

## ANTIOXIDANT POTENTIAL OF SELECTED WHOLE GRAIN CEREALS CONSUMED BY SRI LANKANS: A COMPARATIVE *IN VITRO* STUDY



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### Abstract

Whole grains of rice [improved: Bw 361 (red) and Bg 359 (white)]; traditional: Kalu Heeneti (red) and Suwadal (white)], corn, finger millet, wheat, barley and oat were used in this study. Ethanolic extracts (EEs) and methanolic extracts (MEs) of whole grain cereals were evaluated for total polyphenolic content (TPC), ferric reducing antioxidant power (FRAP), oxygen radical absorbance capacity (ORAC), DPPH and ABTS radical scavenging antioxidant properties (AP) *in vitro* (n=3 each). Results showed significant differences (p<0.05) between extracts and among cereals for investigated AP. In general, MEs of cereals showed significantly higher (p<0.05) activity compared to EEs of cereals. Among the studied cereals, finger millet exhibited the highest AP in both extracts for TPC, FRAP and ORAC while for DPPH and ABTS it's only in MEs. The mean TPC, FRAP and ORAC of MEs of finger millet were 708.45 ± 11.72 mg gallic acid equivalents (GAE), 1413.69 ± 1.61, 1240.45 ± 67.38 mg Trolox equivalents (TE) and 955.76 ± 2.03 mg TE/100 g grain, respectively while for EEs it was 275.36 ± 4.26 mg GAE, 606.34 ± 5.00 mg TE and 1112.52 ± 24.38 TE/100 g grain respectively. For DPPH and ABTS radical scavenging activities the IC<sub>50</sub> values of MEs of finger millet were 49.42 ± 6.65 and 10.74 ± 0.44 µg/mL respectively. Of the cereals analyzed the second highest AP was observed in traditional red rice variety, Kalu Heeneti (except for ORAC) for both extracts tested. Whole grains of wheat and oats had very low AP compared to finger millet and red rice varieties analyzed in the present study. It is concluded that finger millet possesses highest AP.

**Key words:** Antioxidant properties, whole grain cereals, traditional and improved rice, wheat, oat, barley, corn and finger millet

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## 1. Introduction

Excessive generation of free radicals leads to a condition known as oxidative stress (Birben *et al.*, 2012; Rahman *et al.*, 2012) and it is linked to the development and progression of several degenerative diseases such as cancer, diabetes, heart diseases, neurological diseases, inflammatory diseases and ageing (Krishnaiah *et al.*, 2011; Birben *et al.*, 2012; Rahman *et al.*, 2012). Antioxidants are compounds which have the ability to reduce the oxidative stress that is one of the causes that leads to the above mentioned degenerative diseases (Birben *et al.*, 2012; Rahman *et al.*, 2012). Hence, diets rich in antioxidants play a crucial role in the prevention and management of such diseases.

Cereals and cereal-based foods have been the basis of the human diet since ancient times. Current dietary guidelines all over the world recommend the consumption of whole grain cereals as they have the ability to enhance health beyond the basic nutrition (Jones, 2006; Dykes & Rooney, 2007; Okarter & Liu, 2010). The health benefits of whole grain cereals are attributed to the presence of variety of phytochemicals including naturally derived antioxidants (Muntana & Prasong, 2010; Zhang *et al.*, 2010; Laokuldilok *et al.*, 2011; Sompong *et al.*, 2011; Jun *et al.*, 2012; Pengkumsri *et al.*, 2015).

Typical Sri Lankan diet comprises of variety of cereals. Rice is the principle cereal and the dietary staple food for Sri Lankans (Rajapakse *et al.*, 2000; Gunaratne *et al.*, 2013). There are wide varieties of new improved (NI) and especially traditional varieties available in the country (Rajapakse *et al.*, 2000). Some

of the traditional varieties were well known for its health benefits (Dharmasena, 2010; Abeysekera & Premakumara, 2016). Therefore, such varieties still socially acceptable and currently has a good market value. Apart from rice, wheat, finger millets, corn, barley and oats are cereals popular among Sri Lankans. These cereals are consumed mainly as processed refined cereals.

Antioxidant properties of bran, whole and milled grains of some of the commonly consumed cereals in Sri Lanka have been reported (Gunaratne *et al.*, 2013; Premakumara *et al.*, 2013). However, comparison of antioxidant properties of commonly consumed whole grain cereals grown in Sri Lanka and those imported are not reported. The present study reports the comparison of the total polyphenolic content of commonly consumed whole grain cereals in Sri Lanka using Folin-Ciocalteu reagent and the antioxidant properties determined as total reducing capacity by Ferric Reducing Antioxidant Power (FRAP), physiological radical scavenging activity by using peroxy radical by Oxygen Radical Absorbance Capacity (ORAC) as well as non-physiological radical activities by using DPPH Radical and ABTS Radical Scavenging (Phillips *et al.*, 2007; Muntana & Prasong, 2010; Zhang *et al.*, 2010; Laokuldilok *et al.*, 2011; Sompong *et al.*, 2011, Gupta, 2015). Determinations were carried out in the methanolic and ethanolic extracts that have been used worldwide to evaluate the antioxidant properties of cereals (Farrar *et al.*, 2008; Muntana & Prasong, 2010; Zhang *et al.*, 2010; Laokuldilok *et al.*, 2011; Sompong *et al.*, 2011; Premakumara *et al.*, 2013).

## 2. Material and Methods

### 2.1 Cereal samples

Two red *Oryza sativa* (rice) varieties (new improved: Bw 361; traditional: Kalu Heeneti) and two white rice varieties (new improved: Bg 359; traditional: Suwadal) collected from the Rice Research and Development Institute (RRDI), Batalagoda, Sri Lanka and *Eleusine coracana* (finger millet), *Zea mays* (corn), *Triticum aestivum* (wheat), *Avena sativa* (oat) and *Hordeum vulgare* (barley) were collected from the local market, Sri Lanka. These cereals were collected from several traders and pooled random samples were used in the study.

### 2.2 Chemicals and reagents

2,2'-Azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS), 2,2'-azobis (2-amidino-propane) dihydrochloride (AAPH), 1,1-diphenyl-2-picrylhydrazine (DPPH), 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox), gallic acid, 2,4,6-tripyridyl-s-triazine (TPTZ), potassium persulfate, ferric chloride, fluorescein, and Folin-Ciocalteu phenol reagent were purchased from Sigma-Aldrich, USA. All the other chemicals used for the preparation of buffers and solvents were of analytical grade.

### 2.3 Sample preparation

Whole grain cereals were milled (Fritsch, Pulverisette 14, Germany) and passed through 0.5 mm sieve. Cereal flour (1 g) was shaken over night at room temperature ( $28 \pm 2$  °C) with 20 times the sample weight of ethanol and methanol separately. Extracts were then centrifuged (825 g) for 10 min, filtered

through 0.45  $\mu$ m nylon filters and evaporated to dryness under vacuum in a rotary evaporator and used in the determination of antioxidant properties.

### 2.4 Equipment used

Absorbance readings were measured using a 96 well micro plate reader (SpectraMax Plus 384, Molecular Devices, USA).

### 2.5 Total polyphenolic content

Total polyphenolic content (TPC) of cereal extracts were determined using the Folin-Ciocalteu reagent as described by Singleton *et al.* (1999). Twenty microliters of cereal extracts (2 mg/mL) was added to 110  $\mu$ L of ten times diluted freshly prepared Folin-Ciocalteu reagent. Seventy microliters of sodium carbonate solution was added to the mixture and incubated at room temperature ( $25 \pm 2$  °C) for 30 min. The absorbance was recorded at 765 nm using gallic acid as the standard and TPC was expressed as mg gallic acid equivalents (GAE)/100 g dry weight of the whole grain cereal flour.

### 2.6 Ferric reducing antioxidant power

Ferric reducing antioxidant power (FRAP) of cereal extracts was performed according to the method described by Benzie & Szeto (1999). The working FRAP reagent was prepared by mixing 300 mM acetate buffer (pH 3.6), 10 mM TPTZ solution in 40 mM HCl and 20 mM  $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$  in a ratio 10:1:1. The mixture was incubated at 37 °C for 10 min in a shaking water bath. A reaction volume of 200  $\mu$ L, containing 150  $\mu$ L of working FRAP reagent, 30  $\mu$ L acetate buffer and 10  $\mu$ L (2 mg/mL) of sample were incubated at room temperature ( $25 \pm 2$  °C)

for 8 min. The absorbance was recorded at 600 nm using Trolox as the standard and results were expressed as mg Trolox equivalents (TE)/100 g dry weight of the whole grain cereal flour.

### 2.7 DPPH radical scavenging activity

The DPPH radical scavenging activity of cereal extracts was performed according to the method described by Blois (1958). A reaction volume of 200  $\mu$ L, containing 125  $\mu$ M of DPPH radical and 5  $\mu$ L (2 mg/mL) of sample were incubated at 25  $\pm$  2  $^{\circ}$ C for 10 min and the absorbance values were recorded at 517 nm. For dose response studies 3.12, 6.25, 12.5, 25, 50, 100 and 200  $\mu$ g/mL concentrations of selected extracts were used. Trolox was used as the standard antioxidant. DPPH radical scavenging activity (inhibition %) of each cereal extract and Trolox were calculated using the following equation. Results were expressed as % inhibition and IC<sub>50</sub> values.

$$\text{DPPH radical scavenging activity (\%)} = [(A_c - A_s) / A_c] * 100$$

where,  $A_c$  is the absorbance of the control and  $A_s$  is the absorbance of the sample.

### 2.8 ABTS<sup>+</sup> radical scavenging activity

The ABTS<sup>+</sup> radical scavenging activity of cereal extracts were determined according to the method described by Re *et al.* (1999). A stable stock solution of ABTS<sup>+</sup> radical was produced by reacting 7.8 mM of ABTS<sup>+</sup> in potassium persulphate at 37  $^{\circ}$ C for 16 h in dark. A reaction volume of 200  $\mu$ L, containing 40  $\mu$ L of seven times diluted ABTS stock solution, 150  $\mu$ L phosphate buffer and 5  $\mu$ L (2 mg/mL) of sample (n=3) was incubated at 25  $\pm$  2  $^{\circ}$ C for 10 min and the

absorbance values were recorded at 734 nm. Selected cereal extracts were studied for dose response using series of concentrations (1.56, 3.12, 6.25, 12.5, 25 and 50  $\mu$ g/mL; n=3). Trolox was used as the standard. ABTS<sup>+</sup> radical scavenging activity (inhibition %) of each cereal extract and Trolox were calculated using the following equation. Results were expressed as % inhibition and IC<sub>50</sub> values.

$$\text{ABTS radical scavenging activity (\%)} = [(A_c - A_s) / A_c] * 100$$

where,  $A_c$  is the absorbance of the control and  $A_s$  is the absorbance of the sample.

### 2.9 Oxygen radical absorbance capacity

The oxygen radical absorbance capacity (ORAC) of cereal extracts was carried out according to the method described by Ou *et al.* (2001). Trolox (0.75 and 1.5  $\mu$ g/mL), fluorescein (4.8  $\mu$ M) and AAPH (40 mg/mL) solutions were prepared in 75 mM phosphate buffer (pH 7.4). A reaction volume of 200  $\mu$ L, containing 100  $\mu$ L of fluorescein and 50  $\mu$ L of sample (mg/mL) were pre-incubated at 37  $^{\circ}$ C for 10 min. 50  $\mu$ L of AAPH was added to initiate the reaction and decay of fluorescein was measured at excitation and emission wave lengths of 494 nm and 535 nm respectively at 1 min interval for 35 min using a fluorescent micro plate reader (SPECTRAMax- Gemini EM, Molecular Devices Inc, USA). Trolox was used as the standard antioxidant. ORAC activities of the samples were calculated by comparing the net area under curve of fluorescein decay between the control and the samples using the following equation. Results were expressed as mg TE/100 g dry weight of the whole grain cereal flour.

$$\text{ORAC} = \frac{[(\text{AUC}_s - \text{AUC}_c) / (\text{AUC}_t - \text{AUC}_c)] * (\text{Trolox concentration} / \text{Sample concentration})$$

where,  $\text{AUC}_s$  is the area under the curve of the sample,  $\text{AUC}_c$  is the area under the curve of the control and  $\text{AUC}_t$  is the area under the curve of the Trolox.

### 2.10 Statistical analysis

Data of each experiment were statistically analyzed using SAS version 6.12. One way analysis of variance (ANOVA) and the Duncan's Multiple Range Test (DMRT) were used to determine the differences among treatment means. The Pearson's correlation coefficient was used for the correlation analysis.  $P < 0.05$  was regarded as significant.

## 3 Results

### 3.1 Total polyphenolic content

TPC of methanolic and ethanolic extracts of selected whole grain cereals is given in Table 1. Significant differences ( $p < 0.05$ ) were observed among the cereals and between methanolic and ethanolic extracts of cereals for TPC. Methanolic extracts of cereals exhibited significantly higher ( $p < 0.05$ ) TPC than ethanolic extracts of cereals except for extracts of Bg 359 (new improved white rice). The order of potency of methanolic extracts of cereals for TPC were finger millet > Bw 361 = Kalu Heeneti = Suwadal > barley > corn = wheat > Bg 359 > oat while for ethanolic extracts of cereals it was finger millet > Kalu Heeneti > Bw 361 > Suwadal = wheat = barley = 359 = corn > oat. Among the cereals studied finger millet

exhibited the highest TPC while oat had the lowest for both extracts studied.

### 3.2 Ferric reducing antioxidant power

FRAP of methanolic and ethanolic extracts of cereals is given in Table 2. Results demonstrated significant differences ( $p < 0.05$ ) between methanolic and ethanolic extracts and among the cereal extracts for FRAP. Methanolic extracts of cereals exhibited significantly high ( $p < 0.05$ ) FRAP compared to ethanolic extracts of cereals except for the extract of new improved white rice variety Bg 359. Among the cereals tested finger millet showed the highest FRAP while oat had the lowest for the both tested extracts. The order of potency of methanolic extracts of cereals for FRAP was finger millet > Kalu Heeneti > Bw 361 > Suwadal > barley > wheat > corn > Bg 359 = oat. For ethanolic extracts of cereals it was observed as finger millet > Kalu Heeneti > Bw 361 > Suwadal > Bg 359 > barley > wheat > corn > oat.

### 3.3 DPPH radical scavenging activity

DPPH radical scavenging activity of methanolic and ethanolic extracts of cereals is given in Table 3. Significant differences ( $p < 0.05$ ) were observed between methanolic and ethanolic extracts and among the cereal extracts for DPPH radical scavenging activity. Methanolic extracts of cereals exhibited significantly high ( $p < 0.05$ ) DPPH radical scavenging activity compared to ethanolic extracts of cereals except for the extracts of 2 red rices Kalu Heeneti and Bw 361 and white rice variety Suwadal. The order of potency of methanolic extracts of cereals for DPPH radical scavenging

activity was finger millet > Kalu Heeneti > barley > wheat = Bw 361 > oat = corn = Bg 359 > Suwadal while for ethanolic extract it was finger millet = Kalu Heeneti = Bw 361 > barley > Suwadal = Bg 359 = oat = corn = wheat. The cereal extracts which showed highest inhibitory activities at screening were studied for dose response relationship and all selected cereal extracts had significant ( $p < 0.05$ ) and dose dependant DPPH radical scavenging activity. The  $IC_{50}$  values of studied extracts varied from  $49.42 \pm 6.65$  -  $137.18 \pm 7.92$   $\mu\text{g/mL}$ . The methanolic extract of finger millet exhibited the highest DPPH radical scavenging activity while ethanolic extract of finger millet showed the lowest. The second highest activity was observed for Kalu Heeneti, a traditional red rice variety. The dose response relationship of selected cereal extracts for DPPH radical scavenging activity is given in Figure 1.

### 3.4 ABTS<sup>+</sup> radical scavenging activity

ABTS radical scavenging activity of methanolic and ethanolic extracts of cereals is presented in Table 4. ABTS radical scavenging activity between methanolic and ethanolic extracts and among the cereal extracts showed significant differences ( $p < 0.05$ ). Methanolic extracts showed highest activities for the cereal extracts of finger millet, Bw 361 (red rice), corn and oat while cereal extracts of Suwadal (white rice) and barley demonstrated highest activities for ethanolic extracts. Comparable ABTS radical scavenging activities were observed for the cereal extracts of Kalu Heeneti (red rice), Bg 359 (white rice) and wheat. Finger millet showed the highest ABTS radical scavenging activity while oat had the lowest for both extracts tested.

**Table 1.** Total polyphenolic content of methanolic and ethanolic extracts of cereals

Cereal	Total polyphenolic content (TPC)	
	Methanolic extract	Ethanolic extract
Finger millet	$708.45 \pm 11.72^a$	$275.36 \pm 4.26^a$
Red rice (New improved: Bw 361)	$139.79 \pm 0.00^b$	$99.23 \pm 8.32^c$
Red rice (Traditional: Kalu Heeneti)	$129.86 \pm 7.37^{bc}$	$113.56 \pm 1.79^b$
White rice (Traditional: Suwadal)	$116.90 \pm 2.84^c$	$78.60 \pm 1.96^d$
Barley	$93.61 \pm 1.21^d$	$68.69 \pm 1.88^{de}$
Corn	$75.10 \pm 0.00^e$	$52.10 \pm 1.23^f$
Wheat	$71.22 \pm 1.18^e$	$78.05 \pm 6.46^d$
White rice (New improved: Bg 359)	$29.34 \pm 1.70^f$	$59.78 \pm 2.28^{ef}$
Oat	$17.24 \pm 0.73^g$	$15.72 \pm 0.77^g$

Data presented as mean  $\pm$  SE (n=3). Mean values in a column superscripted by different letters are significantly different at  $p < 0.05$ . TPC: mg Gallic acid equivalents/100 g whole grain flour in dry weight basis.

**Table 2.** Ferric reducing antioxidant power of methanolic and ethanolic extracts of cereals

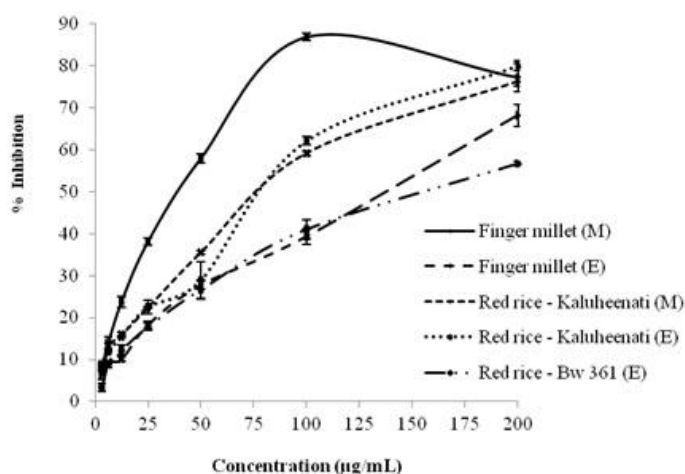
Cereal	Ferric reducing antioxidant power (FRAP)	
	Methanolic extract	Ethanolic extract
Finger millet	1413.69 ± 1.61 <sup>a</sup>	606.34 ± 5.00 <sup>a</sup>
Red rice (Traditional: Kalu Heeneti)	743.18 ± 2.50 <sup>b</sup>	255.34 ± 4.05 <sup>b</sup>
Red rice (New improved: Bw 361)	351.86 ± 4.60 <sup>c</sup>	216.59 ± 0.55 <sup>c</sup>
White rice (Traditional: Suwadal)	252.62 ± 7.65 <sup>d</sup>	166.89 ± 1.47 <sup>d</sup>
Barley	199.45 ± 0.83 <sup>e</sup>	107.21 ± 0.47 <sup>f</sup>
Wheat	177.95 ± 14.70 <sup>f</sup>	84.27 ± 2.63 <sup>g</sup>
Corn	81.06 ± 7.64 <sup>g</sup>	45.09 ± 1.34 <sup>h</sup>
White rice (New improved: Bg 359)	61.79 ± 8.19 <sup>h</sup>	139.36 ± 3.27 <sup>e</sup>
Oat	61.74 ± 2.24 <sup>h</sup>	26.60 ± 2.0 <sup>i</sup>

Data presented as mean ± SE (n=3). Mean values in a column superscripted by different letters are significantly different at  $p < 0.05$ . FRAP: mg Trolox equivalents/100 g whole grain flour in dry weight basis. ND: Not detected.

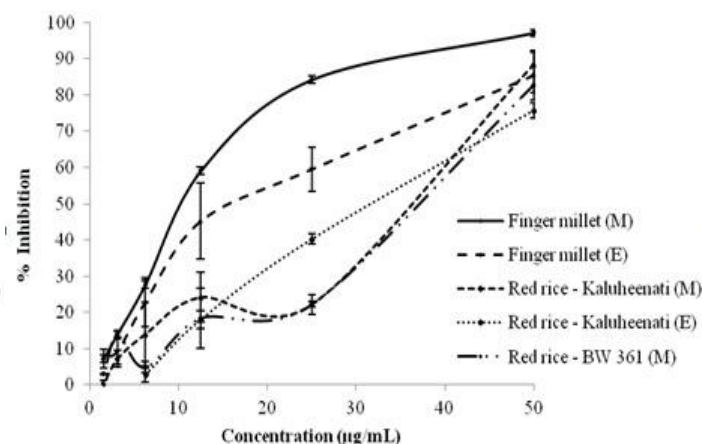
**Table 3.** DPPH radical scavenging activity of methanolic and ethanolic extracts of cereals

Cereal	DPPH radical scavenging activity (% inhibition)	
	Methanolic extract	Ethanolic extract
Finger millet	67.05±1.24 <sup>a</sup>	39.97±0.27 <sup>a</sup>
Red rice (Traditional: Kalu Heeneti)	25.73±1.13 <sup>b</sup>	38.69±1.68 <sup>a</sup>
Barley	22.19±1.09 <sup>c</sup>	13.85±4.33 <sup>b</sup>
Wheat	19.73±0.83 <sup>d</sup>	1.49±2.76 <sup>c</sup>
Red rice (New improved: Bw 361)	19.04±1.00 <sup>d</sup>	32.06±2.19 <sup>a</sup>
Oat	16.33±1.41 <sup>e</sup>	4.93±5.31 <sup>c</sup>
Corn	15.98±0.98 <sup>e</sup>	2.19±4.88 <sup>c</sup>
White rice (New improved: Bg 359)	14.56±0.44 <sup>e</sup>	8.37±3.42 <sup>bc</sup>
White rice (Traditional: Suwadal)	0.60±0.30 <sup>f</sup>	8.78±2.28 <sup>bc</sup>

Data presented as mean ± SE (n=3). Mean values in a column superscripted by different letters are significantly different at  $p < 0.05$ . DPPH radical scavenging activity: % inhibition at 50 µg/mL.



**Figure 1.** Dose response relationship of ethanolic and methanolic extracts of selected cereals for DPPH radical scavenging activity. E: Ethanolic extract; M: Methanolic extract; IC<sub>50</sub> values (µg/mL): Finger millet (M): 49.42 ± 6.65; Finger millet (E): 137.18 ± 7.92; Kalu Heeneti (M): 80.96 ± 0.61; Kalu Heeneti (E): 80.84 ± 2.55; Bw 361 (E): 105.71 ± 0.77.



**Figure 2.** Dose response relationship of ethanolic and methanolic extracts of selected cereals for ABTS<sup>+</sup> radical scavenging activity. E: Ethanolic extract; M: Methanolic extract; IC<sub>50</sub> values (µg/mL): Finger millet (M): 10.74 ± 0.44; Finger millet (E): 37.16 ± 1.15; Kalu Heeneti (M): 32.48 ± 1.40; Kalu Heeneti (E): 28.04 ± 2.63; Bw 361 (M): 33.59 ± 0.64.

**Table 4.** ABTS radical scavenging activity of methanolic and ethanolic extracts of cereals

Cereal	ABTS radical scavenging activity (% inhibition)	
	Methanolic extract	Ethanolic extract
Finger millet	100.47±0.19 <sup>a</sup>	88.52±4.32 <sup>a</sup>
Red rice (Traditional: Kalu Heenati)	82.62±0.59 <sup>b</sup>	83.04±3.36 <sup>a</sup>
Red rice (New improved: Bw 361)	71.25±0.49 <sup>c</sup>	56.87±8.38 <sup>b</sup>
White rice (Traditional: Suwadal)	36.12±4.48 <sup>e</sup>	43.60±1.85 <sup>c</sup>
Barley	34.97±1.68 <sup>e</sup>	63.24±1.96 <sup>b</sup>
White rice (New improved: Bg 359)	45.87±0.95 <sup>d</sup>	40.44±3.19 <sup>c</sup>
Wheat	27.54±1.84 <sup>f</sup>	25.83±1.30 <sup>d</sup>
Corn	21.12±2.79 <sup>g</sup>	8.48±0.45 <sup>e</sup>
Oat	23.64±2.79 <sup>fg</sup>	11.01±0.95 <sup>e</sup>

Data presented as mean ± SE (n=3). Mean values in a column superscripted by different letters are significantly different at  $p < 0.05$ . ABTS radical scavenging activity: % inhibition at 50 µg/mL.



**Table 5.** Oxygen radical absorbance capacity of ethanolic and methanolic extracts of cereals

Cereal variety	Oxygen radical absorbance capacity (ORAC)	
	Methanol extract	Ethanolic extract
Finger millet	1240.45 ± 67.38 <sup>a</sup>	1112.52 ± 24.38 <sup>a</sup>
White rice (Traditional: Suwadal)	439.64 ± 16.24 <sup>b</sup>	272.08 ± 1.70 <sup>b</sup>
Barley	357.51 ± 17.64 <sup>bc</sup>	244.33 ± 9.40 <sup>b</sup>
Red rice (New improved: Bw 361)	323.28 ± 2.06 <sup>cd</sup>	185.09 ± 7.57 <sup>c</sup>
Wheat	322.00 ± 21.77 <sup>cd</sup>	263.90 ± 14.41 <sup>b</sup>
Red rice (Traditional: Kalu Heeneti)	288.84 ± 12.07 <sup>cd</sup>	108.32 ± 3.16 <sup>d</sup>
Corn	249.91 ± 9.61 <sup>d</sup>	84.04 ± 13.22 <sup>d</sup>
White rice (New improved: Bg 359)	150.61 ± 14.32 <sup>e</sup>	180.43 ± 0.73 <sup>c</sup>
Oat	50.71 ± 7.56 <sup>f</sup>	73.20 ± 3.26 <sup>d</sup>

Data presented as mean ± SE (n=3). Mean values in a column superscripted by different letters are significantly different at  $p < 0.05$ . ORAC: mg Trolox equivalents/100 g grain dry weight. ND: Not detected.

studied for dose response relationship and results showed significant ( $p < 0.05$ ) and dose dependant ABTS radical scavenging activity for all the studied cereal extracts. The  $IC_{50}$  values of cereal extracts varied from  $10.74 \pm 0.44$  -  $37.16 \pm 1.15$   $\mu\text{g}/\text{mL}$ . The methanolic extract of finger millet exhibited the highest ABTS radical scavenging activity while ethanolic extract of finger millet showed the lowest. Dose response relationship of selected cereal extracts for ABTS radical scavenging activity is given in Figure 2.

### 3.5 Oxygen radical absorbance capacity

ORAC of methanolic and ethanolic extracts of cereals is given in Table 5. ORAC was significantly different ( $p < 0.05$ ) between methanolic and ethanolic extracts and among the different cereal extracts studied. Results showed significantly high ( $p < 0.05$ ) ORAC in

improved white rice extract of Bg 359 and oat. Among the cereals tested finger millet showed the highest ORAC while oat had the lowest for the both extracts tested. The order of potency of methanolic extracts of cereals for ORAC was finger millet > Suwadal = barley = Bw 361 = wheat = Kalu Heeneti = corn > Bg 359 > oat. For ethanolic extracts of cereals it was observed as finger millet > Suwadal = wheat = barley > Bw 361 = Bg 359 > Kalu Heeneti = corn = oat.

## 4 Discussion

The traditional rice varieties selected for the study was based on the health claims in the Sri Lankan traditional knowledge and the improved varieties selected were widely cultivating rice varieties in the country. The rest of the cereals studied were the commonly consumed cereals by

Sri Lankans. Present study showed that finger millet had the highest antioxidant activities for all the assays tested (TPC, FRAP and ORAC: for both extracts; DPPH and ABTS: only for the methanolic extract). Antioxidant activities of different varieties of finger millets are reported worldwide and it has been shown that antioxidant activity vary depending on the variety (Sreeramulu *et al.*, 2009; Dykes & Rooney, 2007). Phenolic compounds are reported as main antioxidant compounds in finger millets (Chandrasekara & Shahidi, 2011a; Chandrasekara & Shahidi, 2011b; Kumari *et al.*, 2016). TPC of the Sri Lankan finger millet variety tested had more TPC than the TPC of the finger millet varieties studied by Sreeramulu *et al.* (2009) and Dykes & Rooney (2007). This might be due to the varietal difference and/or the extraction methods used. TPC as well as the antioxidant properties measured as DPPH, ABTS and ORAC assays for finger millet varieties from Sri Lanka studied by Chandrasekara & Shahidi (2011a), Chandrasekara & Shahidi (2011b) and Kumari *et al.* (2016) cannot be compared with those of the present study due to the differences in variety and extraction procedures and the results have been expressed for defatted meal and not for the whole grain. A limitation in the present study is that the variety cannot be deduced as a market sample was used.

Of the cereals analyzed second highest antioxidant activities were observed for red rice varieties (except for ORAC). Interestingly, traditional red rice variety Kalu Heeneti had more antioxidant activity than the most popular new improved red rice variety Bw 361. Further, traditional white rice variety

Suwadal also showed comparatively high antioxidant activity (except DPPH) compared to the other cereals tested. Traditionally both Kalu Heeneti and Suwadal are known for variety of health benefits including immune enhancing activity (Dharmasena, 2010; Abeysekera & Premakumara, 2016). This study showed that whole grains of both varieties are better sources of naturally derived antioxidants and thus findings of this study scientifically validated some health claims in the Sri Lankan traditional knowledge. Further, the results of this study revealed that wheat, oats and white rice variety (Bg 359) popularly consumed in Sri Lanka had very low antioxidant activities in contrast to red rice varieties Kalu Heeneti and Bw 361 and finger millets. Consumption of wheat and white rice (Bg 359) in the country is mainly as refined cereals. Thus, antioxidant activities at refined levels may be much less than the findings of the present study. Therefore, it is important to highlight that regular consumption of such refined cereals may lead to less intake of naturally derived antioxidants. Whole grains are rich sources of dietary fiber, vitamins and minerals. Hence, the results of the present study show that regular consumption of whole grains of finger millets, Kalu Heeneti, Bw 361 and Suwadal could be advantages in prevention and dietary management of age-related diseases such as diabetes, cancers, cardiovascular diseases, neurological diseases and inflammatory diseases (Jones, 2006; Dykes & Rooney, 2007; Okarter & Liu, 2010).

Pair-wise correlations between total phenolic content and antioxidant properties of both ethanolic (correlation

coefficients of TPC between ABTS, DPPH ,ORAC and FRAP were 0.75, 0.73, 0.91 and 0.97 respectively) and methanolic (correlation coefficients of TPC between ABTS, DPPH ,ORAC and FRAP were 0.70, 0.87, 0.95 and 0.96 respectively) extracts of cereals studied showed significant positive correlations ( $P < 0.05$ ) indicating that phenolic compounds have vital roles in mediating antioxidant properties in cereals. Our findings are in agreement with the findings of Zhang *et al.* (2010) and Sompong *et al.* (2011).

Considering all, this study clearly showed that whole grains of finger millet and selected red rices are good sources of naturally derived antioxidants. Thus, regular consumption of whole grains of millets and red rices especially traditional red rices may be important in prevention and dietary management of such degenerative diseases. Further, findings also showed that in general antioxidant activities of cereal extracts are higher in methanolic extracts in contrast to ethanolic extracts indicating more compounds are extracted to methanolic phase rather than ethanolic phase.

## 5 Conclusion

From the results of this study it can be concluded that whole grains of finger millet possesses highest antioxidant properties followed by red rice varieties (Bw 361, Kalu Heeneti) studied while whole grains of oats, wheat and new improved white rice variety (Bg 359) had less activity. In general methanol is a better solvent than ethanol for extracting compounds responsible for antioxidant activities in cereals. Therefore,

consumption of whole grain finger millets, Kalu Heeneti and Bw 361 may play an important role in prevention and dietary management of oxidative stress associated chronic diseases.

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