Note

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A correlation between the superoxide anion scavenging capacity of antioxidants and their antioxidant capacity as measured by the galvinoxyl or DPPH method

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Abstract

The superoxide anion scavenging capacity of 11 antioxidants (AH) was observed, and the results were compared with the antioxidant capacity as measured by DPPH and galvinoxyl methods. The superoxide anion was generated by the xanthine/xanthine oxidase system. 4-[3-(4-iodophenyl)- 2-(4- nitrophenyl)-2H-5-tetrazolio]-1,3-benzene disulfonate sodium salt (WST-1) was used as a probe of the superoxide anion. The color development rates of WST-1 were measured under various [AH]/[WST-1] ratios, and the data obtained were analyzed according to the following formula.

$R = \Delta A_0/\Delta A = 1 + (k_A/k_W)*([AH] / [WST\text{-}1])$

, where ΔA_0 and ΔA are the color development rates of WST-1 in the absence and presence of antioxidants, respectively, and k_A and k_W are the rate constants of the reaction of superoxide anion with the antioxidants and WST-1.

The slope k_A/k_W calculated from the linear regression of the plot of [AH]/[WST-1] vs. V_o/V, indicated the relative superoxide anion scavenging capacity of each antioxidant.

The superoxide anion scavenging capacity decreased in the order of caffeic acid, n-propyl gallate, 7,8-dihydroxyflavone, gallic acid, catechin, pyrogallol, quercetin, L-ascorbic acid, BHA, 4-hydroxycoumarin and ferulic acid. Correlation coefficients between the superoxide anion scavenging capacity and the antioxidant capacity as measured by the galvinoxyl or DPPH method were as low as 0.448 (galvinoxyl method) and 0.368 (DPPH method), and it was shown that the antioxidant capacity measured by DPPH method or galvinoxyl method does not necessarily reflect the scavenging capacity of superoxide anion.

Keywords : antioxidant capacity, superoxide anion, DPPH, galvinoxyl, WST-1

I Introduction

Over the past few decades, many researchers have focused on natural antioxidants in food not only because of the impact of oxidation on the flavor of food, but also because it had been widely recognized that antioxidants are important heath-protecting factors. However, screening antioxidants in food, which involves the separation of each antioxidant compound and studying it individually, is costly and inefficient. Therefore, a convenient method for the quick quantitation of antioxidant effectiveness in preventing diseases is appealing for researchers, and many methods to measure antioxidant capacity have been reported. Depending upon the reactions involved, these assays can roughly be classified into two types: assays based on hydrogen atom transfer (HT) reactions and assays based on electron transfer (ET) reactions.

Many hydrogen atom transfer based assays have been reported and include the inhibition of induced low-density lipoprotein autoxidation, oxygen radical absorbance capacity (ORAC)[1], total radical trapping antioxidant parameter (TRAP)[2], and crocin bleaching assays[3].

Electron transfer reaction based assays include the total phenols assay using Folin–Ciocalteu reagent[4], Trolox equivalence antioxidant capacity (TEAC)[5], and ferric ion reducing antioxidant power (FRAP)[6].

Assays using stable radicals such as α,α-diphenyl-β-picrylhydrazyl (DPPH)[7], 2,2'-azinobis(3-ethylbenzothiazoline-6-sulfonic acid)[5], or N,N-dimethyl-p-phenylene- diamine

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