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Assaying antioxidants for nitrous acid scavenging activity with four assay systems: a comparative analysis

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Abstract: Four different assay systems for determination of nitrous acid have been optimized to evaluate a group of potential antioxidants for their nitrous acid scavenging activity (NASA). The methods show linear ranges for detection within 100 nmole nitrite with relative sensitivity order based on their regression estimates (mean values within parenthesis): iron (II) oxidation (38.1) > diazotization (8.9) > iodometric (6.5) > ferrocyanide (5.55). Relative response order of the assays to test antioxidants, based on per cent test agents responding to the given assay, has been (approximate values within parenthesis): ferrocyanide (95) >diazotization (68) > iron (II) oxidation (53)> iodometric (42). Highly active agents with potency calculated as mean NASA (nmoles nitrite scavenged per µmole test agent) by different methods (the mean values within parenthesis) and their relative order has been found as: TGA (413) > ascorbic acid (56.4) > quercetin (39.3)> gallic acid (31.9)> resorcinol (26)> morin (24.8)> curcumin (23.7)> daflon (22.6)> diosmin (21.4). Weak to moderately active agents included hydroquinone (14.2) ≥ thiourea (14) > rutin (13.7) > catechol (5.8) > guaiacol (3.3)>phenol (1.8)>oxalic acid (0.6). DMSO (0.11), citric (0.09) and tartaric acids (0.15) have shown negligible activity those too detectable with only ferrocyanide method while other methods failed to detect using 50 µmole mass each. Solvents viz. ethanol, methanol, acetone and dilute sodium hydroxide have shown characteristic affects in one or more test assay systems. The study provides opportunity to screen test antioxidants for their NASA using a variety of assay protocols.

Key words: Nitrous acid scavengers, Assays, Flavonoids, Phenolics, Ascorbic acid, Thiourea, TGA, Curcumin.

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