Electronic Supplementary Material (ESI) for RSC Advances.

Supporting Information

Cork extracts reduce UV-mediated DNA fragmentation and cell death

Ana R. Araújo[1,2*], David M. Pereira[1,2,4], Ivo M. Aroso[1,2], Tânia Santos[3], Maria T. Batista[3], Mariana T. Cerqueira[1,2], Alexandra P. Marques[1,2], Rui L. Reis[1,2] and Ricardo A. Pires[1,2*]

[1] 3B's Research Group - Biomaterials, Biodegradables and Biomimetics, University of Minho, Headquarters of the European Institute of Excellence on Tissue Engineering and Regenerative Medicine, AvePark, 4806-909 Taipas, Guimarães, Portugal

[2] ICVS/3B's - PT Government Associate Laboratory, Braga/Guimarães, Portugal

[3] Centro de Estudos Farmacêuticos - Faculdade de Farmácia, Universidade de Coimbra, Pólo das Ciências da Saúde, Azinhaga de Santa Comba, 3000-548 Coimbra, Portugal

[4] Present address: REQUIMTE/LAQV, Laboratório de Farmacognosia, Departamento de Ciências Químicas, Faculdade de Farmácia, Universidade do Porto, Rua de Jorge Viterbo Ferreira, n° 228, 4050-313 Porto, Portugal

* Corresponding authors: Ricardo A. Pires (rpires@dep.uminho.pt) and Ana R. Araújo (anarita.araujo@dep.uminho.pt)

HPLC characterization of samples CWE, CWEE and fCWEE

The chromatographic analysis of the cork extracts CWE, CWEE and fCWEE is presented in Figure S1. The main peaks, indicated by the letters, are assigned, respectively, to A – Vescalagin, B – Castalagin, C – Gallic acid and D – Ellagic acid. The identification was performed by comparison of the retention time and spectroscopic data from the photodiode array (PDA) with that of the standards compounds. The quantification was performed from calibration curves obtained with standard solutions of the pure compounds. The vescalagin and castalagin was previously isolated in our lab and their HPLC-ESI/MS analysis is presented in Table S1.

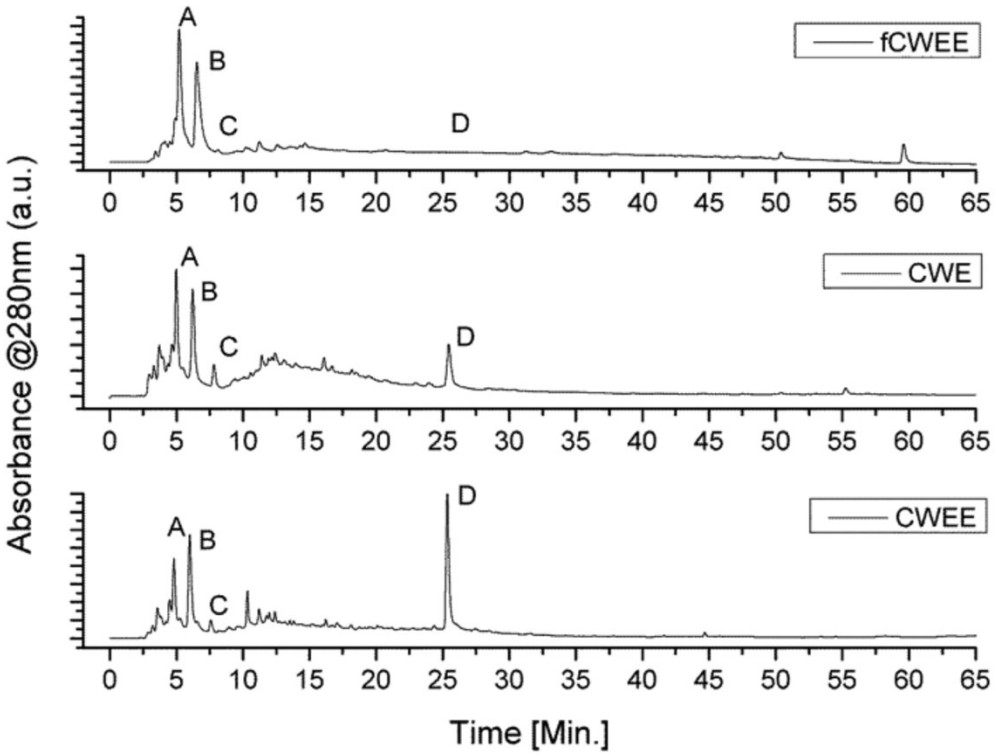

Figure S1. *HPLC chromatographic profiles of samples CWE, CWEE and fCWEE obtained using UV detection at λ=280nm; the letters indicate the most relevant peaks assigned and identified as: A – vescalagin; B – castalagin; C – gallic acid and D – ellagic acid.*

Table S1. *HPLC-ESI/MS analysis and identification of main cork extract compounds*

Retention time (min)	PDA – λ max (nm)	HPLC-ESI/MS tandem			Compound
		$[M-H]^-$	MS^2	MS^3	
5.1	251	933	915 871 569 613	871 897 569 613	Vescalagin (A)
6.5	254	933	915 631 569	613 569	Castalagin (B)
7.8	272	--	--	--	Gallic acid (C)
25.4	255; 366	--	--	--	Ellagic acid (D)

Cytotoxicity evaluation of cork extracts in L929 cells

The cytotoxicity of each extract was assessed by MTS assay. The range of concentration used for study the work concentration was from 300μg/ml until 25μg/ml. Once cells were confluent, each extract was added to the culture medium. After 24h the metabolic activity was determined. The data (Figure S2) showed no cytotoxicity up to a working concentration of 75μg/ml for the three extracts.

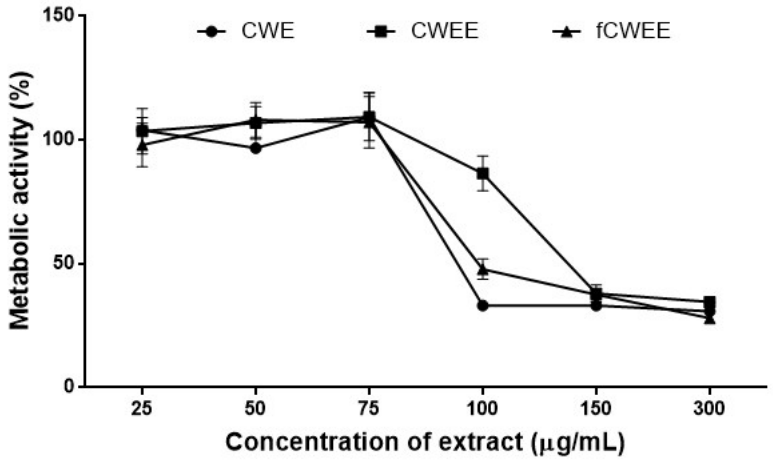

Figure S2. *Metabolic activity of L929 cells cultured with 300, 150, 100, 75, 50 and 25µg/ml of each cork extract for 24h.*

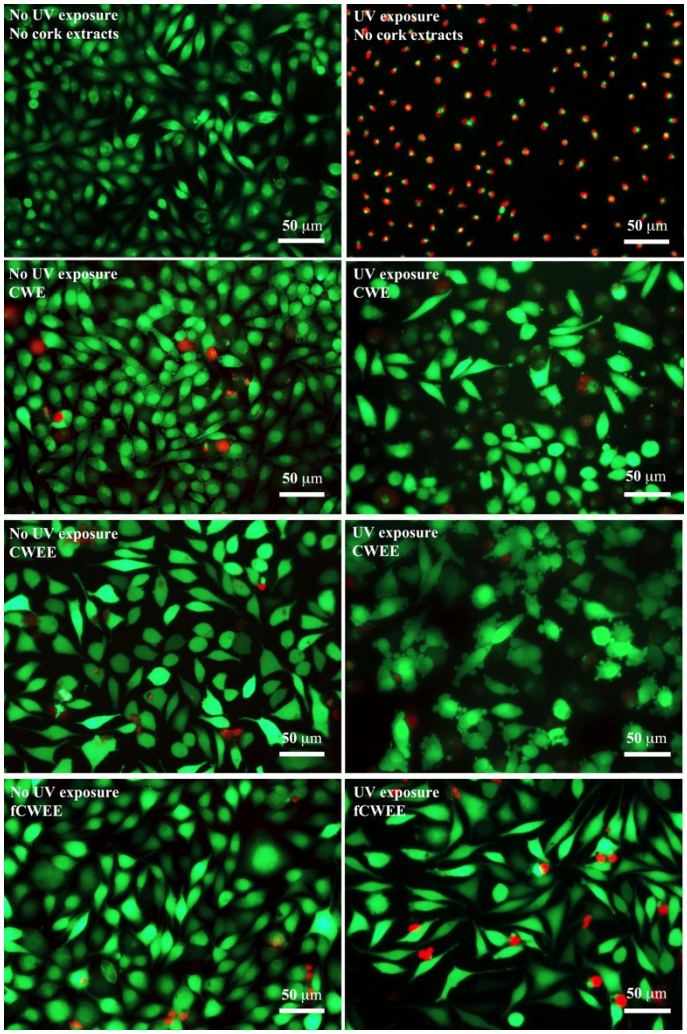

Figure S3. *Assessment of viability of the cells cultured in the absence and presence of cork extracts as well as in the presence or absence of UV-exposure. Live cells are stained in green and dead cells are stained in red.*