Electronic Supplementary Material (ESI) for Food and Function

Supplementary Information

New insights on the interaction mechanism between tau protein and Oleochantal, an extra-virgin olive-oil bioactive component.

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K18 modified peptides after OLC treatment

	4°C		37°C
K18 (uM)	20	20	20
OLC excess	3 and 5 fold	5 fold	5 fold
Incubation time	3 min	15 min	15 min
K18 Peptides	ΔMw 272 Da	ΔMw272 Da	ΔMw91 Da
1-12			
1-15		x	
13-15			
13-17		x	
15-38			
16-18			
16-32		x	
17-38			
18-25			
18-32		x	x
18-38		x	
26-32			
26-38		x	x
26-39		x	
26-48		x	x
32-52			
33-38			
33-39		x	
39-48		x	
39-56			x
49-52			
49-75			x
53-56			
53-75		x	
56-78			
57-75		x	
57-97		x	x
76-79			
80-98		x	
99-101			
99-107		x	
99-111			
102-105			
106-111			
112-127			
128-130			
*53-69+128-130	x	x	
*40-56+102-107	x		
*76-105+13-17	x		
*76-101+16-25	x		

Fig. S1. Proteolytic map of K18-OLC complex at 4 and 37 degree. * Peptides found to be cross-linked by OLC giving rise to a double imine macrocyclic system

NMR spectra of OLC and OLC-Propylamine adduct

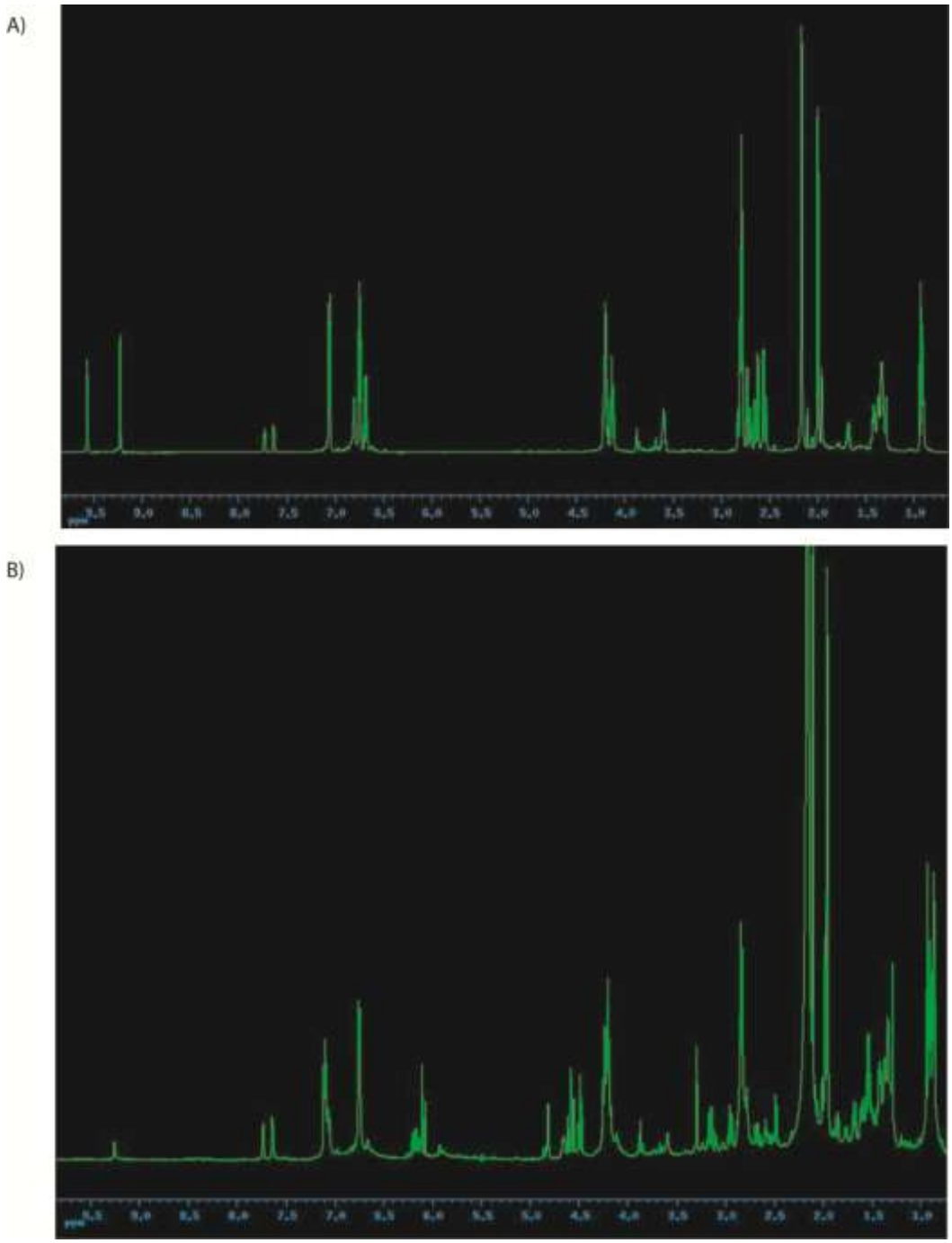
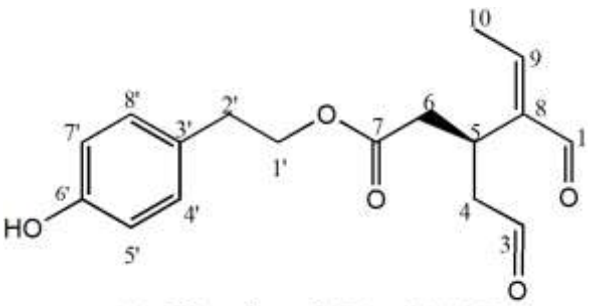


Fig.S2. Comparison between OLC ^{1}H NMR spectra (**A**) and OLC- Propylamine adduct ^{1}H NMR spectra (**B**). The loss of the signal assigned to the H(C-3) aldehyde was clearly measured confirming that first imine bond formation occurs through C-3 aldehyde.

Spectroscopic characterization of OLC



(-)- Oleochantal (Mw 304.2 Da)

Oleocanthal: colourless solid; IR (KBr) *ν*max 3030, 2830, 2740, 1740, 1725, 1200 cm-1; UV ($CDCl_3$) λmax 330, 242 nm; $[\alpha]^{25}_{D}$ -0.9, c = 2, $CHCl_3$; ESIMS *m/z* 327.3 $[M\text{-}Na]^+$; ESIMS/MS *m/z* 206.8, 121.5;.^{1}H NMR δ ($CDCl_3$, 600.13 MHz) 9.24 (H-1,d, J=1.8 Hz), 9.64 (H-3, bs), 2.74 (H-4a**,** dd, J=18.4, 5.7 Hz), 2.98 (H-4b**,** dd, J=18.4, 8.8 Hz), 3.61 (H-5, m), 2.62 (H-6a, dd, J= 15.8, 6.6 Hz), 268 (H-6b, dd, J= 15.8, 8.3 Hz), 6.63 (H-8, q, J= 7.0 Hz), 2.08 (H-10, d, J = 7.0 Hz), 4.18 (H-1'a, m), 4.22 (H-1'b, m), 2.80 (H-2', t, J = 7.0 Hz), 7.05 (H-4'/8', d, J = 8.0 Hz), 6.76 (H-5'/7', d, J = 8.0 Hz); ^{13}C NMR δ($CDCI_3$, 150.9 MHz) 195.1 (C-1), 200.4 (C-3), 46.0 (C-4), 27.0 (C-5), 36.5 (C-6), 171.9 (C-7), 154.4 (C-8), 143.4 (C-9), 15.3 (C-10), 65.2 (C-1'), 34.3 (C-2'), 129.5 (C-3'), 130.1 (C-4'/8'), 115.4 (C-5'/7'), 154.6 (C-6').

LC-MS analysis of K18 protein

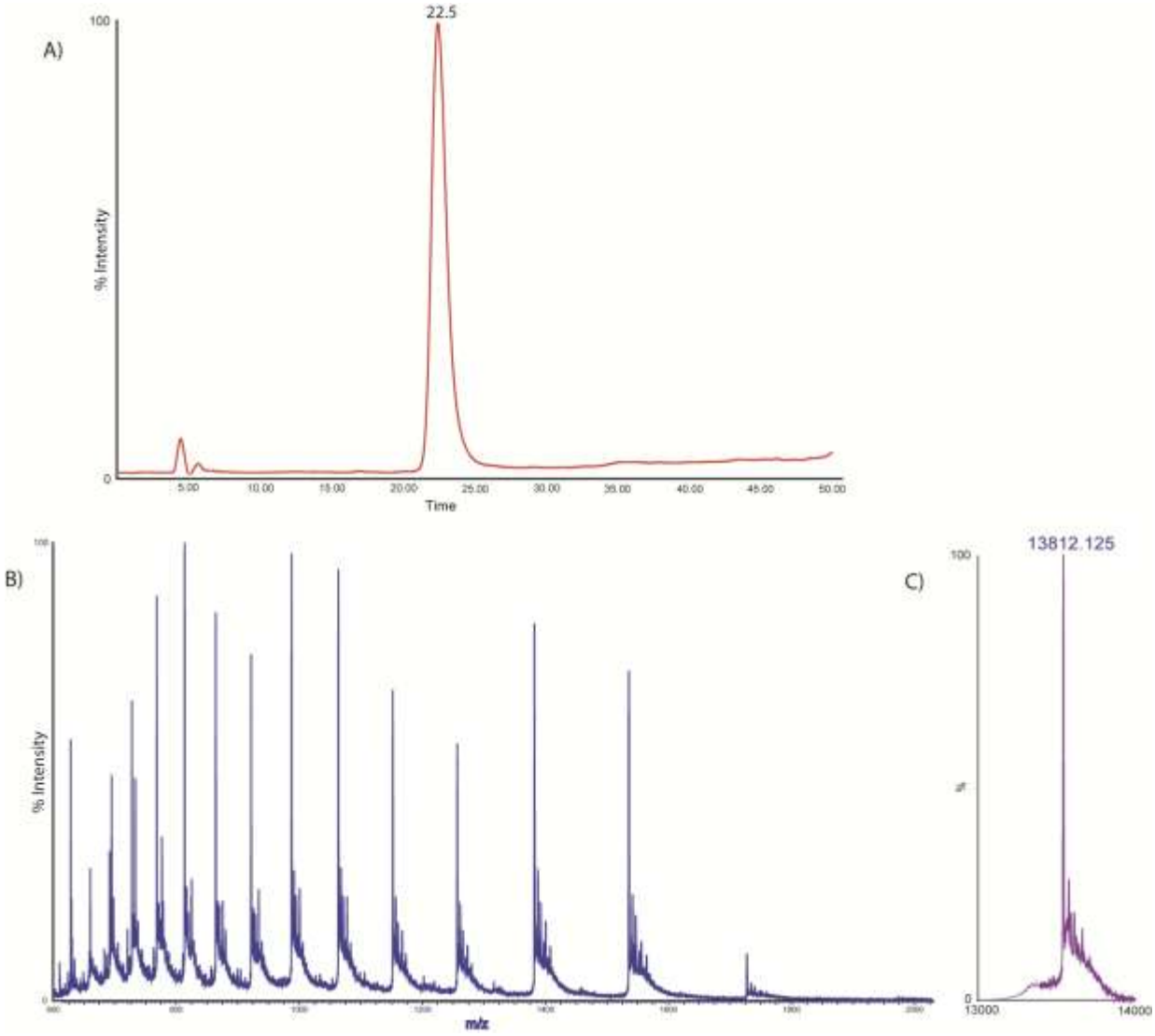


Fig.S3. LC-MS analysis of the recombinant K18 protein. K18 chromatographic profile is reported in panel **A,** while the multi-charged K18 MS spectra and deconvoluted mass spectra are reported in panel **B** and **C** respectively.