

Evaluation of Antioxidant Properties of Six Underutilized Edible Wild Fruits in Sri Lanka

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Abstract

Protection against free radicals can be impeded by improving the dietary intake of antioxidants. Wild fruits and their products are a valuable source of antioxidant. Therefore, objectives of this study were to determine and compare different mode of antioxidant activities of six different underutilized wild fruits i.e. *Calophyllum calaba* (Keena), *Syzygium cumini* (Madan), *Drypetes sepiaria* (Weera), *Syzygium caryophyllatum* (Dan), *Ficus racemose* (Attikka) and *Careya arborea* (Kahata) found in natural habitats of Sri Lanka. Antioxidant properties of wild fruits were tested using different *in vitro* assays using 96 well micro plates. Free radical scavenging activity (FRSA), Ferric reducing antioxidant power (FRAP) and oxygen radical absorbance capacity (ORAC), total phenolic contents (TPC) and total flavonoid content (TFC) was determined by using standard procedures. Softmax Pro 5.2 v software was used to calculate antioxidant values. Results showed that 2,2-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS⁺) and 1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity in all fruit types. DPPH and ABTS⁺ radical scavenging activity of selected wild fruits were in the range of 30.49±4.87 to 585.40±6.43 mg Trolox equivalents (TE)/g of extract and 43.08±02.89 to 435.30±6.62 mg (TE)/g of extract respectively. *Calophyllum calaba* recorded the lowest FRSA with the highest IC₅₀ value in each assay. Total reducing potential of fruit extracts that was measured using FRAP revealed that significantly (P<0.05) higher value in *Careya arborea* (455.91 ± 33.94 mg TE/g of extract) while the lowest in *Calophyllum calaba* (12.35 ± 0.10 mg TE/g of extract). ORAC of selected fruit extracts were ranged from 7 to 25 mg TE/g extract. The highest TPC was recorded in *Careya arborea* (590.95 ± 22.17 mg gallic acid eq/g extract) followed by *Ficus racemose* (180.04 ± 5.09 mg gallic acid eq/g extract). *Syzygium caryophyllatum* recorded the highest TFC (13.56 ± 0.26 quercetin/g of

extract) while *Calophyllum calaba* and *Syzygium cumini* showed the lowest TPC and TFC respectively. Overall results suggested that selected fresh wild fruits possess marked antioxidant activities and *Careya arborea* and *Calophyllum calaba* L recorded the highest and the lowest antioxidant activity showing the use of these wild fruits for prevention and management of oxidative stress associated chronic diseases in Sri Lanka.

Keywords: antioxidant activities, free radicals, wild fruits

1. Introduction and research problem

It has been proven that wild fruits and their products are a valuable source of protective agents against cancer, cardiovascular diseases, neurodegenerative diseases and ageing, due to the bioactive properties of them. Underutilized fruit crops are characterized by the fact that they are abundant locally but scarce globally, and less scientific information is available. Over 60 varieties of underutilized fruit crops are found in Sri Lanka. Protection against free radicals can be enhanced by improving the dietary intake of antioxidants that are capable of preventing or slowing the oxidation of other molecules and protect cells from damages caused by exposure to reactive oxygen species which are produced during normal oxidation reactions in biological cells. Detailed studies on comparison of antioxidant properties of underutilized wild fruits in Sri Lanka, which explore the medicinal potential is almost non-existence. Therefore, the present study was focused on determination and comparison of different mode of antioxidant activities of six different underutilized wild fruits grown in Sri Lanka.

2. Research Methodology

Ethanollic extracts (95 % v/v) of freeze dried, six edible wild fruits namely Keena (*Calophyllum calaba* L; Family-Guttiferae), Madan (*Syzygium cumini* L; FamilyMyrtaceae), Weera (*Drypetes sepiaria*; Family-Euphorbiaceae), Dan (*Syzygium caryophyllatum* L; Family-Myrtaceae), Attikka (*Ficus racemose* L; Family-Moraceae) and Kahata (*Careya arborea*; Family-Lecythidaceae) were used in this study. Samples were collected from Rathnapura (Keena, Madam and Kahata), Anuradhapura (Weera) and

Colombo (Dan and Attikka) districts. Antioxidant properties of wild fruits were tested using different *in vitro* assays using 96 well micro plates. Free radical scavenging activity was measured using 2,2-azinobis-3ethylbenzothiozoline-6-

sulfonic (ABTS⁺) radicals and 1diphenyl-2picryl-hydrazyl (DPPH) based on the methods described by Re *et al.*,(1999) and Blois (1958) respectively. Five concentrations of each fruit pericarp extract (31.25, 62.5, 125.0, 250.0 and 500.0 µg/mL) were used to calculate IC₅₀ value. Trolox was used to construct the standard curve. Ferric reducing antioxidant power (FRAP) (Kucuk *et al.*, 2007) and oxygen radical absorbance capacity ORAC (Ou *et al.*, 2001) were measured using Trolox as the standard. The Folin-Ciocalteu method (Singleton *et al.*, 1999) was adopted to determine total phenolic contents (TPC) in fruit pericarp extracts using Gallic acid standard curve. Total flavonoid content (TFC) was determined by Aluminium chloride method (Siddhuraju and Becker, 2003). Quercetin was used to construct the standard curve. Three replicates were used in each assays. Softmax Pro 5.2 v software of the micro plate reader (SPECTRAMaxPLUS³⁸⁴ Molecular Devices, California, USA) was used to calculate antioxidant values of all samples.

3. Results and findings

Pericarp extracts of wild fruits showed ABTS⁺ and DPPH radical scavenging activity at all tested concentrations regardless of fruit species. Free radical scavenging activity increased with the increment of sample concentration. Mean DPPH and ABTS⁺ radical scavenging activity of six wild fruits were in the range 30.49±4.87 into 585.40±6.43 mg Trolox equivalents (TE)/g of extract and 43.08±02.89 into 435.30±6.62 mg (TE)/g of extract respectively. Keena recorded the lowest free radical scavenging activity in each assay. Among tested fruit extracts, reduction of metal ions that was measured using FRAP revealed a significantly ($P < 0.05$) higher value in Kahata extracts (455.91 ± 33.94 mg TE/g of extract) while the lowest was from Keena (12.35 ± 0.10 mg TE/g of extract). FRAP of Kahata, Attikka, Dan and Keena showed significant difference ($P < 0.05$) among each other. Oxygen radical absorbance capacity of selected fruit pericarp extracts ranged from 07.03 ± 0.88 mg TE /g to 25.54 ± 3.36 mg TE /g in Kahata and Weera respectively. ORAC values of Kahata, Keena and Attikka did not show a significant difference ($P > 0.05$) among each other. The highest TPC was recorded in Kahata (590.95 ± 22.17 mg gallic acid eq/g extract) followed by Attikka (180.04 ± 5.09 mg gallic acid eq/g extract). Dan recorded the highest TFC (13.56 ± 0.26 mg quercetin/g of extract). Keena and Madan showed the lowest TPC and TFC respectively.

4. Conclusions, implications and significance

Overall results suggested that *Calophyllum calaba* L, *Syzygium cumini* L, *Drypetes sepiaria*, *Syzygium caryophyllatum* L, *Ficus racemose* L, and

Careya arborea fresh wild fruits possess marked antioxidant properties such as free radical scavenging activity, Ferric reducing antioxidant power, oxygen radical absorbance capacity, total polyphenolic and flavanoid content. Out of six underutilized wild fruits, *Careya arborea* and *Calophyllum calaba* L recorded the highest and the lowest antioxidant activity. Further, the present study provides scientific basis of the use of these wild fruits for prevention and management of oxidative stress associated chronic diseases in Sri Lanka.

5. References

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Extended Abstract