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Antioxidant properties of polyphenols and synthetic compounds

My doctoral dissertation consists of 3 papers published in the journal *Food Chemistry* devoted to (i) comparison of antioxidant properties of flavanols and other natural and synthetic antioxidants, (ii) studies of the effect of binding Fe^{2+} ions on the antioxidant properties of flavanols, and (iii) comparison of generation of hydrogen peroxide by various antioxidant and an insight into the mechanism of this phenomenon.

The flavanols studied [(+)-catechin, (-)-epicatechin, (-)-epigallocatechin, (-)-epicatechin gallate and (-)-epigallocatechin gallate] are very good antioxidants in various cell-free systems, protecting against such oxidants as peroxyxynitrite, hypochlorite and peroxy radicals in the tests of inhibition of dihydrorhodamine 123 oxidation and inhibition of oxidation-induced fluorescein bleaching. In the majority of these tests as well as in the test of reduction of ABTS[•] radical, the antioxidant activities of the flavanols were much higher than those of standard antioxidants (glutathione and ascorbate). Flavanols protected also erythrocytes against oxidant-induced hemolysis.

Fe^{2+} ions were found to form complexes with flavanols; stoichiometry of these complexes was different for various compounds. Flavanol complexes formed by addition of substoichiometric amounts of Fe^{2+} (1 mol Fe^{2+} per 4 moles of a flavanol) had antioxidant properties similar to those of flavanols alone. Complexes of flavanols with Fe^{2+} did not show pseudoenzymatic activities of superoxide dismutase or catalase.

Comparison of hydrogen peroxide generation in cell culture media by 54 natural and synthetic antioxidants showed that half of them (27 compounds) produced hydrogen peroxide. Generation of hydrogen peroxide was lower in media used for yeast culture than in media used for culture of mammalian cells and phosphate-buffered saline (PBS). The highest amounts of hydrogen peroxide was generated by propyl gallate, pyrogallol, (-)-epigallocatechin gallate and quercetin. Ascorbate generated hydrogen peroxide but when combined with polyphenols it decreased hydrogen peroxide generation by the polyphenols.

Apparently, hydrogen peroxide is formed mainly by dismutation of the superoxide radical anion generated in two successive one-electron reaction of polyphenol oxidation to semiquinone radicals and subsequent oxidation of semiquinone radicals. In agreement with this mechanism, the formation of semiquinone radicals during oxidation of propyl gallate and (-) epigallocatechin gallate was demonstrated by EPR spectroscopy and superoxide formation was demonstrated by superoxide-dismutase inhibitable NBT reduction and dihydrorhodamine 123 oxidation.

The factors determining the amount of hydrogen peroxide are: the concentration of trace amounts of transition metal ions and pH of the solution. Generation of hydrogen peroxide in the tea

was confirmed. It was found that addition of lemon to the tea considerably decreases the amount of hydrogen peroxide generated.

Hydrogen peroxide generated in the DMEM medium contributed to the cytotoxic action of propyl gallate, (-)-epigallocatechin gallate and quercetin since the presence of catalase in the medium decreased the cytotoxic action of these compounds on DU-145 cells.