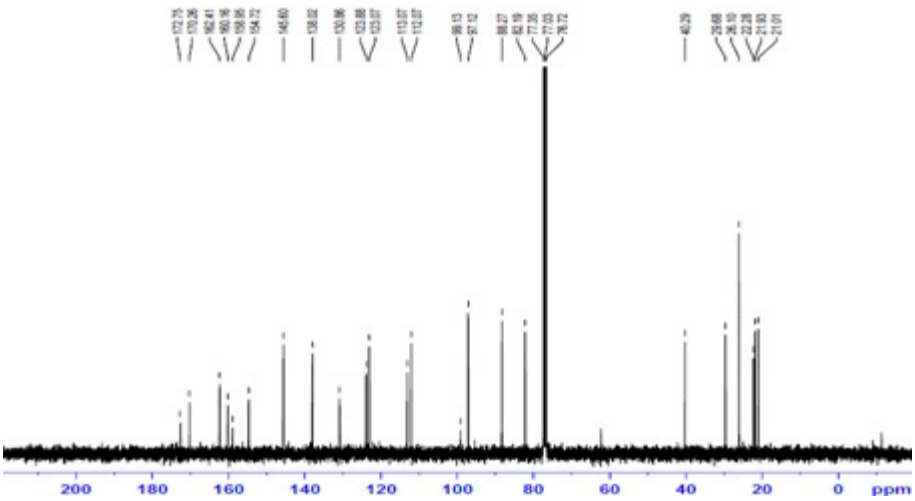


**Figure S2.4.1.** $^{13}$C-NMR spectrum of RTM ($CDCl_3$, 100 MHz).

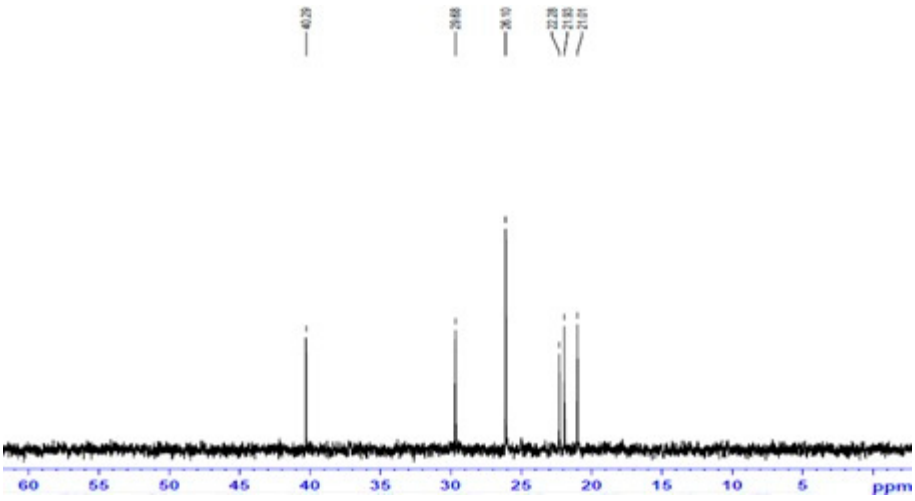


**Figure S2.4.2.** $^{13}$C-NMR spectrum of RTM ($CDCl_3$, 100 MHz). Expansion in the area from 0.5 - 60 ppm.

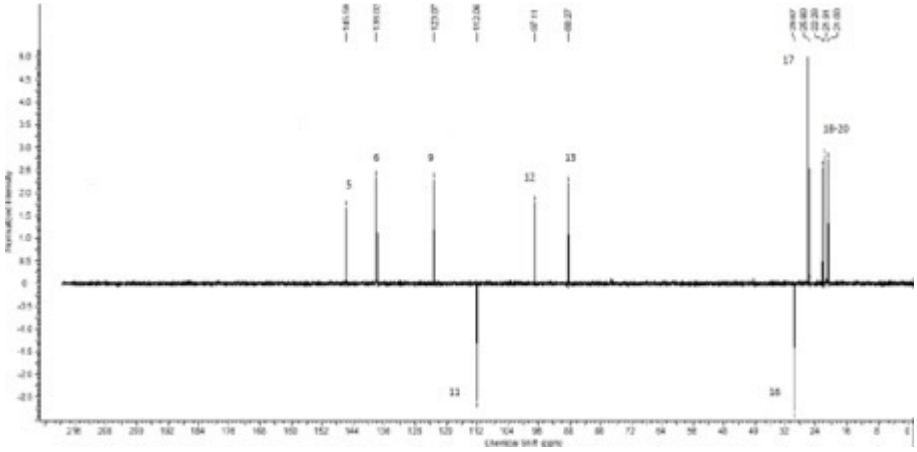


**Figure S2.5.** DEPT 135, $^{13}$C-NMR spectrum of RTM ($CDCl_3$, 100 MHz).

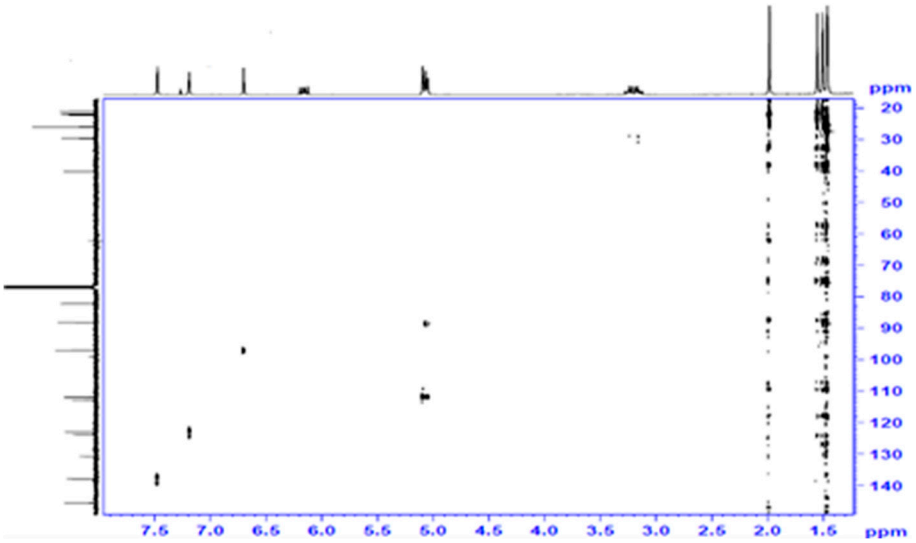


**Figure S2.5.1.** $^{1}H$-$^{13}C$-HSQC spectrum of RTM ($CDCl_3$, 400 MHz).

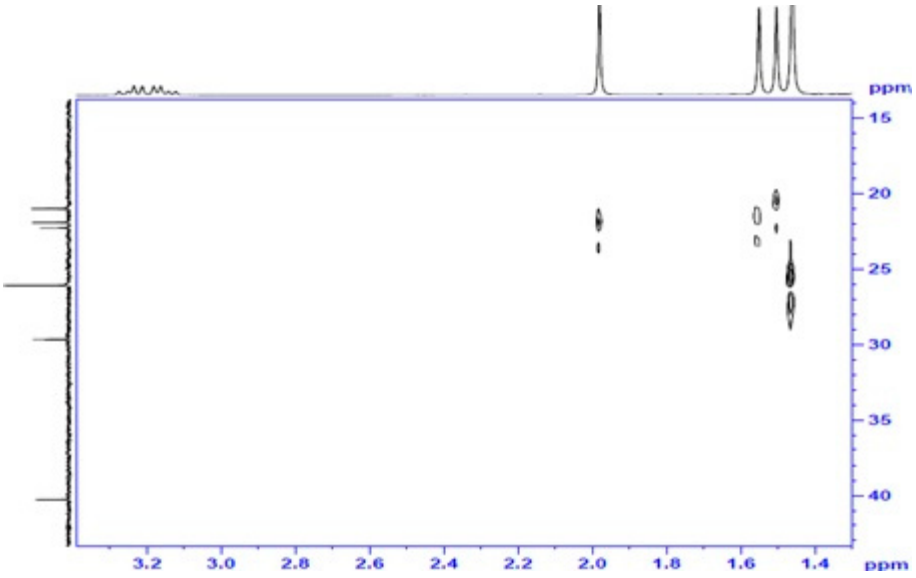


**Figure S2.5.2.** $^{1}H$-$^{13}C$-HSQC spectrum of RTM ($CDCl_3$, 400 MHz). Expansion of the area $^{1}H$: 1.3 - 3.4 ppm, $^{13}C$: 15 - 45 ppm.

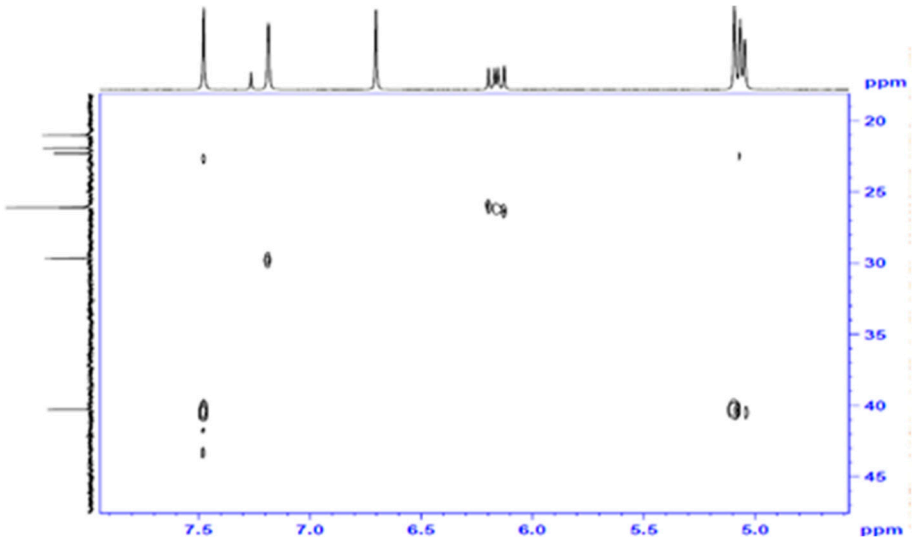


**Figure S2.5.3.** $^{1}H$-$^{13}C$-HSQC spectrum of RTM ($CDCl_3$, 400 MHz). Expansion of the area $^{1}H$: 4.6 - 8.0 ppm, $^{13}C$: 20 – 47 ppm.

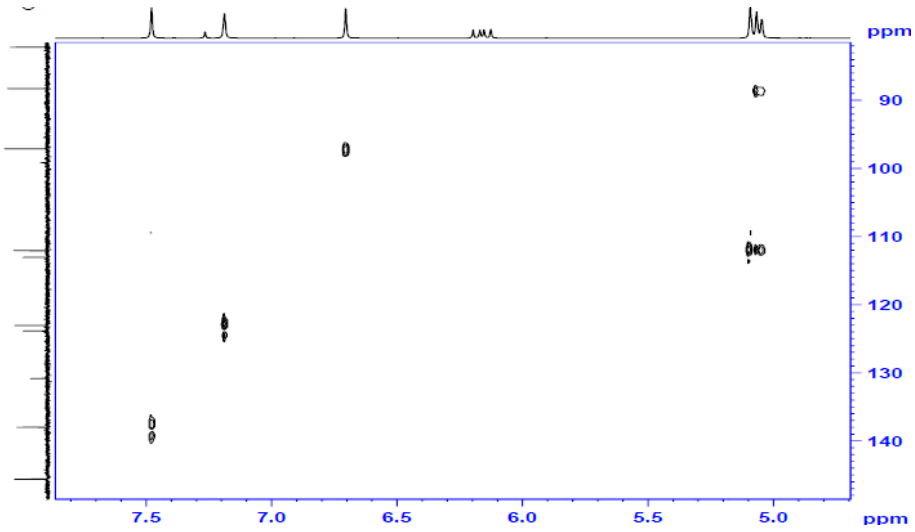


**Figure S2.5.4.** $^{1}H$-$^{13}C$-HSQC spectrum of RTM ($CDCl_3$, 400 MHz). Expansion of the area $^{1}H$: 4.6 - 8.0 ppm, $^{13}C$: 80 - 150 ppm.

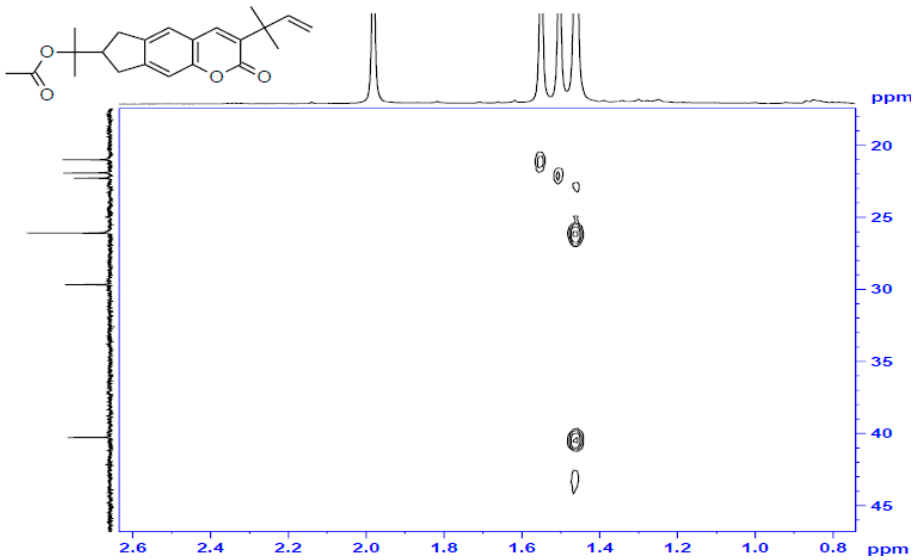


**Figure S2.6.1.** $^{1}$H-$^{13}$C-HMBC spectrum of RTM ($CDCl_3$, 400 MHz). Expansion of the area $^{1}$H: 0.8 - 2.6 ppm, $^{13}$C: 18 - 46 ppm.

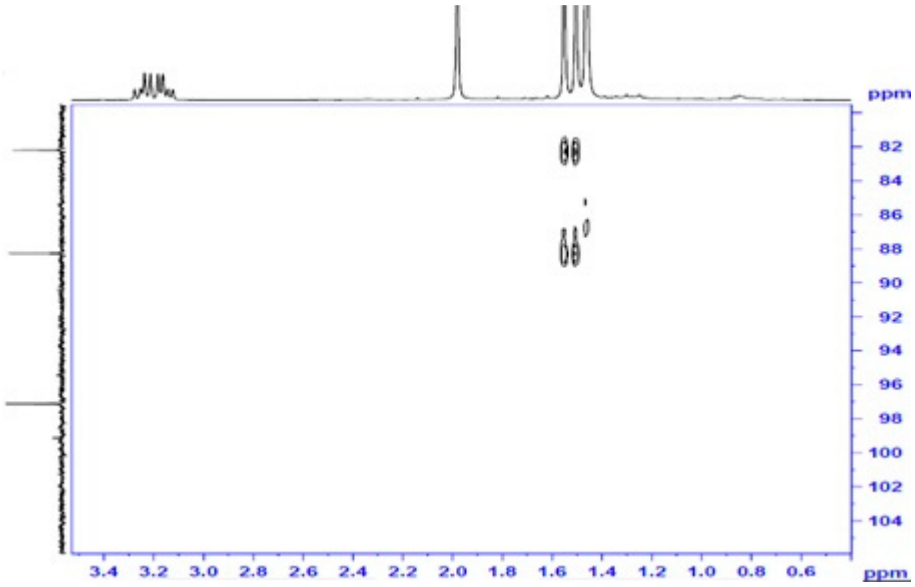


**Figure S2.6.2.** $^{1}$H-$^{13}$C-HMBC spectrum of RTM ($CDCl_3$, 400 MHz). Expansion of the area $^{1}$H: 0.4 - 3.5 ppm, $^{13}$C: 80 - 106 ppm.

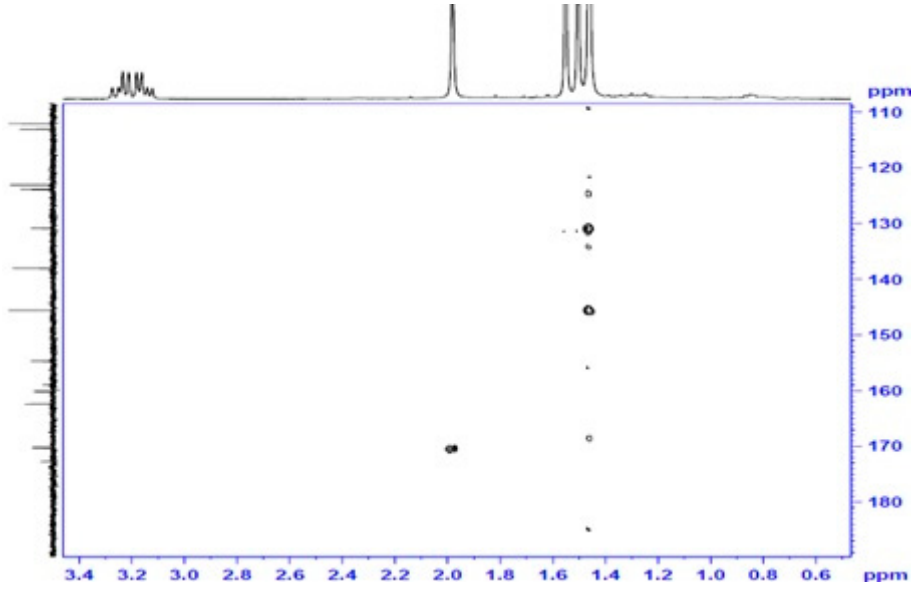


**Figure S2.6.3.** $^{1}H$-$^{13}C$-HMBC spectrum of RTM ($CDCl_3$, 400 MHz). Expansion of the area $^{1}H$: 0.4 - 3.4 ppm, $^{13}C$: 109 - 190 ppm.

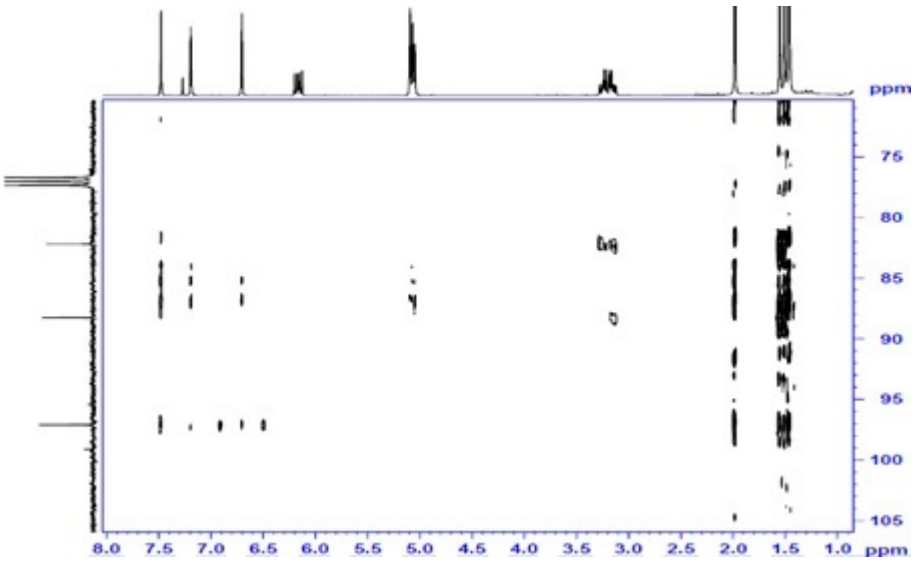


**Figure S2.6.4.** $^{1}H$-$^{13}C$-HMBC spectrum of RTM ($CDCl_3$, 400 MHz). Expansion of the area $^{1}H$: 0.8 – 8.0 ppm, $^{13}C$: 70 - 106 ppm.

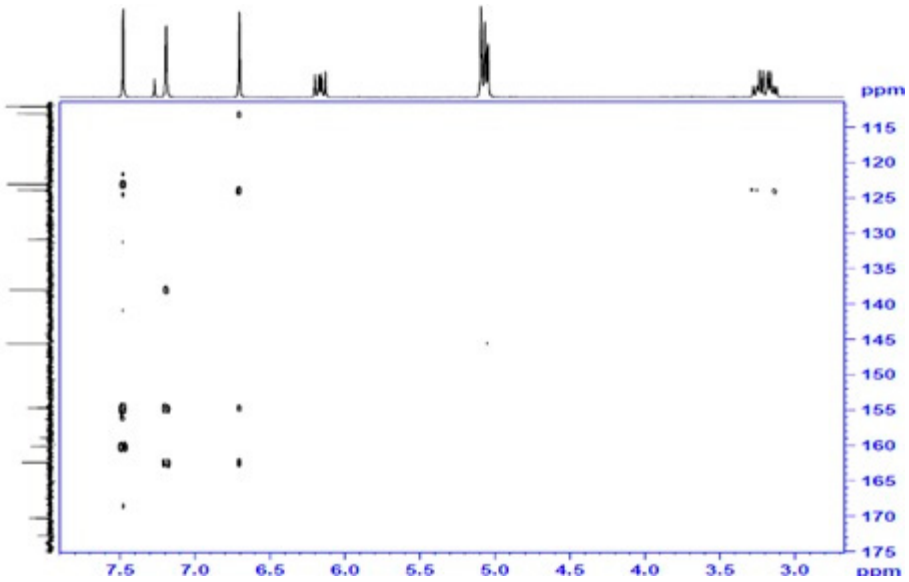


**Figure S2.6.5.** $^{1}$H-$^{13}$C-HMBC spectrum of RTM ($CDCl_3$, 400 MHz). Expansion of the area $^{1}$H: 2.5 – 8.0 ppm, $^{13}$C: 110 - 175 ppm.

***Graveolin (GRV)***

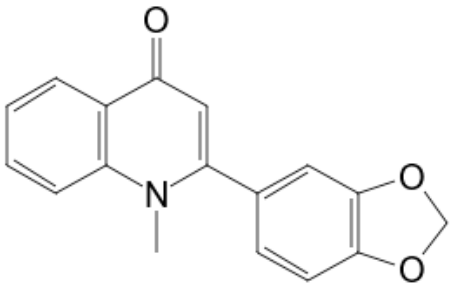


**Figure S3.1.** Structure of graveoline (GRV). PubChem CID = 353825 (https://pubchem.ncbi.nlm.nih.gov/compound/353825).

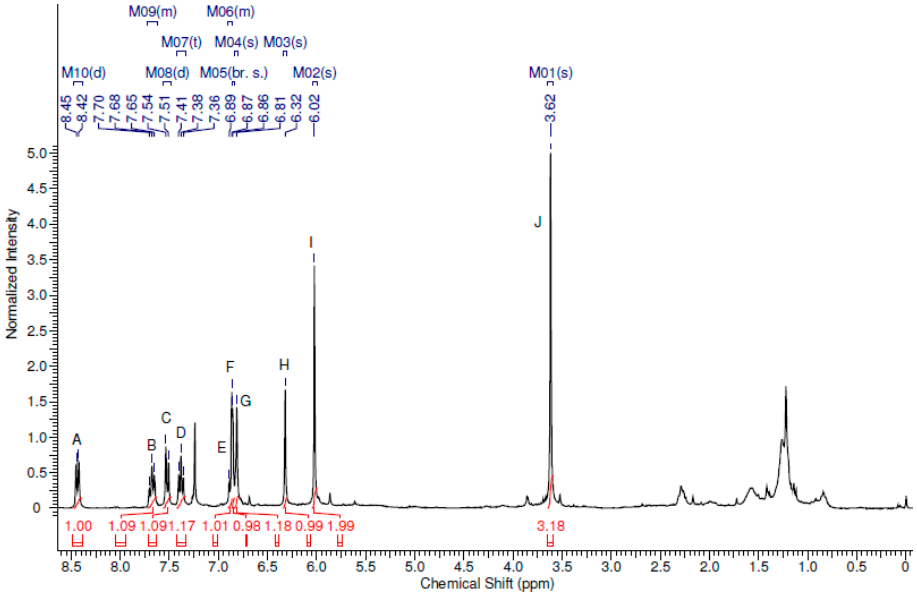


**Figure S3.2.1.** $^{1}$H-NMR spectrum of GRV ($CDCl_3$, 400 MHz).

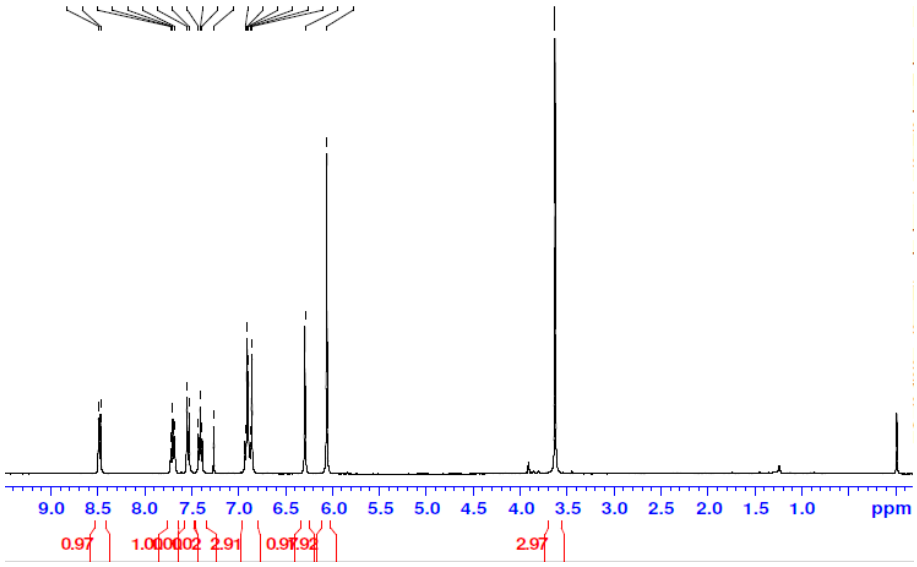


**Figure S3.2.2.** $^{1}$H-NMR general spectrum of GRV ($CDCl_3$, 400 MHz).

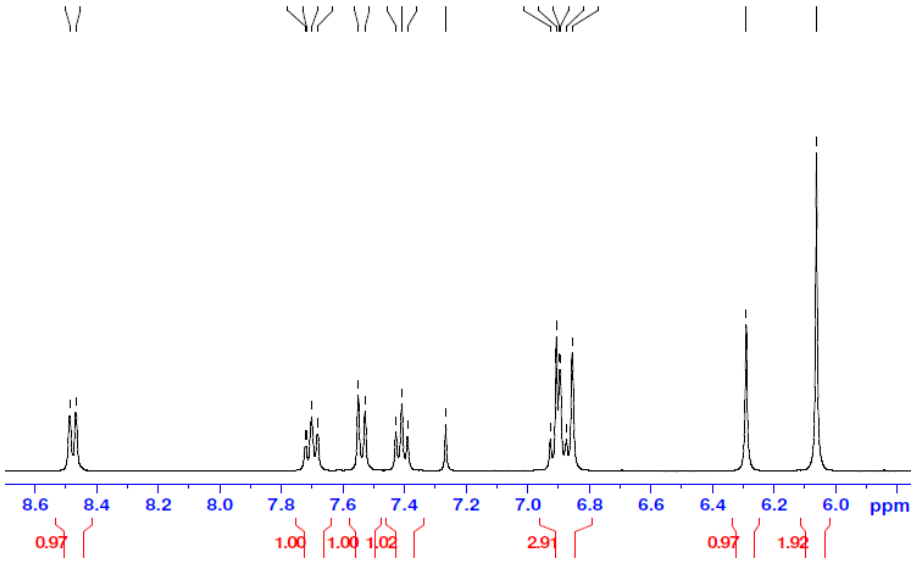


**Figure S3.2.3.** $^{1}$H-NMR spectrum of GRV ($CDCl_3$, 400 MHz). Expansion of the area $^{1}$H: 5.7 – 8.7 ppm.

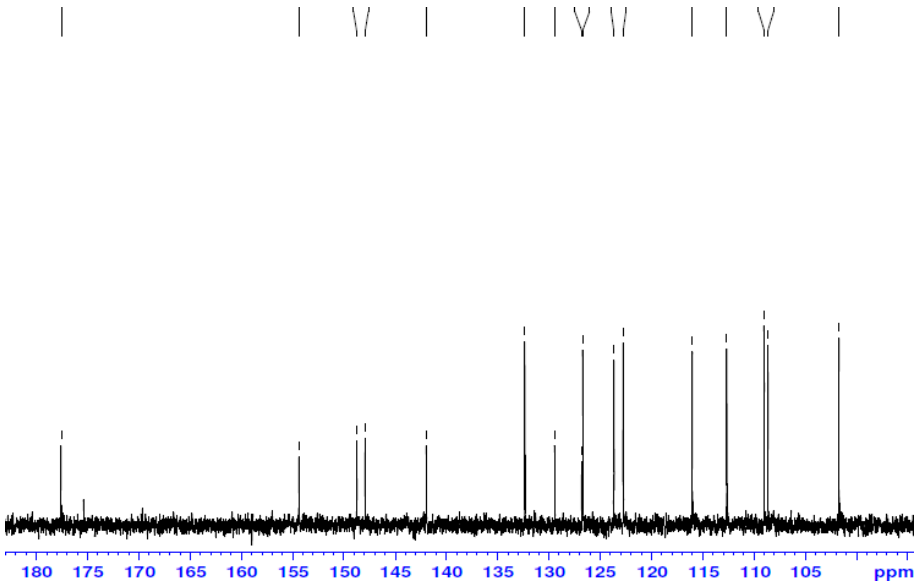


**Figure S3.2.4.** $^{1}$H-NMR spectrum of GRV ($CDCl_3$, 100 MHz). Expansion of the area $^{1}$H: 94 – 183 ppm.

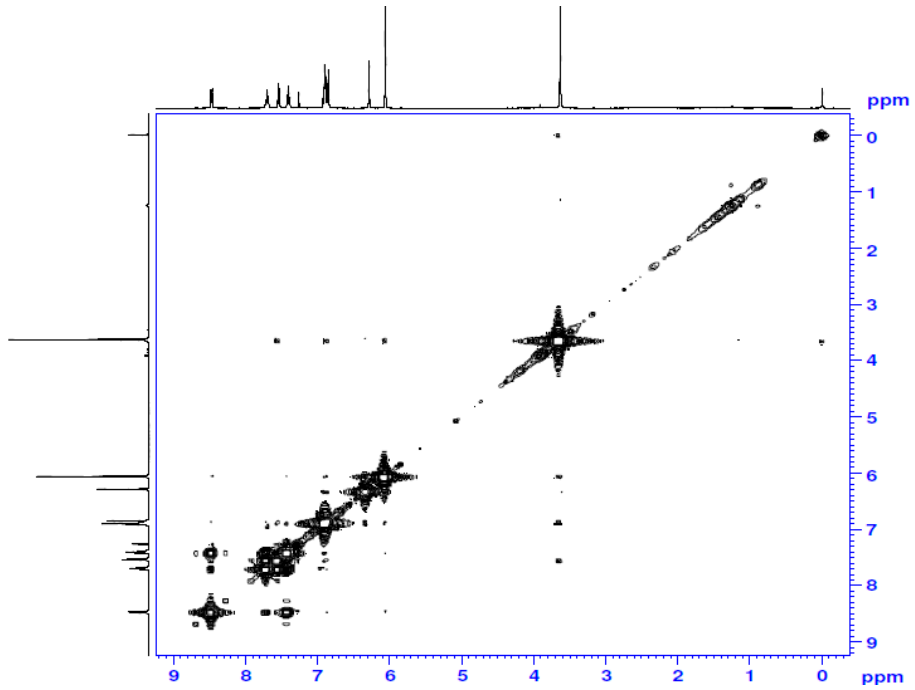


**Figure S3.3.1.** $^{1}$H-$^{1}$H COSY general spectrum of GRV ($CDCl_3$, 400 MHz).

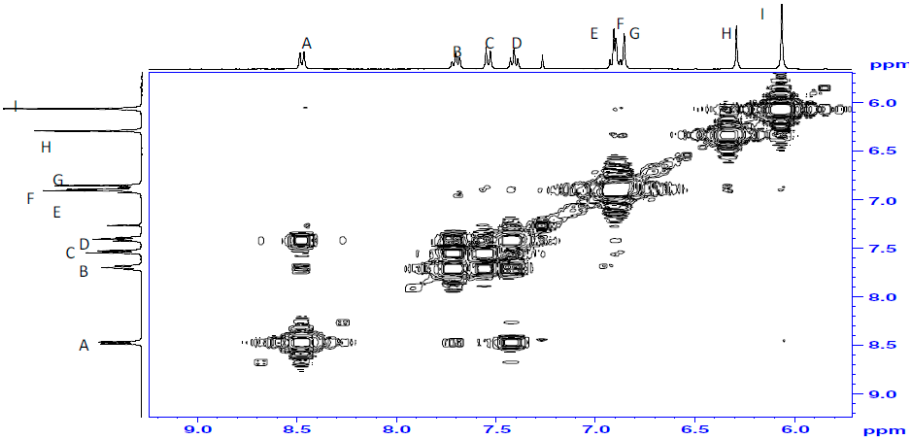


**Figure S3.3.2.** $^{1}$H-$^{1}$H COSY spectrum of GRV ($CDCl_3$, 400 MHz). Expansion of the area $^{1}$H: 5.7 – 9.2 ppm, $^{1}$H: 5.7 – 9.2.

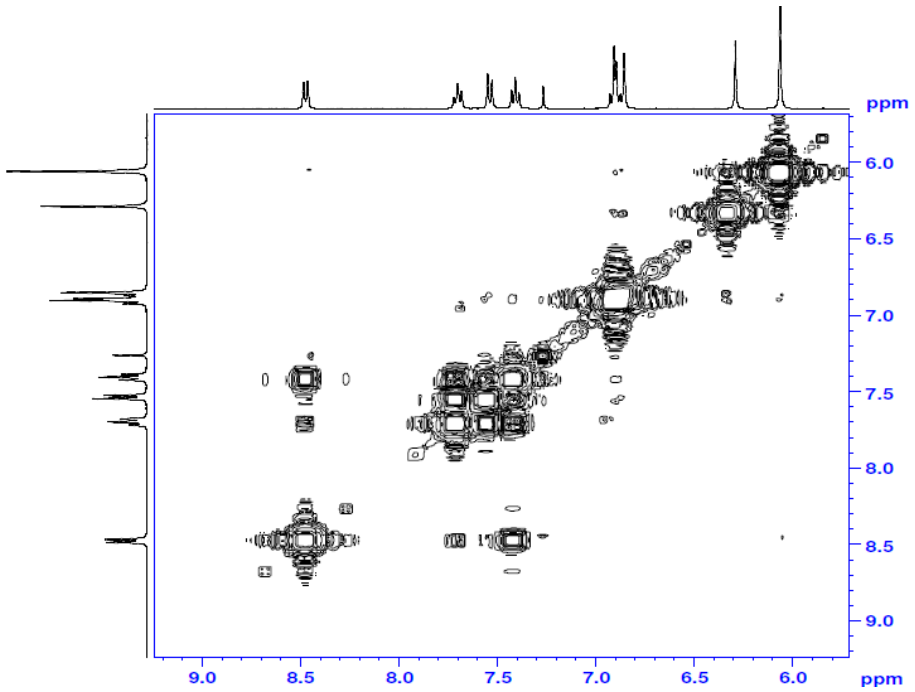


**Figure S3.3.3.** $^{1}$H-$^{1}$H COSY spectrum of GRV ($CDCl_3$, 400 MHz). Expansion of the area $^{1}$H: 5.7 – 9.2 ppm, $^{1}$H: 5.7 – 9.2.

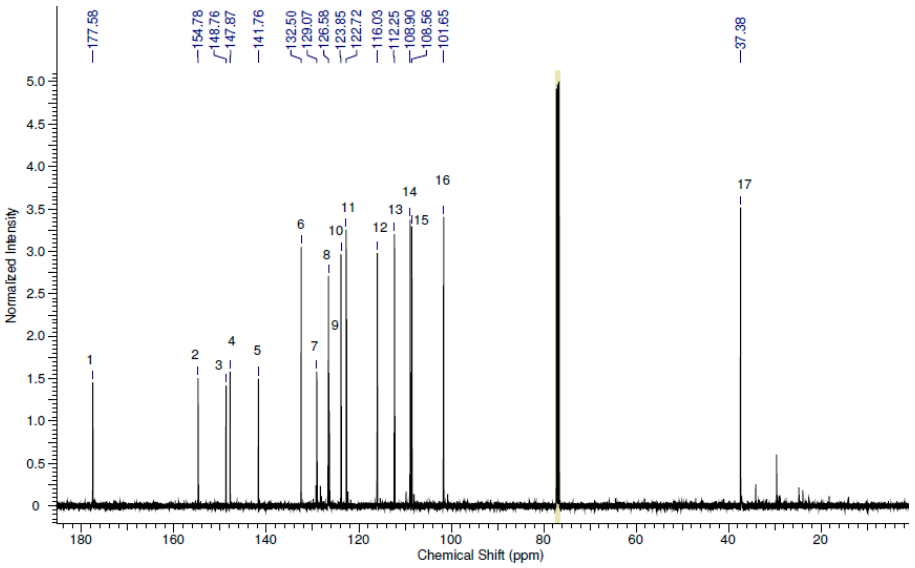


**Figure S3.4.1.** $^{13}$C-NMR spectrum of GRV ($CDCl_3$, 100 MHz).

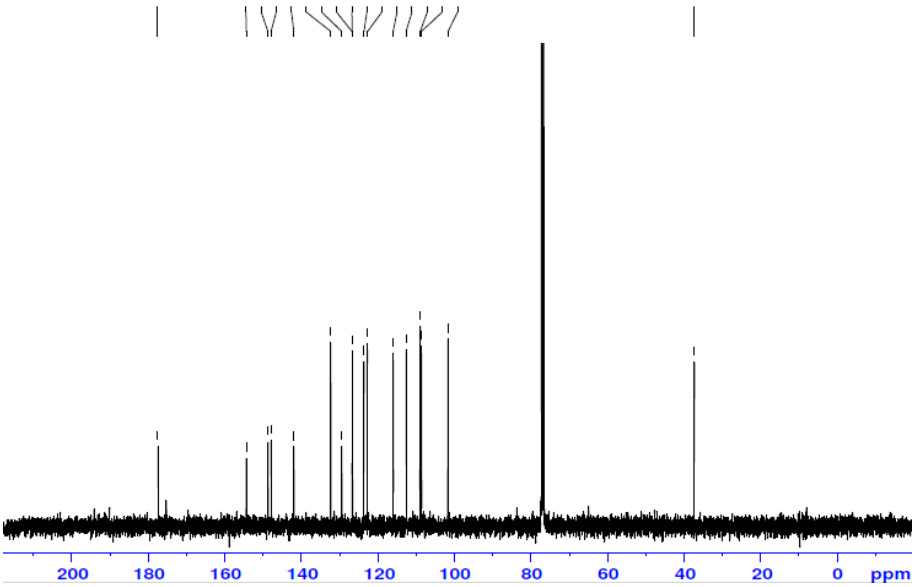


**Figure S3.4.2.** $^{13}$C-NMR spectrum of GRV ($CDCl_3$, 100 MHz).

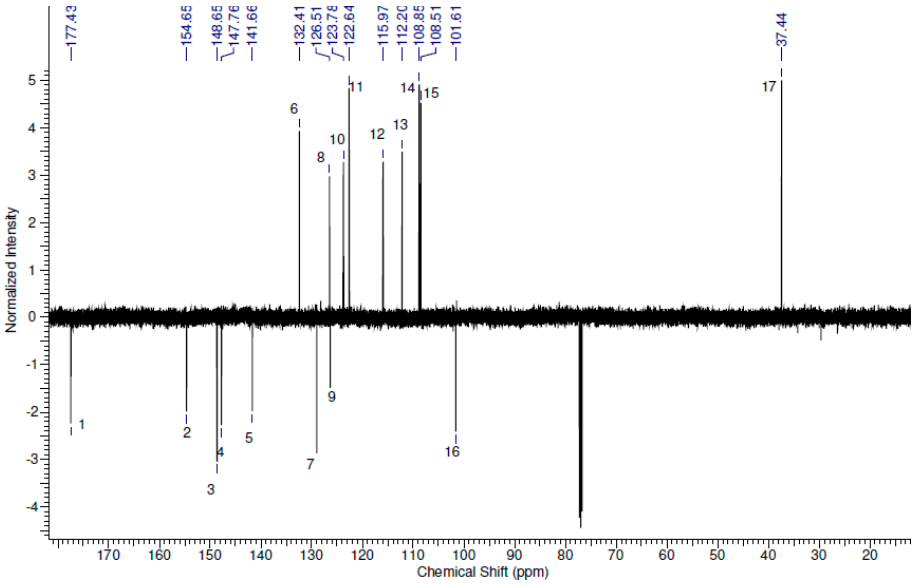


**Figure S3.5.** APT, $^{13}$C-NMR spectrum of GRV ($CDCl_3$, 400 MHz).

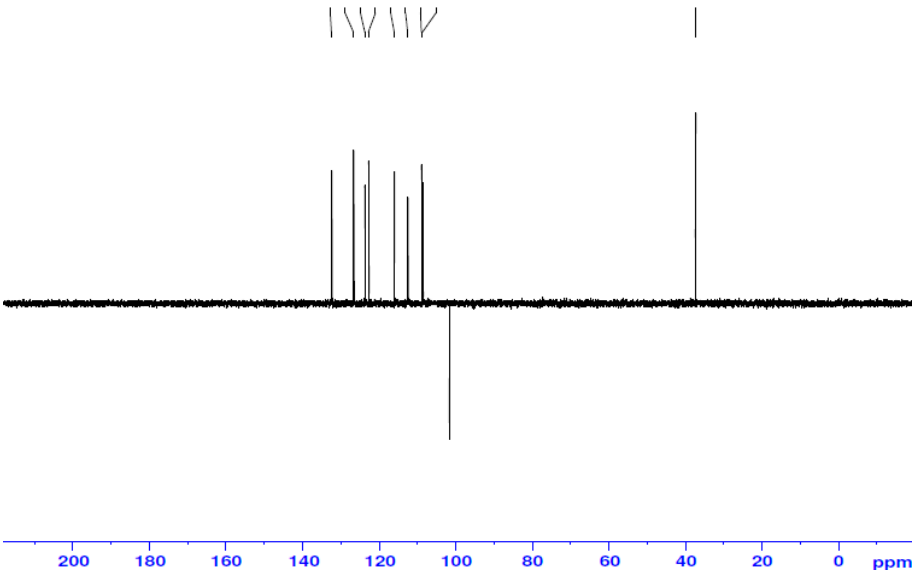


**Figure S3.6.** DEPT 135, $^{13}$C-NMR spectrum of GRV ($CDCl_3$, 100 MHz).

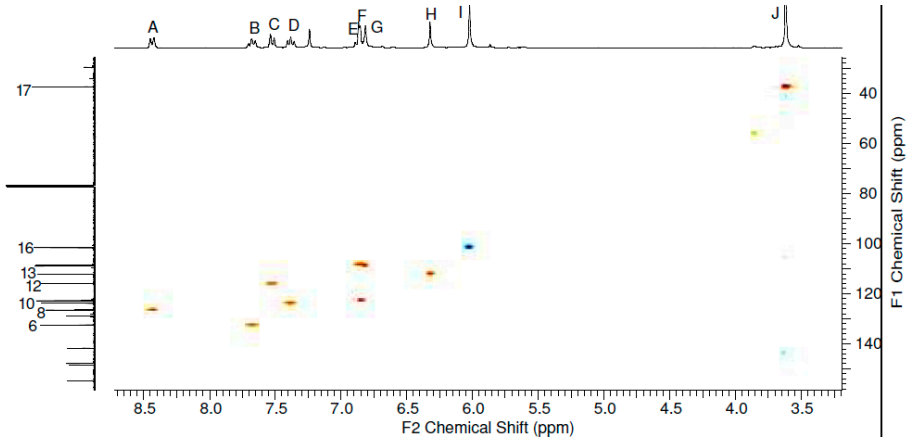


**Figure S3.7.1.** $^{1}$H-$^{13}$C-HSQC spectrum of GRV ($CDCl_3$, 400 MHz).

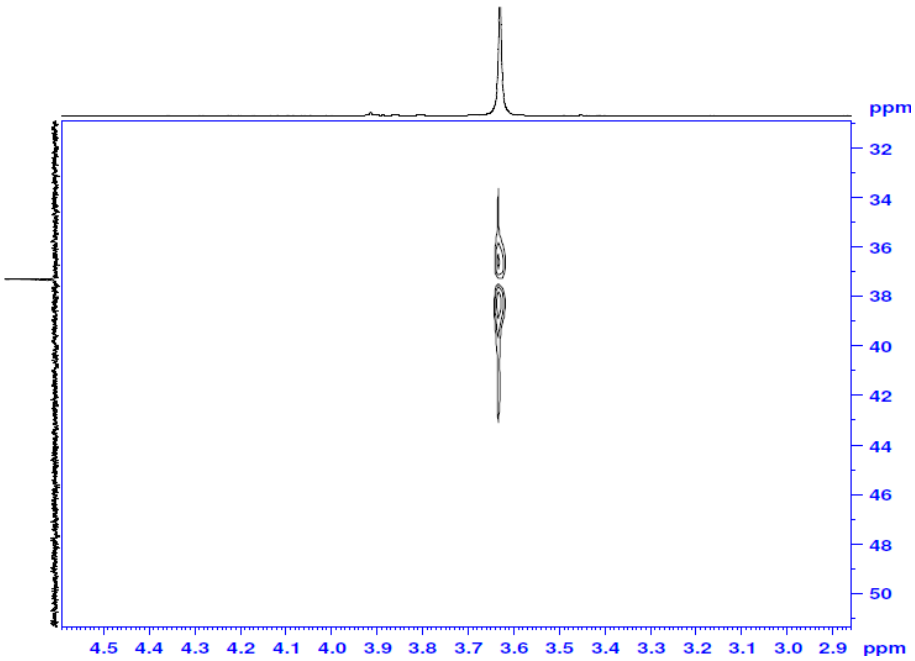


**Figure S3.7.2.** $^{1}$H-$^{13}$C-HSQC spectrum of GRV ($CDCl_3$, 400 MHz). Expansion of the area $^{1}$H: 2.9 - 4.6 ppm, $^{13}$C: 31 - 51.

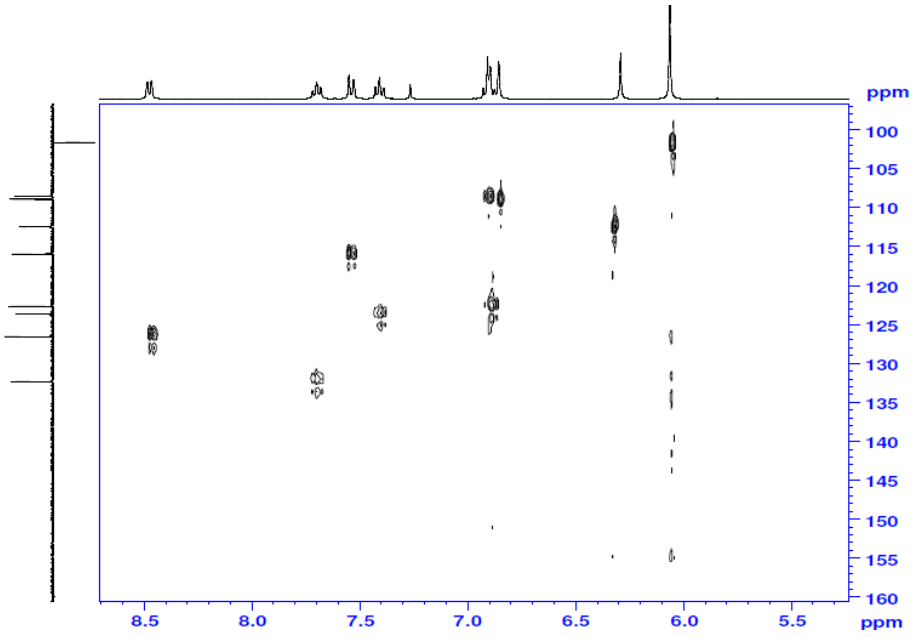


**Figure S3.7.3.** $^{1}$H-$^{13}$C-HSQC spectrum of GRV ($CDCl_3$, 400 MHz). Expansion of the area $^{1}$H: 5.3 - 8.7 ppm, $^{13}$C: 95 - 160.

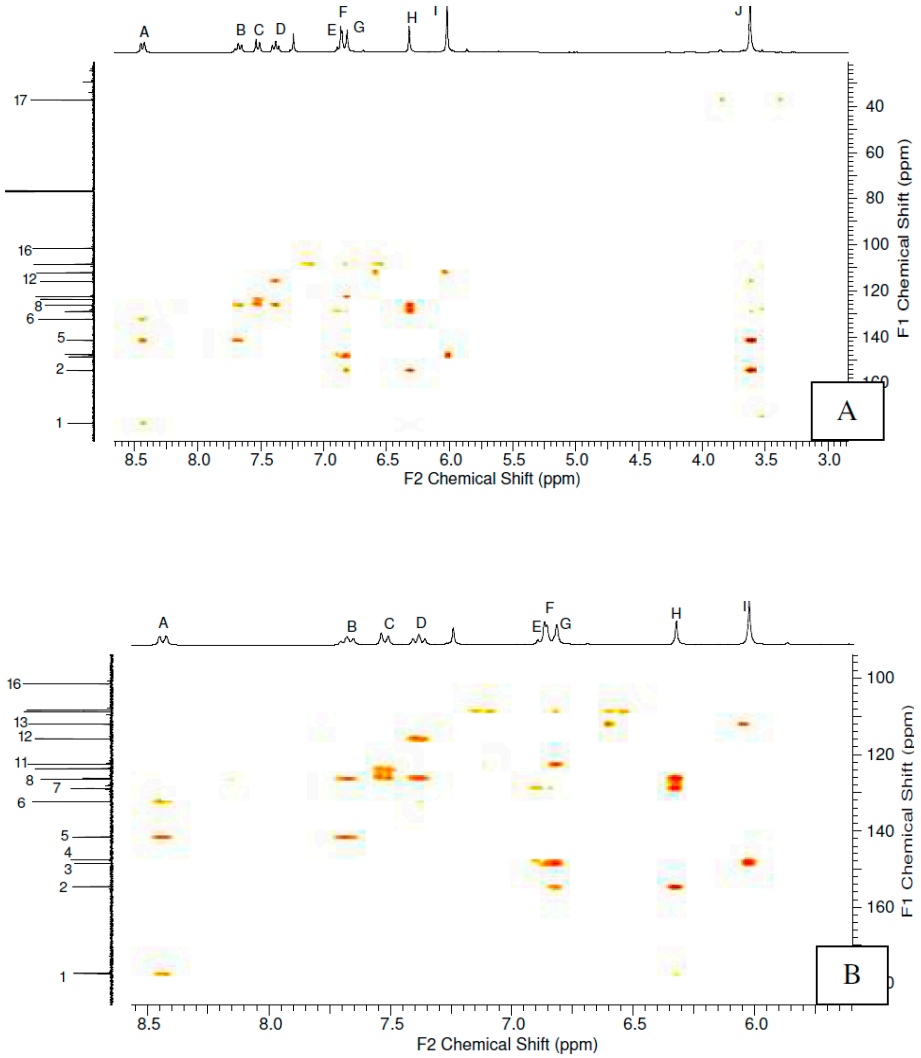


**Figure S3.8.1.** $^{1}H$-$^{13}C$-HMBC spectrum of GRV ($CDCl_3$, 400 MHz). A: General spectrum.; B: Expansion of the area $^{1}H$: 5.0 - 8.8 ppm, $^{13}C$: 90 - 185 ppm.

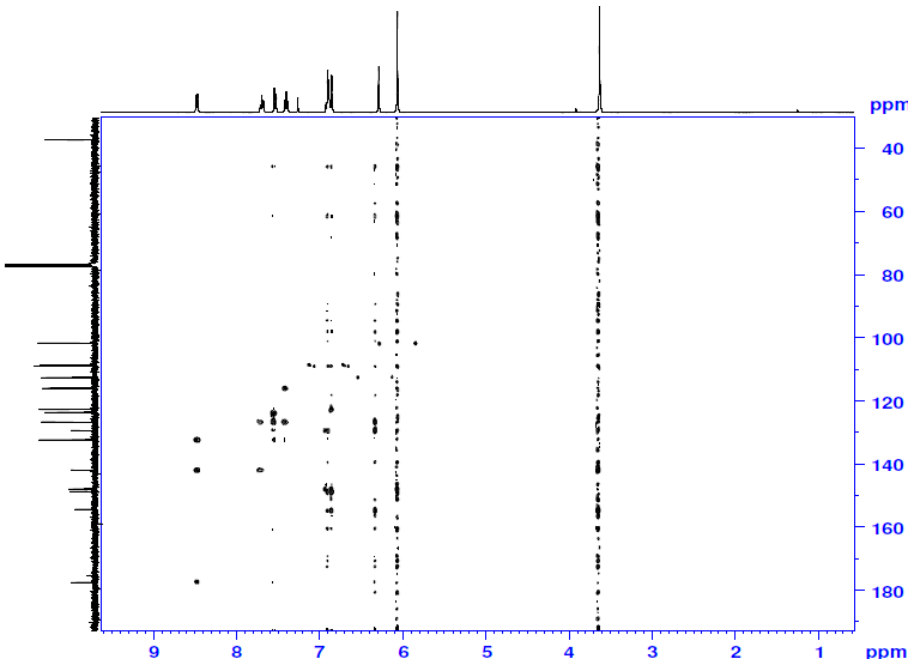


**Figure S3.8.2.** $^{1}H$-$^{13}C$-HMBC general spectrum of GRV ($CDCl_3$, 400 MHz).

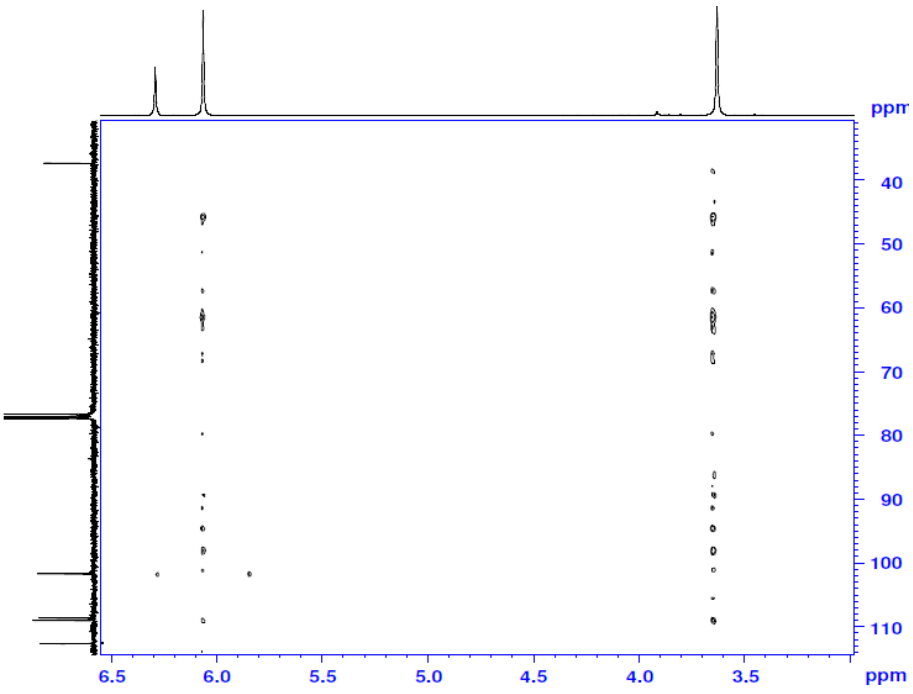


**Figure S3.8.3.** $^{1}H$-$^{13}C$-HMBC spectrum of GRV ($CDCl_3$, 400 MHz). Expansion of the area $^{1}H$: 3.0 - 6.6 ppm, $^{13}C$: 30 - 115 ppm.

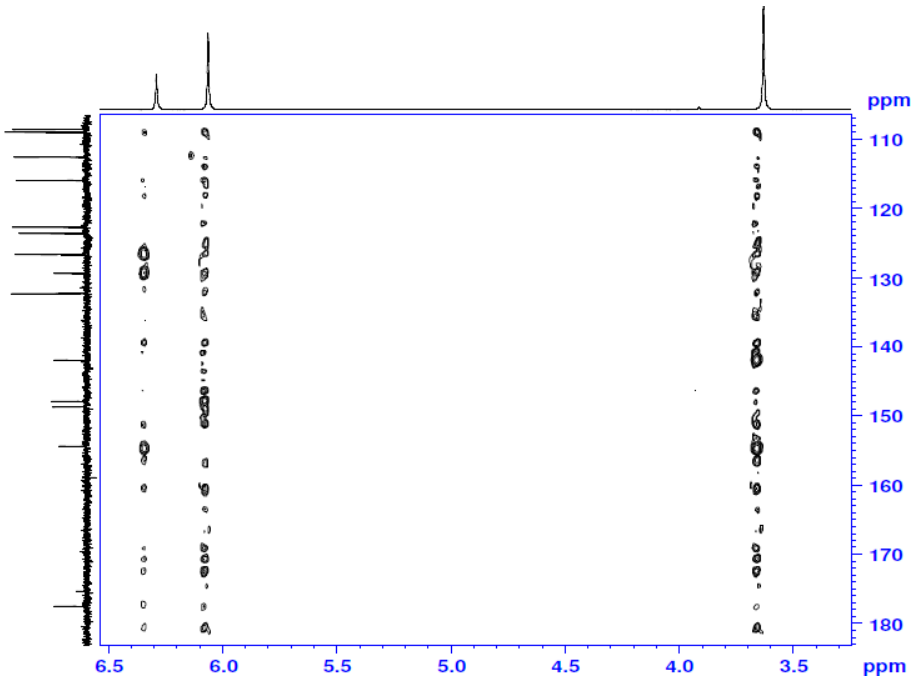


**Figure S3.8.4.** $^{1}H$-$^{13}C$-HMBC spectrum of GRV ($CDCl_3$, 400 MHz). Expansion of the area $^{1}H$: 3.0 - 6.6 ppm, $^{13}C$: 107 - 183 ppm.

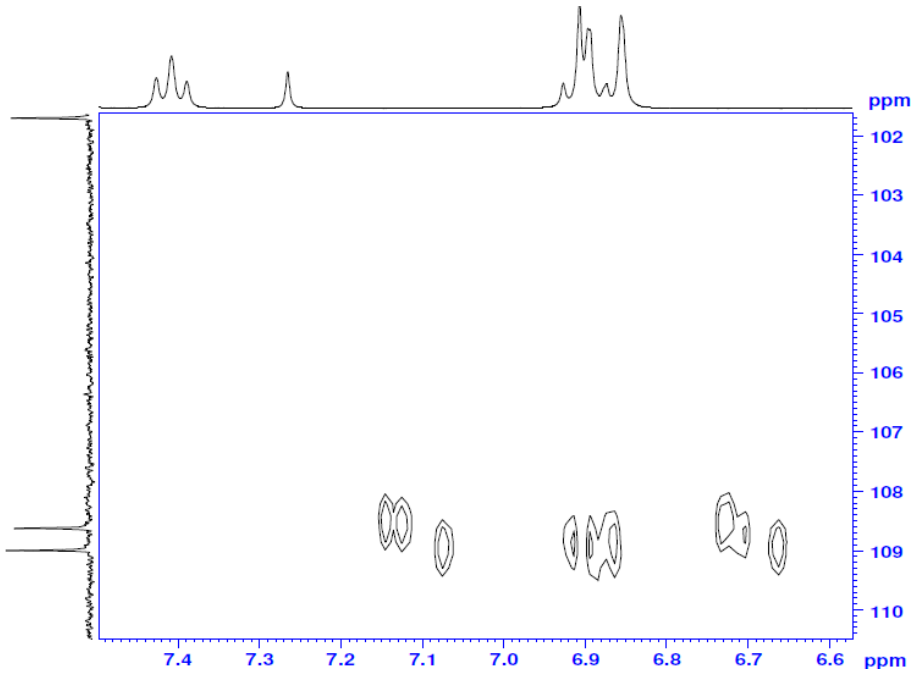


**Figure S3.8.5.** $^{1}H$-$^{13}C$-HMBC spectrum of GRV ($CDCl_3$, 400 MHz). Expansion of the area $^{1}H$: 6.4 - 7.5 ppm, $^{13}C$: 99 - 115 ppm.

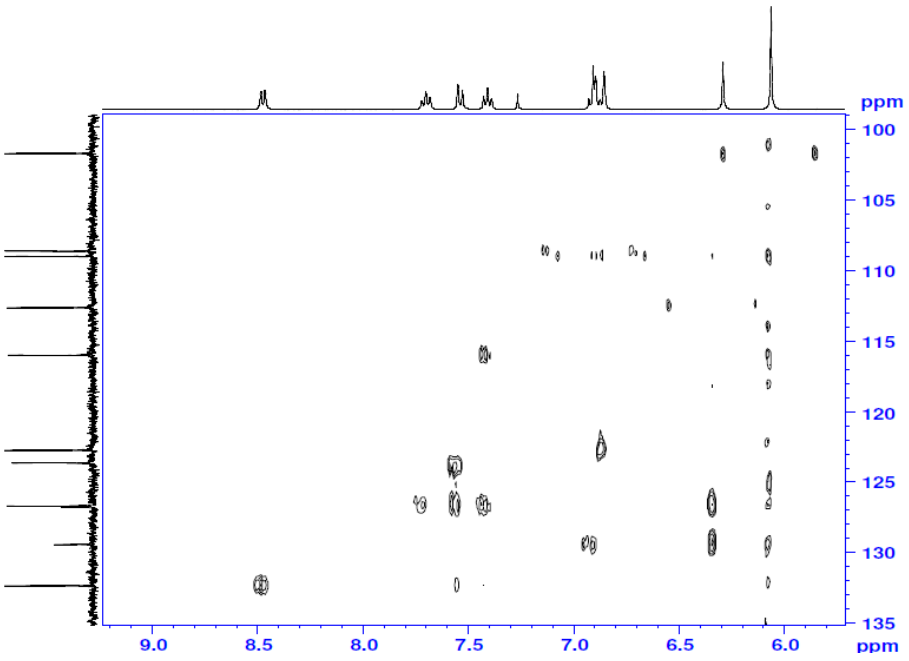


**Figure S3.8.6.** $^{1}$H-$^{13}$C-HMBC spectrum of GRV ($CDCl_3$, 400 MHz). Expansion of the area $^{1}$H: 5.7 – 9.2 ppm, $^{13}$C: 99 - 135 ppm.

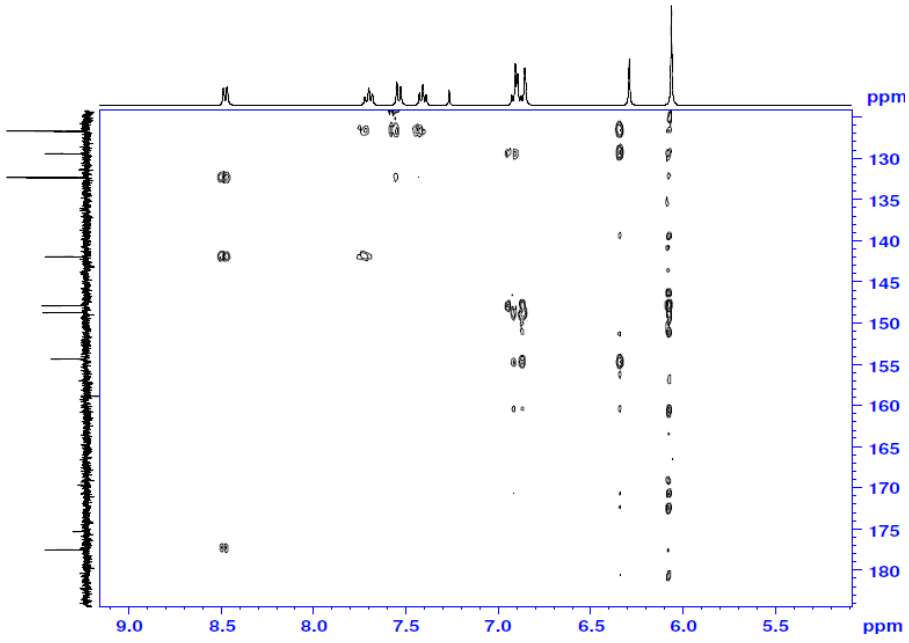


**Figure S3.8.7.** $^{1}$H-$^{13}$C-HMBC spectrum of GRV ($CDCl_3$, 400 MHz). Expansion of the area $^{1}$H: 5.1 – 9.1 ppm, $^{13}$C: 124 - 185 ppm.

**Data Availability Statement:** The datasets generated or analyzed during the present study are available from the corresponding authors.

**Conflicts of Interest:** The authors declare no conflict of interest.

**References**

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