Development of FIA-chemiluminescence methods to evaluate quenching effects against reactive oxygen species

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Recently, special attention has been focused on quenching effects for reactive oxygen species (ROS) in foods. A simple and rapid evaluation method for quenching effect against ROS is required to control food quality and human health. In this study, FIA-chemiluminescence (CL) methods to evaluate quenching effects of functional foods against ROS such as singlet oxygen (1O2) and hydroxyl radicals (OH) are developed.

The proposed methods are based on CL reaction between luminol and ROS. A Fenton-reaction and a H2O2-NaBr-lactoperoxidase (LPO) system were utilized to prepare hydroxyl radical and singlet oxygen, respectively.

FIA conditions: Carrier solution, 0.1 M acetate buffer (pH 7.4) for 1O2 and 0.1 M Hepes buffer (pH 7.4) for OH; CL reagent, 10 µg/ml LPO in acetate buffer for 1O2 and 8 mM diethylenetriaminepentaacetic acid and 200 µM FeCl2 for OH. Quenching effect was calculated by the following equation: quenching effect % = 100 - [{(CL intensity of sample/CL intensity of blank)} x 100]. The increased value means an increase quenching effect.

Optimization of FIA conditions (concentration of luminol, flow rate of carrier solution and CL reagent) improved the precision of measurement for OH and gave the relative standard deviation of less than 2.5% (n=5). Applicability of the proposed methods was confirmed by measuring quenching effects of grape seed extracts, chalcone, pelargonidine, cyaniding, delphinidine and resveratrol. Quenching effects against 1O2 were ranging from 3.1 to 100%. A rapid measurement for both ROS could be achieved by the proposed method (2 or 3 injections/min).