

Electronic Supplementary Material

Article Title: Structure elucidation of the novel carotenoid gemmatoxanthin from the photosynthetic complexes of *Gemmatimonas phototrophica* AP64

Authors: Nupur¹, Marek Kuzma², Jan Hájek^{1,3}, Pavel Hrouzek^{1,3}, Alastair T. Gardiner¹, Martin Lukeš¹, Martin Moos⁴, Petr Šimek⁴, Michal Koblížek^{1*}

Affiliation: ¹Centre Algatech, Institute of Microbiology, The Czech Academy of Sciences (CAS), 379 81, Třebon, Czechia. ²Laboratory of Molecular Structure Characterization, Institute of Microbiology of the Czech Acad Sci, Prague, Czechia. ³University of South Bohemia, Faculty of Science, Branišovská 1760, České Budějovice, Czechia. ⁴ Biology Centre of the Czech Academy of Sciences, Branišovská 1160/31, České Budějovice 370 05, Czechia.

Corresponding Author: koblizek@alga.cz

	m/z	Summ formula	error		annotation
[M+H] ⁺	581.4023	C ₄₀ H ₅₂ O ₃ +H ⁺	-5.9ppm	methoxy- loss	X ₁ ⁺
[M+H] ⁺	563.3910	C ₄₀ H ₅₀ O ₂ +H ⁺	-4.7ppm	water loss	not explained
[M+H] ⁺	553.4113	C ₃₉ H ₅₂ O ₂ +H ⁺	-13.2ppm	methoxy- & formyl- loss	X ₁ ⁺ =>X ₃ ⁺
[M+H] ⁺	525.3387	C ₃₆ H ₄₄ O ₃ +H ⁺	-4.5ppm	terminal C ₅ H ₁₀ O loss	X ₂ ⁺
[M+H] ⁺	399.2691	C ₂₉ H ₃₄ O+H ⁺	-2.2ppm	not assigned	not assigned
[M+H] ⁺	359.2369	C ₂₆ H ₃₀ O+H ⁺	+0.2ppm	not assigned	not assigned
[M+H] ⁺	341.2288	C ₂₆ H ₂₈ +H ⁺	-7.2ppm	not assigned	not assigned

Table S1: Major HRMS-APCI fragments (positive mode)

	m/z	Summ formula	error		annotation
[M] ⁻	579.38459	C ₄₀ H ₅₀ O ₃ ⁻	0.4 ppm	methoxy- loss	X ₁ ⁻
[M] ⁻	561.37438	C ₄₀ H ₄₈ O ₂ ⁻	1.0 ppm	water loss	not explained
[M] ⁻	535.39335	C ₃₉ H ₅₀ O ⁻	-2.2 ppm	CO ₂ loss	X ₁ ⁻ =>X ₂ ⁻
[M] ⁻	445.3105	C ₃₁ H ₄₀ O ₂ ⁻	-1.7 ppm		X _{3zzzz} ⁻
[M] ⁻	287.2021	C ₁₉ H ₂₆ O ₂ ⁻	1.5 ppm		X ₄ ⁻
[M] ⁻	247.1700	C ₁₆ H ₂₂ O ₂ ⁻	-1.4 ppm		X ₅ ⁻
[M] ⁻	221.1549	C ₁₄ H ₂₀ O ₂ ⁻	-0.8 ppm		X ₆ ⁻
[M] ⁻	197.0970	C ₁₄ H ₁₂ O ⁻	-0.8 ppm	not assigned	not assigned
[M] ⁻	187.1126	C ₁₃ H ₁₄ O ⁻	-1.2 ppm	not assigned	not assigned
[M] ⁻	171.0815	C ₁₂ H ₁₀ O ⁻	-0.2 ppm	not assigned	not assigned
[M] ⁻	99.0450	C ₅ H ₆ O ₂ ⁻	-3.4 ppm		X ₇ ⁻

Table S2: The major HRMS-APCI fragments (negative mode)

Supplementary Figures

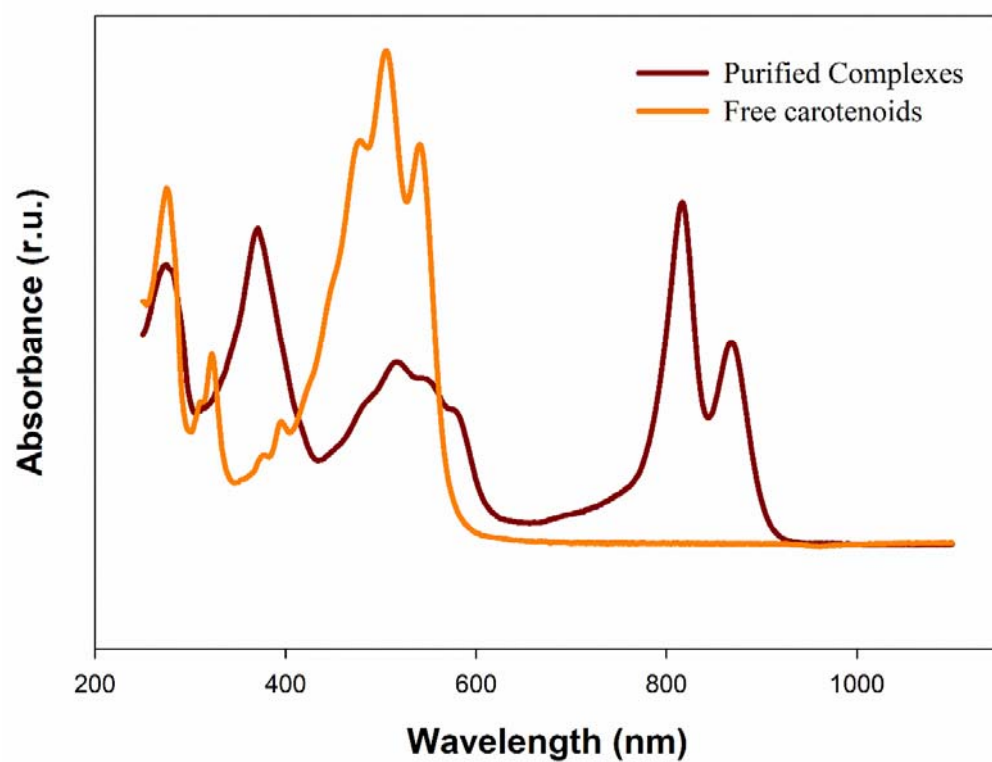


Figure S1: A UV-VIS absorption spectrum of the free carotenoid fraction (orange) and the complexes (purple). The pure PS complexes were obtained during the sucrose gradient centrifugation and then with subsequent chromatographic steps and carotenoids not bound to complexes were removed.

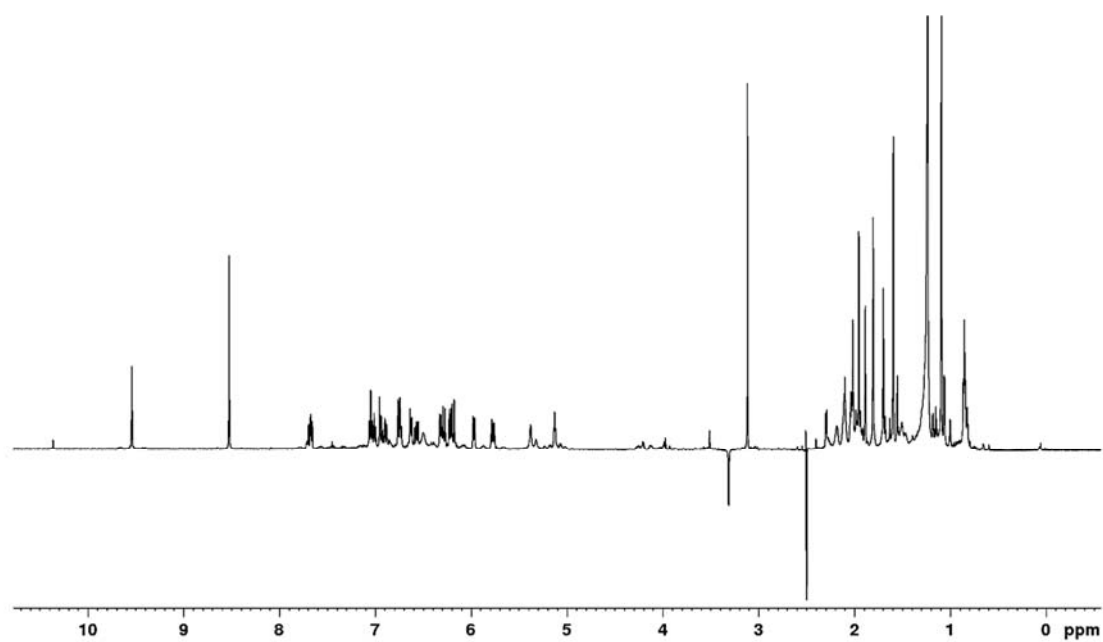


Figure S2: A ^1H NMR spectrum of gemmatoxanthin (700.13 MHz, DMSO- d_6 , 303.1 K)

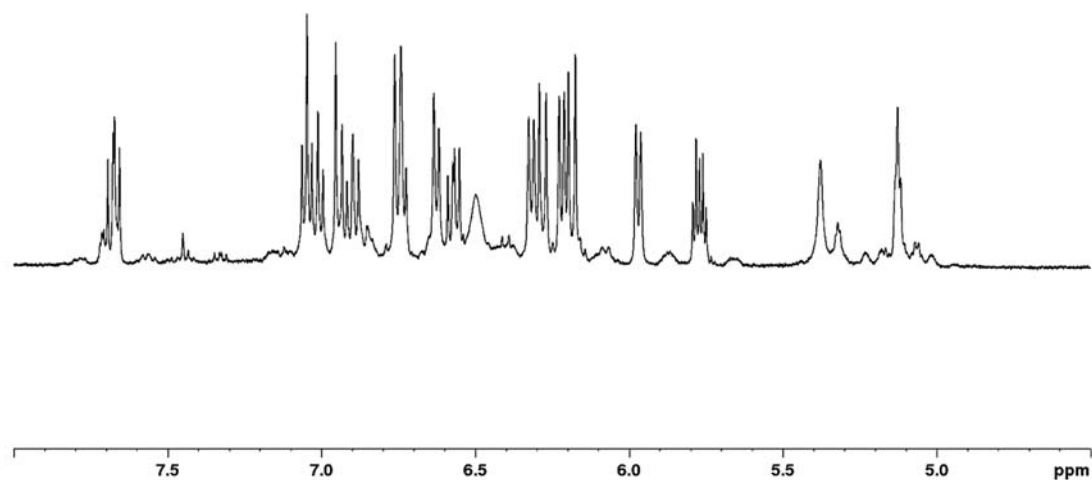


Figure S3: A detail of the ^1H NMR spectrum of gemmatoxanthin (700.13 MHz for ^1H , $\text{DMSO-}d_6$, 303.1 K) – 8 to 4.5 ppm

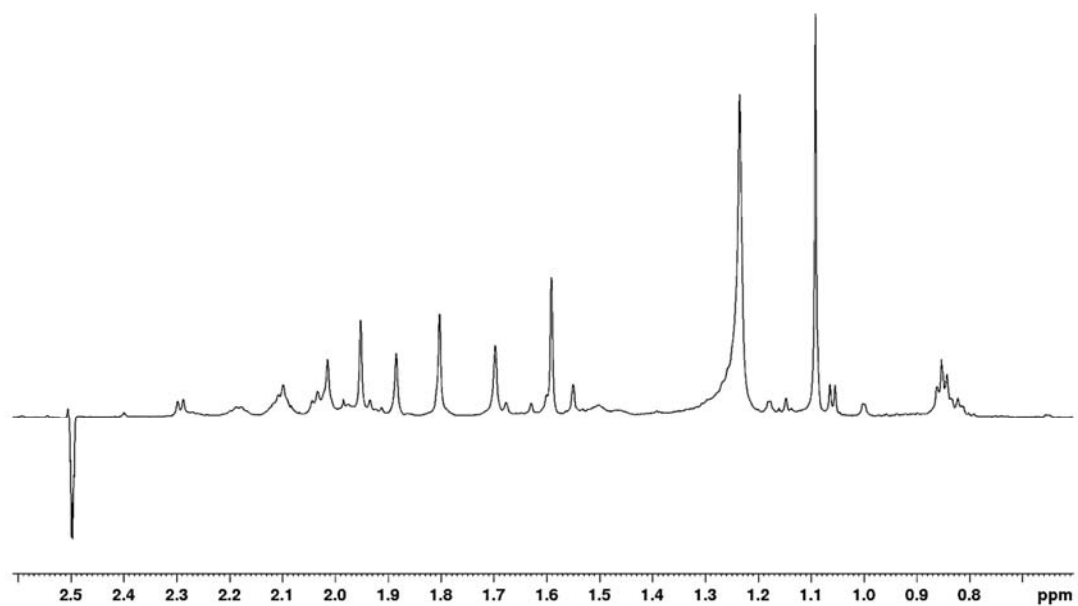


Figure S4: A detail of the ^1H NMR spectrum of gemmatoxanthin (700.13 MHz for ^1H , $\text{DMSO-}d_6$, 303.1 K) – 2.4 to 0.7 ppm

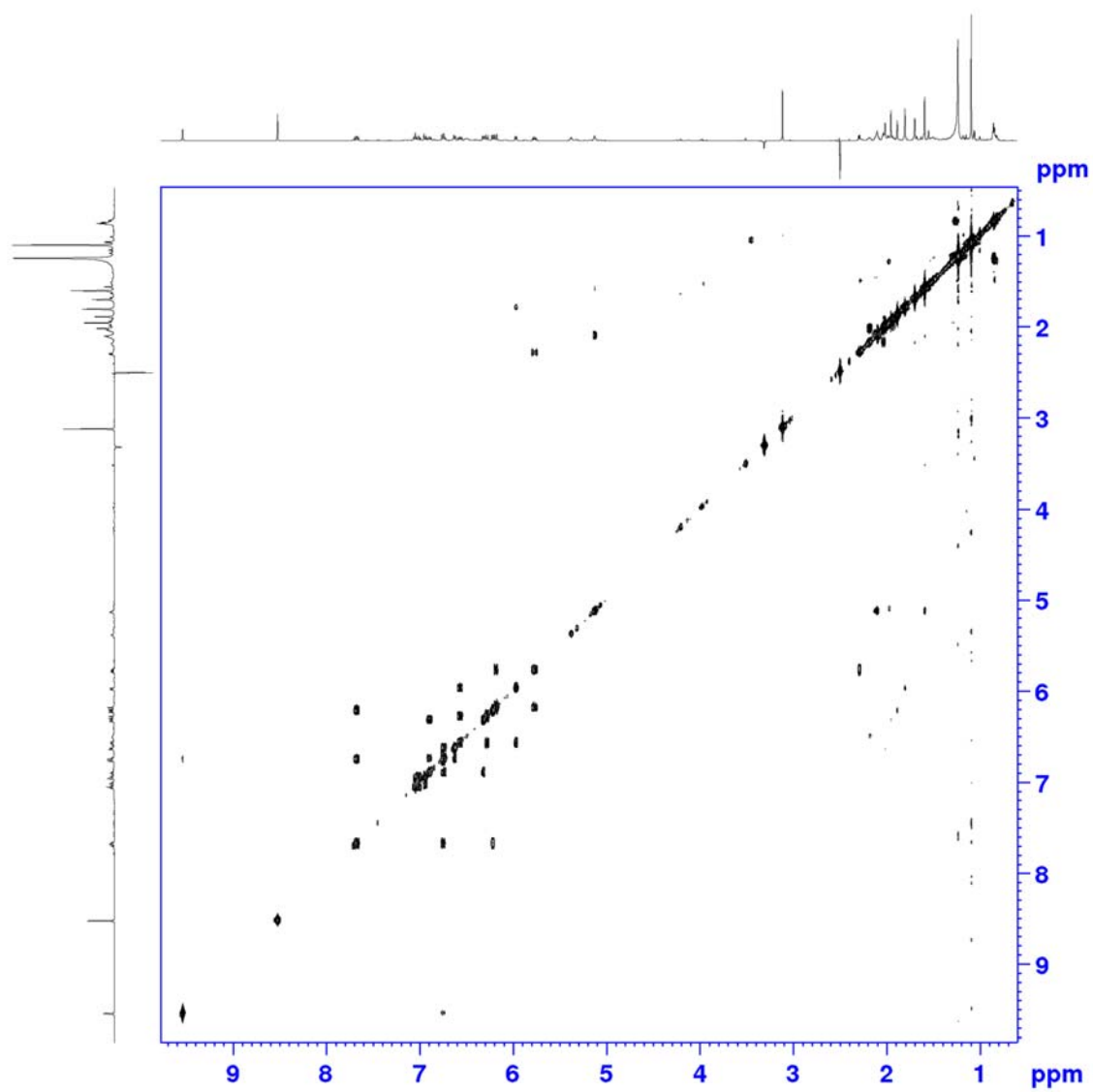


Figure S5: A COSY spectrum of gemmatoxanthin (700.13 MHz for ¹H, DMSO-*d*₆, 303.1 K)

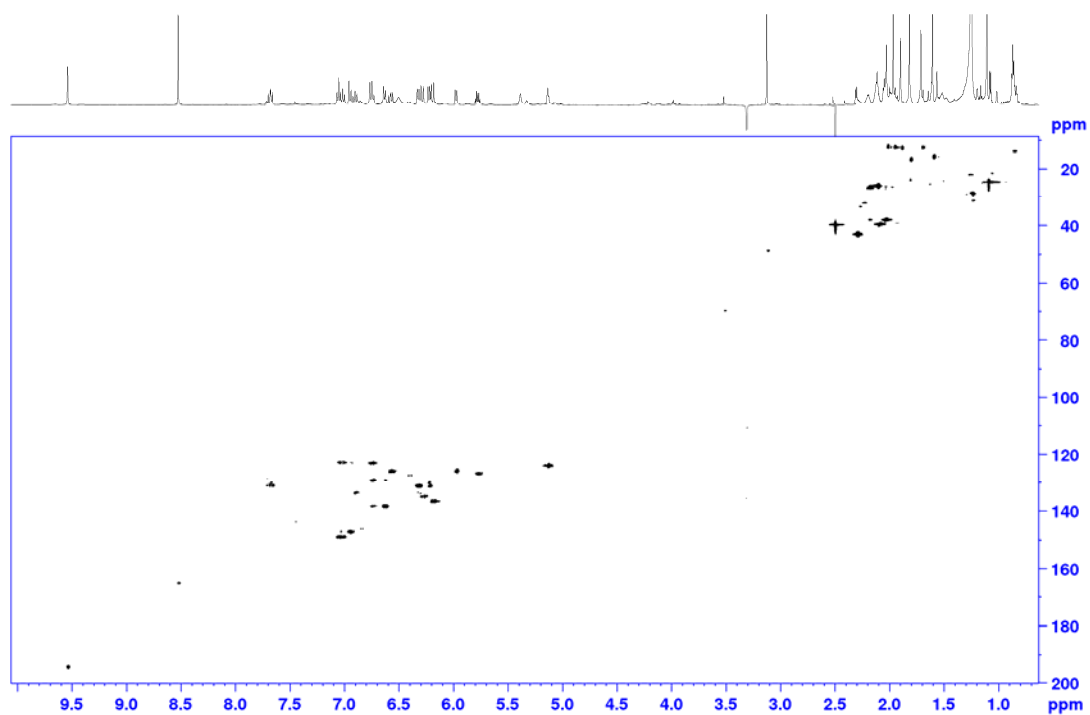


Figure S6: A ^1H - ^{13}C HSQC spectrum of gemmatoxanthin (700.13 MHz for ^1H , 176.05 MHz for ^{13}C , DMSO- d_6 , 303.1 K)

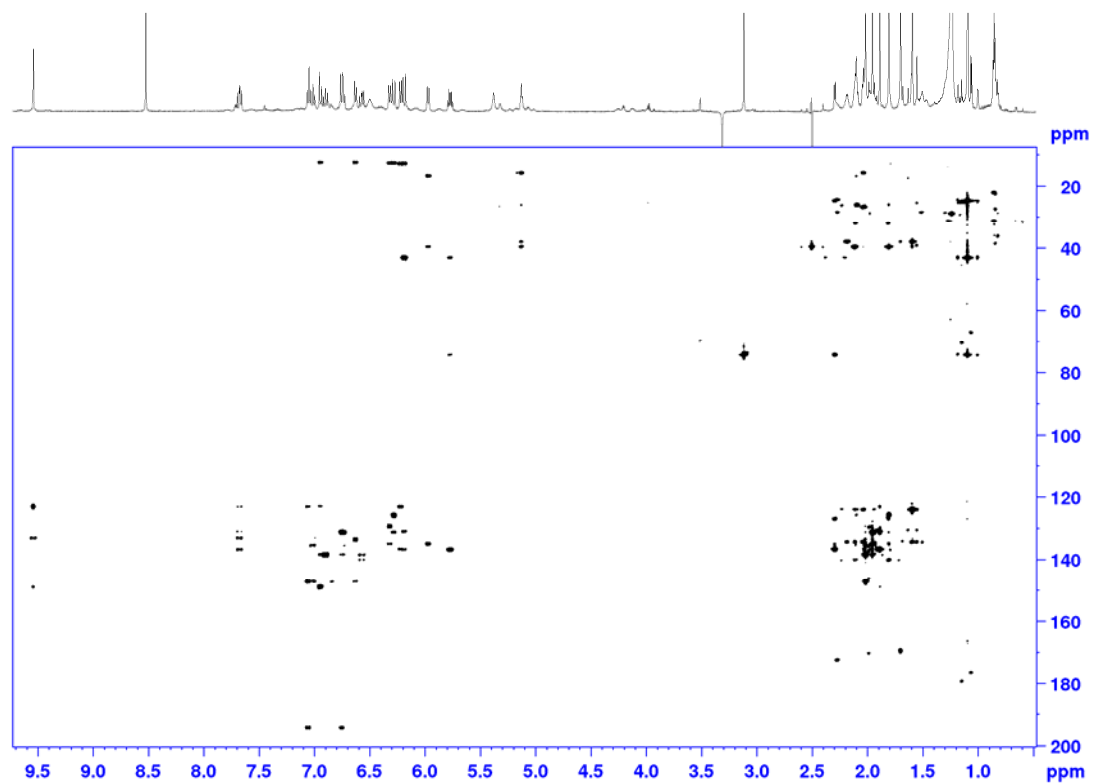


Figure S7: A ^1H - ^{13}C HMBC spectrum of gemmatoxanthin (700.13 MHz for ^1H , 176.05 MHz for ^{13}C , DMSO- d_6 , 303.1 K)

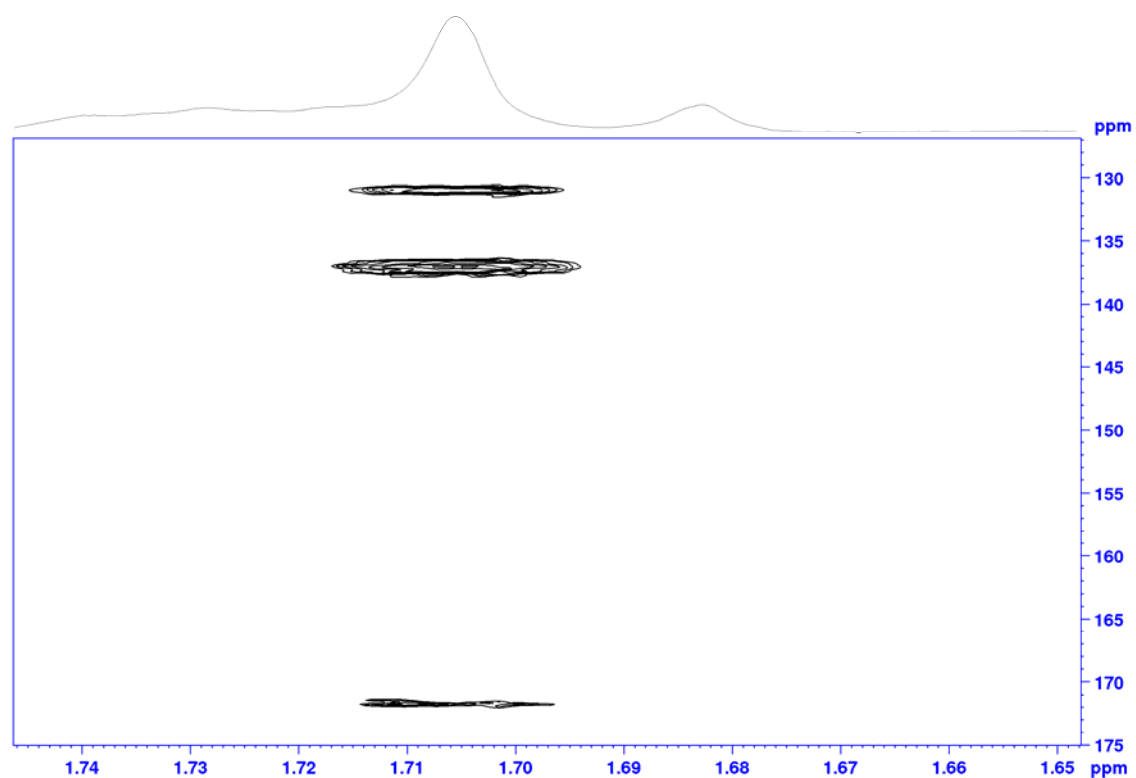


Figure S8: A detail of ^1H - ^{13}C HMBC spectrum of gemmatoxanthin (700.13 MHz for ^1H , 176.05 MHz for ^{13}C , DMSO- d_6 , 303.1 K) from the second batch of *G. phototrophica* cultivated in ^{13}C labelled SILEX *E. coli* media

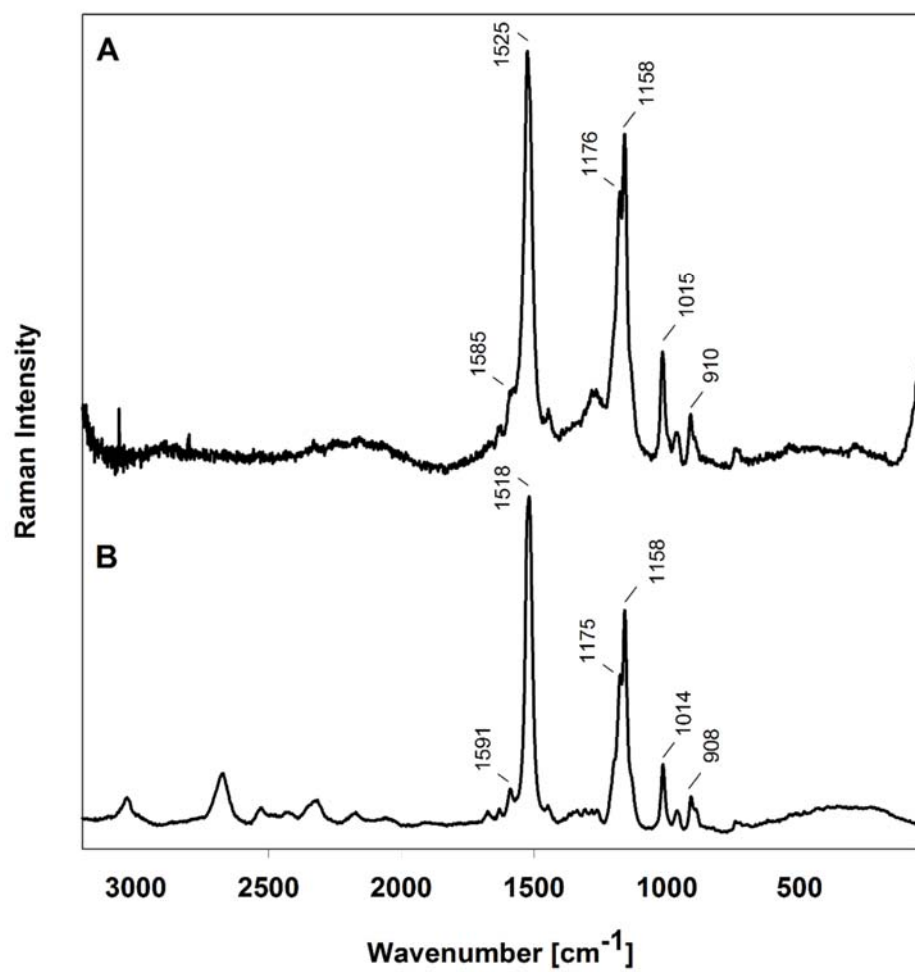


Figure S9: Raman spectra of gemmatoxanthin at (A) 785nm and (B) 532nm.