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May 17–19, 2010, Joensuu, Finland*

*Abstracts*



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## **(40) Red clover (*Trifolium pratense* L.): Identification and characterization of phenolic compounds and protective action against oxidative stress and inflammation**

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Red clover (*Trifolium pratense* L.), a widely used feed crop, has recently received considerable interest as a valuable source of phenolic compounds with multiple potential protective functions. It is a rich source of isoflavonoids, plant secondary metabolites widely distributed in the Leguminosae family. Phenolic compounds in red clover were identified and characterized by high-performance liquid chromatography and mass spectrometry. A total of 31 and 28 phenolic compounds were tentatively identified in leaves and roots, respectively. Leaves were rich in glycoside malonates of biochanin A, formononetin and quercetin.

Quercetin (flavonol) and biochanin A (isoflavone) were selected for the evaluation of their ability to inhibit oxidative stress and inflammation using cell culture models (human ARPE-19 cells and N9 murine cells). Inflammation and oxidative stress responses were analyzed by measuring the expression of interleukin-6 (IL-6), IL-1 $\beta$ , TNF- $\alpha$  (tumor necrosis factor alpha), GSTP1 (Glutathione S-transferase P1) and HO-1 (heme oxygenase 1). The expression of nuclear factor-erythroid 2-related factor-2 (Nrf2), a key transcription factor involved in defense against oxidative stress, was also analyzed.

Our results indicate that quercetin protected ARPE-19 cells from oxidative stress-induced cell death, whereas biochanin A had no statistically significant protective effect. Quercetin also effectively protected N9 cells from LPS-induced inflammation in a dose-dependent manner, measured as the levels of secreted IL-6, whereas the effects of biochanin A were minimal. Quercetin slightly reduced the expression of IL-6 mRNA in cells treated with LPS, whereas biochanin A showed no protective effects. Neither compound decreased the expression of TNF- $\alpha$ . Quercetin also reduced the gene expression of IL-6 and IL-1 $\beta$ , and increased the expression of transcription factor Nrf2 and protective enzymes HO-1 and GSTP in cells treated with H<sub>2</sub>O<sub>2</sub>. Our results suggest that the flavonol quercetin possesses higher protective function against oxidative stress and inflammation related diseases than the isoflavone biochanin A.