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## The influence of the extraction method on the DNA protective effects of seaweed extracts in Caco-2 cells

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Brown seaweeds contain a variety of compounds such as phlorotannins, carotenoids, vitamins, phospholipids and peptides that may benefit human health<sup>(1)</sup>. The extraction method and type of solvent used influences the nature of compounds extracted from seaweeds<sup>(2)</sup>. The solvents in the present study (water, ethanol and methanol) are of a polar nature and extract a range of hydrophilic compounds including the phlorotannins.

The objective of the present study was to determine the potential protective effect of extracts obtained from *Ascophyllum nodosum* (AN) and *Fucus serratus* (FS) against hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) and tert-butylhydroperoxide (tert-BOOH)-induced DNA damage in Caco-2 cells. Compounds were extracted using 100% H<sub>2</sub>O, 60% ethanol (EtOH) or 60% methanol (MeOH). Caco-2 cells were pre-treated with each seaweed extract for 24 h followed by exposure to either 50 μM H<sub>2</sub>O<sub>2</sub> or 200 μM tert-BOOH for 30 min. DNA damage was assessed by the comet assay.

	tert-BOOH (% tail DNA)		H <sub>2</sub> O <sub>2</sub> (% tail DNA)	
	Mean	SE	Mean	SE
Control	10.0	2.0	10.0	2.0
Positive control	30.0	3.5	55.0	3.0
AN (100% H <sub>2</sub> O)	20.5	2.0	35.0#	6.0
AN (60% EtOH)	18.5#	1.0	46.0	3.0
AN (60% MeOH)	30.5	1.5	61.0	1.0
FS (100% H <sub>2</sub> O)	17.5†	1.5	47.0	5.5
FS (60% EtOH)	21.5	2.5	48.0	2.0
FS (60% MeOH)	26.0	1.5	45.0	5.5

#Denotes significant protection ( $P < 0.05$ ) compared to oxidant control. †Denotes significant protection ( $P < 0.01$ ) compared to oxidant control.  $N = 4$  individual experiments. Statistical analysis was by ANOVA followed by the Dunnett's test.

The addition of 50 μM H<sub>2</sub>O<sub>2</sub> and 200 μM tert-BOOH increased the DNA damage in Caco-2 cells to 55 and 30%, respectively. Preincubation of Caco-2 cells with AN (60% EtOH) and FS (100% H<sub>2</sub>O) extracts offered significant protection against tert-BOOH-induced DNA damage. Only the AN (100% H<sub>2</sub>O) extract significantly reduced H<sub>2</sub>O<sub>2</sub>-induced DNA damage. The MeOH extracts of AN and FS did not protect against either H<sub>2</sub>O<sub>2</sub> or tert-BOOH-induced DNA damage. The DNA protective effects of the seaweeds may indicate their potential use in the pharmaceutical and functional food industry. The presence of hydrophilic polysaccharide compounds may account for the antioxidant ability of the 100% H<sub>2</sub>O extracts, whereas the antioxidant behaviour of the aqueous ethanol extracts may be due to the presence of a mixture of polar and less polar compounds.

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