Protective effects of olive leaf extract on inflammatory activation of endothelial cells

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BACKGROUND

Olive leaf extract (OLE) has been traditionally used due to its anti-inflammatory and anti-atherogenic effects. The mechanism through which OLE modulates atherogenesis is unclear. Serum amyloid A (SAA) is a major inflammatory mediator in humans linking inflammation and atherosclerosis.

OBJECTIVE

To explore the anti-inflammatory properties of OLE in the SAA-activated endothelial cells from the human coronary arteries (HCAEC), the most susceptible vascular site in atherosclerosis.

METHODS

HCAEC were treated with SAA (1000 nM; 24h) and/or OLE (0.5 and 1.0 mg/ml; 45 min prior SAA). The amount of secreted IL-6 and IL-8 was quantified by ELISA. The expression of VCAM-1, E-selectin, matrix metalloproteinases (MMP2 and MMP9) and miRNAs 146a and let-7e was determined by qPCR. Phosphorylation of NF-κB was assessed by Western blot and DNA damage was measured using the COMET assay.

OLE regulates SAA-induced miR-146a and let-7e

The mean fold changes of mRNA expression levels of miR-146a (A) and let-7e (B) are shown for 3 biological replicates.

OLE attenuates SAA-induced and H2O2-induced DNA damage in HCAEC

COMET assay was performed without (A) and with oxidative treatment (3% H₂O₂) (B). DNA damage was evaluated according to the following categories: A - no DNA damage, B - slight tail, C - strong signal in tail and nucleus, D - majority of DNA in tail, E - no DNA in round nucleus.

CONCLUSION

OLE significantly attenuated certain pro-inflammatory and pro-adhesive responses and decreased DNA damage in HCAEC upon stimulation with SAA. The reversal of SAA-driven endothelial activation by OLE might contribute to its anti-inflammatory and anti-atherogenic effects in HCAEC.

OLE decreases SAA-driven release of pro-inflammatory cytokine IL-6 and chemokine IL-8 from HCAEC

The amount of IL-6 (A) and IL-8 (B) in cell culture supernatants was measured by ELISA. The mean of 3 biological replicates is shown.

OLE decreases mRNA expression of adhesion molecules VCAM-1 and E-selectin and MMP2 in HCAEC

The expression of VCAM-1 (A), E-selectin (B), MMP2 (C) and MMP9 (D) mRNA was determined by qPCR. Mean of 3 biological replicates is shown.

RESULTS

OLE decreased significantly released protein levels of IL-6 and IL-8, as well as mRNA expression of E-selectin in SAA-stimulated HCAEC and reduced MMP2 levels in unstimulated cells. Phosphorylation of NF-κB was upregulated in the presence of SAA, with OLE significantly attenuating this effect. OLE stabilized SAA-induced upregulation of microRNA-146a and let-7e and protected HCAEC from SAA- and H₂O₂-driven DNA damage.

OLE attenuates SAA-induced phosphorylation of NF-κB.

Western blot shows phosphorylation of NF-κB in HCAEC pretreated with OLE for 1 h and then treated with/without SAA for 1h.