

Evaluation of the anti-inflammatory activity of raisins (*Vitis vinifera* L.) in human gastric epithelial cells: a comparative study

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Gastritis is an inflammatory disease involving millions of people in the world; about 90% of cases of gastritis are caused by the presence of the bacterium *Helicobacter pylori* (*H. pylori*) while only 10% is due to other risk factors including autoimmune reactions, drugs, and alcohol. During *H. pylori* infection, macrophages release several cytokines, such as TNF α , which in turn lead to the activation of the transcription factor NF- κ B in epithelial gastric cells (Mai et al., 1991). NF- κ B is strictly involved in the expression of a variety of inflammatory genes, including IL-8 (Yasumoto et al., 1992). This chemokine is mostly released by gastric epithelial cells during gastric inflammation (Shimada et al., 1998). Raisins are dried grapes largely consumed in the Mediterranean area; they are an important dietary source of potassium, fibers and polyphenols. In literature, different studies have documented the potential health benefits of these fruits; methanol extracts from raisins already showed anti-inflammatory properties in human gastric epithelial cells (AGS), these extracts were able to decrease protein and mRNA levels of ICAM-1 in TNF α stimulated cells (Kaliora et al., 2008). However, no studies regarding the anti-inflammatory activity of the hydro-alcoholic or water extracts, that is the mostly used for food supplements preparation, have been reported until now.

The aim of this study was to compare the anti-inflammatory activity of the water and hydro-alcoholic extracts of raisins from different origins (var. sultana, *Vitis vinifera* L.) at gastric level.

The extracts were prepared from five different raisins: one from Portugal (sample 1), one from Turkey (sample 2) and three commercially available (sample 3,4 and 5); the extracts were screened for their ability to inhibit IL-8 release in AGS cells stimulated with TNF α (10 ng/mL); IL-8 secretion and NF- κ B nuclear translocation were assayed by ELISA assays. NF- κ B driven transcription and IL-8 promoter activity were assayed transfecting cells with reporter plasmids containing luciferase gene, in particular: NF- κ B-Luc plasmid containing E-selectin promoter characterized by the presence of three responsive elements κ B and IL-8-Luc plasmid containing human IL-8 gene characterized by the presence of a binding site for NF- κ B.

Among raisin extracts, only sample 2 significantly inhibited IL-8 secretion at 100 μ g/mL; in particular, the hydro-alcoholic extract showed higher reduction of TNF α -induced IL-8 release with respect to the water extract (IC₅₀: 3.34 vs. 94.18 μ g/mL, respectively). Macroscopic evaluation of sample 2 raisin showed the presence of seeds, whereas the other raisins samples were seedless.

In order to investigate the contribution of seeds, the hydro-alcoholic extracts from fruit and seeds were prepared and individually tested on IL-8 release, IL-8 promoter activity, NF- κ B driven transcription and nuclear translocation; seeds extract inhibited all the previous parameters in a concentration-dependent manner, showing higher efficacy than fruit extract. IC₅₀ from the seeds extract was lower than the fruit extract in each parameter evaluated.

	IL-8 secretion IC ₅₀ \pm s.d. (μ g/mL)	IL-8 promoter activity IC ₅₀ \pm s.d. (μ g/mL)	NF- κ B driven transcription IC ₅₀ \pm s.d. (μ g/mL)	NF- κ B nuclear translocation IC ₅₀ \pm s.d. (μ g/mL)
Seeds extract	0.49 \pm 0.05	0.75 \pm 0.25	1.21 \pm 0.21	2.62 \pm 0.35
Pulp extract	35.8 \pm 6.5	51.3 \pm 8.43	> 250	> 100

These results suggest that seeds could have an important contribution in the anti-inflammatory activity of raisins and their consumption could confer a beneficial effect against gastric inflammatory diseases.

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Yasumoto et al. (1992). *The Journal of biological chemistry.* 267, 22506-22511

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