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Nutritional value, phytochemicals and antioxidant properties of two wild edible fruits (*Eugenia operculata* Roxb. and *Antidesma bunius* L.) from Assam, North-East India

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Abstract. The purpose of present study was to investigate the nutritional composition, phytochemical contents and antioxidant capacities of two wild edible fruits *viz*. *Eugenia operculata* Roxb. and *Antidesma bunius* L. from Assam of North-East India. The fruits showed variable amounts of proximate and mineral compositions which are reported herein and discussed. The phytochemical screening of different solvent extracts exhibited the presence of many phytochemicals which are biologically important. The antioxidant activities were examined using DPPH (1, 1-diphenyl-2-picrylhydrazyl), ABTS (2, 2′-Azinobis (3-ethylbenothiazoline-6-sulfonic acid) diammonium salt), H_2O_2 (Hydrogen peroxide) and FRAP (Ferric reducing antioxidant power) assays. The fruits showed antioxidant properties with DPPH IC_{50} value of 92.330 ± 4.163 μg/mL (*E. operculata*) and 395.002 ± 3.605 μg/mL (*A. bunius*), ABTS IC_{50} value of 52.660 ± 1.154 μg/mL (*E. operculata*) and 105.331 ± 3.055 μg/mL (*A. bunius*), H_2O_2 IC_{50} value of 20.566 ± 0.208 μg/mL (*E. operculata*) and 24.366 ± 0.057 μg/mL (*A. bunius*), and FRAP value of 281.583 ± 8.799 μM TE/g DE (dry extract) in *E. operculata* and 61.583 ± 3.818 μM TE/g DE in *A. bunius*. The total phenolic content (TPC) in *E. operculata* and *A. bunius* fruits were found to be 226.741 ± 2.099 mg GAE/g DE and 119.356 ± 1.395 mg GAE/g DE, respectively, while the total flavonoid content (TFC) were 108.761 ± 7.015 mg QE/g DE and 64.323 ± 8.828 mg QE/g DE, respectively. The fresh fruits of *A. bunius* and *E. operculata* were found to contain vitamin C of 7.30 ± 1.452 mg/100 g and 6.60 ± 1.123 mg/100 g, respectively. The studies revealed that *E. operculata* fruit had stronger antioxidant activity than *A. bunius* fruit showing better DPPH, ABTS and H_2O_2 scavenging activities, and higher FRAP value, TPC and TFC. TPC and TFC showed a strong positive correlation with antioxidant activity assayed by FRAP. A strong positive correlation of antioxidant activity (DPPH, ABTS and H_2O_2) was also found with vitamin C.

Keywords: Wild edible fruits, *Eugenia operculata*, *Antidesma bunius*, proximate, minerals, phytochemicals, antioxidant

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1. Introduction

Wild edible fruits are excellent sources of nutrients and used as an important food items in the diets of indigenous people and consequently, these fruits are making an important contribution to the health of local communities. Wild fruits contain numerous natural antioxidant compounds such as carotenoids, vitamins, phenolic compounds, and many more which have the capability to scavenge reactive oxygen species such as free radicals, inhibit the oxidation process and slow down the growth of various human diseases like cancer, heart disease, diabetes, stroke, rheumatoid arthritis, obesity, cataracts, and Alzheimer's disease [1–3]. Antioxidants include both natural and synthetic antioxidants. The free radicals generated in the body due to various reasons are responsible for the damage of nucleic acids, proteins, and lipids in the cells that lead to various human diseases and the plant based natural antioxidants can prevent this to happening by scavenging this active radicals [4]. It was reported that plant products are also potential sources of skin-whitening compounds in cosmetic and medicinal fields [5]. Synthetic antioxidants have widely been used in the food industry as food additives to protect food items against oxidative degradation and to prolong the shelf-life, but some of the synthetic antioxidants are harmful as they possess potential toxicity and carcinogenicity [6, 7]. Fruits, spices, vegetables, medicinal plants and even microalgae have been reported to contain good sources of natural antioxidants [7–10] and the natural antioxidants are considered as potential alternatives to synthetic ones because of their numerous health profits [11]. Hence, the researchers throughout the world are being attracted to explore natural antioxidants from fruits, vegetables and medicinal plants and the search is still increasing.

Eugenia operculata Roxb. is a perennial tree, extensively distributed and propagated in China, Vietnam and some other tropical countries [12]. The plant is found growing in Bihar, Orissa, and Northeast region of India [13]. The plant produces fruits which are eaten by local communities in Assam. Its leaves, bark and buds were studied and showed medicinal and food properties [12, 14, 15]. Devi et al. [13] also studied aqueous and ethanol leave extracts of *E. operculata* and reported that the leave contains different phytochemicals and possess antioxidant property. However, still there is no information available on nutrient and antioxidant properties from the fruits of *E. operculata. Antidesma bunius* (L.) Spreng. is a wild plant belonging to the family Euphorbiaceae and distributed in Thailand, Philippines, and Southeast Asia [16]. The plant has antioxidant, anticancer and antidiabetic properties [17–19]. Its fruit has a sweet-sour taste and is popular among rural populaces. A. *bunius* fruits found in Northeast Thailand are good sources of nutritional and phytochemical constituents [20]. Its fruit extract also showed antibacterial activities [21]. This plant is also found in Assam of Northeast of India and no available information on food properties of its fruit is reported from this region. The aim of this study was to evaluate nutritional, mineral, phenolic and flavonoid contents along with the investigation of antioxidant properties of two wild edible fruits *viz*. *E. operculata* and *A. bunius.*

2. Materials and methods

2.1. Chemicals

DPPH (1, 1-diphenyl-2-picrylhydrazyl), ABTS (2, 2′-Azinobis (3-ethylbenothiazoline-6-sulfonic acid) diammonium salt) and quercetin were obtained from Himedia Laboratories Pvt. Ltd., Mumbai (India), trolox from Sigma Aldrich, Bangalore (India), H_2O_2 (Hydrogen peroxide), ascorbic acid and Folin-Ciocalteu's reagent from Merck, Mumbai (India) and gallic acid from Central Drug House Pvt. Ltd., New Delhi (India).

2.2. Collection and identification of plants

Ripened fruits of *E. operculata* and *A. bunius* were obtained in the month of April, 2015 from the Chirang district of Assam and the plants were identified with the help of Botanical Survey of India, Shillong (Meghalaya).

2.3. Sample preparation

The fresh fruits were washed thoroughly under tap water followed by distilled water and then moisture and vitamin C contents of the fresh fruits were determined on the same day. The remaining fruits were then freeze dried (FD) for 72 h. The dried fruits were pulverized and the powdered materials were kept in a container. For screening of phytochemical constituents, the powdered materials were extracted separately with methanol, hexane, chloroform, acetone and water in 1:10 ratio (w/v), shivered, stored for 72 h, filtered (Whatman No. 1), filtrate evaporated to dryness using Buchi Rotavapor R-215 (Switzerland) and the dry extracts were kept in air-tight containers at 4°C till further analyses.

2.4. Analyses of proximate composition

Proximate compositions were determined following the methods of the Association of Official Analytical Chemists [22]. Total carbohydrate and dry matter were calculated using the methods of James [23]. Calorific value was calculated following the method of Food and Agriculture Organization (FAO) [24].

2.5. Determination of mineral contents

Mineral contents were investigated at Sophisticated Analytical Instrumentation Centre (SAIC), Tezpur University using Atomic Absorption Spectrometer (AAS-ICE 3500, Thermo Scientific, UK). The powder of freeze dried (FD) fruits were digested with concentrated HNO_3. The mineral contents were presented in mg/100 g of FD sample.

2.6. Qualitative phytochemical study

The qualitative phytochemical screening of different solvent extracts of FD fruits was performed using the standard procedures [25, 26].

2.7. Evaluation of antioxidant properties

Using methanol extract of FD fruit, antioxidant activities were determined with an UV-VIS spectrophotometer (Lambda 35, Perkin Elmer, USA) following the previously reported DPPH and ABTS assays [26]. Hydrogen peroxide scavenging activity was investigated with an UV-VIS spectrophotometer (Lambda 35, Perkin Elmer, USA) at 230 nm following the method of Ruch et al. [27]. Ferric reducing antioxidant power (FRAP) value was obtained using the method of Benzie and Strain [28]. The values were presented in μM trolox equivalent (TE)/g of dry extract (DE).

2.8. Investigation of total phenolic content (TPC) and total flavonoid content (TFC)

TPC and TFC were determined from methanol extract of FD fruits using an UV-VIS spectrophotometer (Lambda 35, Perkin Elmer, USA) following the previously reported procedures [26].

2.9. Estimation of vitamin C content

Vitamin C content of the fresh fruits was estimated following the procedure of Suntornsuk et al. [29].

2.10. Statistical analysis

The experimental results were presented as mean of triplicate results ± standard deviation. Microsoft Excel was used for calculation of standard deviations. Statistical analyses of results were performed by the one-way ANOVA *t*-test at $p < 0.05$ using OriginPro 8.5 software (OriginLab Corporation, MA 01060 USA). SPSS 13.0 software was used for the study of Pearson's correlation.

3. Results and discussion

3.1. Proximate composition

The results of proximate composition of two wild edible fruits are shown in Table 1. *A. bunius* had the higher moisture content of 4.53 ± 0.351 g/100 g of FD and 64.466 ± 0.251 g/100 g fresh fruit while *E. operculata* showed moisture content of 3.343 ± 0.004 g/100 g of FD and 52.53 ± 0.404 g/100 g of fresh sample. The moisture content in *A. bunius* is similar to that of *Arbutus pavarii* (68.06 ± 1.65 g/100 g) and *Ficus palmata* (67.82 ± 2.07 g/100 g) [30] and the moisture content of *E. operculata* is also comparable to that of *Melastoma malabathricum* (56.6 ± 0.71 g/100 g) [31]. The ash content which is an index of mineral contents was found to be 0.516 ± 0.003 g in *A. bunius* and 0.343 ± 0.004 g in *E. operculata*. The crude fat content of the fruits investigated were found to be 0.97 ± 0.026 g in *A. bunius* and 1.86 ± 0.02 g in *E. operculata* which is almost similar to the values of *Rosa dumalis, Rosa pulverulenta* and *Rosa canina* reported by Ercisli [32]. The crude protein content in *A. bunius* was 1.231 ± 0.050 g and that in *E. operculata* was 1.323 ± 0.035 g per 100 g of FD which are comparable to the values of *Elaeagnus conferta* fruit reported by Rai et al. [33] and *Ziziphus spinachristi* fruit reported by Feyssa et al. [34]. Higher value of crude fibre was found in *E. operculata* fruit (17.566 ± 0.351 g/100 g) than *A. bunius* fruit (9.433 ± 0.305 g/100 g). Fibre rich diets are essential for digestion and effective removal of waste. Consumption of fruits and vegetables with rich fibres can lower the risk of coronary heart disease, constipation, serum cholesterol, diabetes, hypertension, and breast and colon cancer [35, 36]. The total carbohydrate content was found to be 92.743 ± 0.428 g in *A. bunius* and 93.123 ± 0.084 g in *E. operculata* which is higher than the values reported by Gnansounou et al. [37]. The fruits having good composition of carbohydrates are very nutritious for health products and responsible for their high calorific value [38]. The calorific value of *E. operculata* and *A. bunius* fruits were 394.586 ± 0.025 kcal/100 g and 384.651 ± 1.296 kcal/100 g, respectively which is higher to that of *Grewia sapida* fruit (346.34 ± 0.04 kcal/100 g) presented in the previous report [26].

3.2. Mineral composition

The mineral contents of *E. operculata* and *A. bunius* fruits per 100 g of FD sample are shown in Table 2. The results of mineral analyses in the two wild edible fruits revealed that *E. operculata* had the sodium content of 4.640 ± 0.046 mg and *A. bunius* had 5.377 ± 0.032 mg per 100 g which is comparable to that of *Ziziphus mauritiana* (5.03 mg/100 g) [39]. High content of potassium was found in *A. bunius* (3043.852 ± 6.088 mg/100 g) and *E. operculata* (2219.736 ± 6.659 mg/100 g) which is in agreement with results reported by Saka et al. [40] and Amarteifio et al. [41]. Potassium is one of the most essential and major plant nutrients and foods rich in potassium are generally used for the treatment of rheumatoid arthritis and heart disease [42]. In this study, calcium was found to be 714.820 ± 8.578 mg in *E. operculata* and 787.900 ± 14.182 mg in *A. bunius*. Leterme et al. [43] reported calcium content of *Annona squamosa* L. fruit as 991 mg/100 g which is higher in comparison to this report. The recommended daily intake of calcium for adult ranges from 1000 – 1500 mg. The present study showed the satisfactory amount of magnesium and found to be 172.387 ± 0.517 mg/100 g in *E. operculata* and 250.703 ± 0.251 mg/100 g in *A. bunius*. Magnesium is very important metal for many enzymatic reactions and magnesium deficiency causes various health disorders including asthma, high blood pressure, angina pectoris,

Table 1
Proximate composition of *E. operculata* and *A. bunius* fruits per 100 g of DW

Parameters	*E. operculata*	*A. bunius*
Moisture (g)	3.343 ± 0.004^{a}	4.530 ± 0.351^{b}
	$52.530 \pm 0.404^{*a}$	$64.466 \pm 0.251^{*b}$
Ash (g)	0.343 ± 0.004^{a}	0.516 ± 0.003^{a}
Acid insoluble ash (g)	0.235 ± 0.003^{a}	0.337 ± 0.004^{a}
Acid soluble ash (g)	0.108 ± 0.005^{a}	0.178 ± 0.006^{a}
Crude fat (g)	1.860 ± 0.020^{a}	0.970 ± 0.026^{b}
Crude protein (g)	1.323 ± 0.035^{a}	1.231 ± 0.050^{a}
Crude fiber (g)	17.566 ± 0.351^{a}	9.433 ± 0.305^{b}
Total carbohydrate (g)	93.123 ± 0.084^{a}	92.743 ± 0.428^{b}
Dry matter (g)	96.656 ± 0.025^{a}	95.466 ± 0.351^{b}
Calorific value (kcal)	394.586 ± 0.025^{a}	384.651 ± 1.296^{b}

*Moisture content of fresh fruit; DW, dry weight; The results followed by different letters along a row are significantly different from each other at $p < 0.05$.

Table 2
Mineral contents of *E. operculata* and *A. bunius* fruits

Minerals	*E. operculata* (mg/100 g FD)	*A. bunius* (mg/100 g FD)
Sodium	4.640 ± 0.046^{a}	5.377 ± 0.032^{b}
Potassium	2219.736 ± 6.659^{a}	3043.852 ± 6.088^{b}
Calcium	714.820 ± 8.578^{a}	787.900 ± 14.182^{b}
Magnesium	172.387 ± 0.517^{a}	250.703 ± 0.251^{b}
Iron	8.279 ± 0.033^{a}	7.579 ± 0.015^{b}
Zinc	1.828 ± 0.011^{a}	2.903 ± 0.012^{b}
Copper	1.493 ± 0.051^{a}	1.774 ± 0.060^{a}
Manganese	2.817 ± 0.020^{a}	7.616 ± 0.023^{b}
Cobalt	0.352 ± 0.050^{a}	0.390 ± 0.019^{a}

FD, freeze dried; The results followed by different letters along a row are significantly different from each other at $p < 0.05$.

cardiac arrhythmias, coronary artery disease, all types of musculoskeletal disorders, mitral valve prolapse, panic disorder, epilepsy, anxiety, chronic fatigue syndrome and psychiatric conditions [37, 39, 42]. The iron detected in *E. operculata* and *A. bunius* fruits were 8.279 ± 0.033 mg and 7.579 ± 0.015 mg per 100 g, respectively which are comparable to that of *Melastoma malabathricum* (8.00 ± 0.19 mg) and *Calamus guruba* (8.50 ± 0.19 mg) reported by Nayak el al. [31]. Iron is required for haemoglobin synthesis in red blood cells which is needed for oxygen transportation to all parts of the body. Iron deficiency causes anemia and immune system dysfunction [26]. The level of copper, zinc and manganese in *A. bunius* were 1.774 ± 0.060 mg, 2.903 ± 0.012 mg and 7.616 ± 0.023 mg, respectively and in *E. operculata* were 1.493 ± 0.051 mg, 1.828 ± 0.011 mg and 2.817 ± 0.020 mg, respectively. Nayak el al. [31] reported copper content of *Careya arborea* as 1.90 ± 0.04 mg which is comparable to the fruits of this study. The zinc level found in *A. bunius* is close to that of the fruits of blackberry (2.30 ± 0.35 mg/100 g), raspberry (2.97 ± 0.1 mg/100 g) and red currant (2.11 ± 0.54 mg/100 g) reported by Plessi et al. [44] and these values are slightly higher to that of *E. operculata*. Copper is an essential element of many enzyme systems such

as cytochrome oxidase, lysyl oxidase and ceruloplasmin, manganese is important for haemoglobin formation and the deficiency of zinc leads to impaired growth and malnutrition [45]. The level of cobalt in *E. operculata* and *A. bunius* fruits were 0.352 ± 0.050 mg/100 g and 0.390 ± 0.019 g/100 g, respectively. Cobalt is an essential part of vitamin B_{12} also known as cyanocobalamin, and its deficiency can cause pernicious anemia. As human body is not capable to synthesize vitamins, the consumption of diets containing these compounds is essential.

3.3. Phytochemical screening

In this study, five types of solvents including both polar and non-polar *viz*. methanol, chloroform, hexane, acetone and water were used to get fruit extracts and to screen the presence of different phytochemicals in the solvent extracts. The results of phytochemical screening of different solvent extracts from the fruits of *E. operculata* (Table 3) and *A. bunius* (Table 4) showed the presence of many phytochemicals. Both the fruits indicated the presence of alkaloids, saponins and carbohydrates (Fehling's test) in all the five extracts. However, *A. bunius* showed the presence of steroids in all the extracts whereas in *E. operculata*, steroids were not detected in acetone extract. Cardiac glycoside in *E. operculata* was not detected in water extract, while in *A. bunius*, it was detected in methanol, chloroform, and acetone extracts. In *E. operculata*, methanol, acetone and water extracts showed positive test for anthraquinones and coumarins while in *A. bunius*, anthraquinone was not detected in chloroform extract and coumarin was not detected in methanol, chloroform and hexane extracts. Positive test for tannins was observed in the acetone extract of *E. operculata* and in the methanol and acetone extracts of *A. bunius*. Chloroform and acetone extracts showed negative results for flavonoids in *E. operculata* and found to be present in all other three extracts. Chloroform and hexane extracts showed negative results for flavonoids in

Table 3
Qualitative phytochemical analysis of *E. operculata* fruit with different solvent extracts

Phytochemical constituents	Test	Methanol	Chloroform	Hexane	Acetone	Water
Alkaloids	Wagner's reagent	+	+	+	+	+
	Dragendroff's reagent	+	+	+	+	+
Saponins	Froth test	+	+	+	+	+
Cardiac glycosides	Keller-Killiani's test	+	+	+	+	–
Steroids (Terpenoids)	Liebermann-Burchard test	+	+	+	–	+
	Salkowski's test	+	+	+	+	–
Anthraquinones	Modified Borntrager's test	+	–	–	+	+
Coumarins		+	–	–	+	+
Phenols	$FeCl_3$ test	+	+	+	+	+
Tannins	Gelatin test	–	–	–	+	–
Flavonoids	Shinoda's test	+	–	+	–	+
Carbohydrates	Molisch's test	+	+	–	+	–
	Fehling's test	+	+	+	+	+
Starch	Iodine test	–	+	+	–	+
Anthocyanins		+	–	+	+	–
Proteins	Ninhydrin test	–	–	+	–	–
	Millon's test	+	–	–	–	+
Phlobatannins		–	–	–	+	+
Lignin		–	+	+	+	+

(+), present; (–), absent.

Table 4
Qualitative phytochemical analysis of *A. bunius* fruit with different solvent extracts

Phytochemical constituents	Test	Methanol	Chloroform	Hexane	Acetone	Water
Alkaloids	Wagner's reagent	+	+	+	+	+
	Dragendroff's reagent	+	+	+	+	+
Saponins	Froth test	+	+	+	+	+
Cardiac glycosides	Keller-Killiani's test	+	+	–	+	–
Steroids (Terpenoids)	Liebermann-Burchard test	+	+	+	+	+
	Salkowski's test	+	+	+	+	–
Anthraquinones	Modified Borntrager's test	+	–	+	+	+
Coumarins		–	–	–	+	+
Phenols	$FeCl_3$ test	+	–	+	–	+
Tannins	Gelatin test	+	–	–	+	–
Flavonoids	Shinoda's test	+	–	–	+	+
Carbohydrates	Molisch's test	+	+	–	+	–
	Fehling's test	+	+	+	+	+
Starch	Iodine test	+	+	+	+	+
Anthocyanins		–	–	+	+	–
Proteins	Ninhydrin test	–	–	+	–	–
	Millon's test	+	–	–	–	+
Phlobatannins		–	–	–	+	+
Lignin		–	+	–	+	+

(+), present; (–), absent.

A. bunius. Chloroform, hexane and water extracts indicated positive results for starch in *E. operculata*, while starch was detected in all the five extracts in *A. bunius*. Methanol, hexane, and acetone extracts were found to contain anthocyanins in *E. operculata* and in *A. bunius*, only hexane and acetone extracts showed the presence of anthocyanins. Both the fruits showed positive results for proteins (Millon's test) in methanol and water extracts and phlobatannins were detected in acetone and water extracts of both the fruits. Lignin was detected in four different solvent extracts except methanol in *E. operculata*, whereas in *A. bunius*, chloroform, acetone and water extracts showed the presence of lignin. Phytochemicals found in the plant materials are known to possess many biologically active compounds and they are responsible for several biological activities such as antioxidant, antimicrobial, antifungal, antiinflammatory, and anticancer activities [8, 26, 46].

3.4. Antioxidant properties

Most of the polyphenol and flavonoid compounds found in plants are soluble in methanol which is a polar solvent and these compounds display various biological properties including the antioxidant activity [8, 26]. In this study, the methanol extracts of *E. operculata* and *A. bunius* FD fruits were investigated for antioxidant activities using DPPH, ABTS, H_2O_2 and FRAP methods. DPPH is a stable radical species with a maximum absorption at 517 nm that can readily undergo scavenging by antioxidants [26]. This assay has been used commonly to investigate the capacity of compounds as free radical scavengers or hydrogen donors and to assess the antioxidant property of food and plant extracts [47]. The DPPH free radical scavenging activities of different concentrations of methanol extracts of *E. operculata* and *A. bunius* fruits, and standard ascorbic acid are presented in Table 5 and it was observed that the radical scavenging capacity increased with concentration of sample. At 500 μg/mL concentration, *E. operculata* fruit extract ($89.651 \pm 0.552\%$) showed higher percentage of inhibition than *A.*

Table 5
DPPH, ABTS and H_2O_2 scavenging activities of methanolic extracts of *E. operculata* and *A. bunius* fruits

	E. operculata	*A. bunius*	Ascorbic acid**
Conc. (μg/mL)		Inhibition (%) of fruits for DPPH assay	
2	17.973 ± 0.642^a	17.481 ± 0.320^a	15.800 ± 0.556^b
5	22.496 ± 0.960^a	18.903 ± 0.475^b	27.100 ± 0.754^c
10	25.636 ± 0.825^a	19.381 ± 0.161^b	36.433 ± 0.702^c
50	61.960 ± 0.187^a	22.936 ± 0.555^b	93.233 ± 0.404^c
100	80.551 ± 1.011^a	29.373 ± 0.636^b	93.600 ± 0.501^c
200	83.421 ± 0.371^a	31.403 ± 0.475^b	94.166 ± 0.550^c
400	85.012 ± 0.642^a	48.441 ± 0.363^b	94.333 ± 0.650^c
500	89.651 ± 0.552^a	61.413 ± 0.398^b	95.066 ± 0.450^c
IC_{50}	92.330 ± 4.163^a	395.002 ± 3.605^b	16.666 ± 2.516^c
		Inhibition (%) of fruits for ABTS assay	
25	28.93 ± 0.351^a	24.771 ± 0.752^b	36.093 ± 0.875^c
50	41.006 ± 0.467^a	36.912 ± 1.193^b	38.520 ± 1.176^c
75	66.033 ± 0.503^a	46.363 ± 0.521^b	55.551 ± 1.023^c
100	78.69 ± 0.739^a	52.483 ± 1.058^b	66.856 ± 0.661^c
150	81.613 ± 0.452^a	63.726 ± 0.295^b	73.506 ± 0.810^c
250	90.170 ± 0.655^a	80.743 ± 0.895^b	79.426 ± 1.168^c
IC_{50}	52.660 ± 1.154^a	105.331 ± 3.055^b	73.666 ± 3.214^c
		Inhibition (%) of fruits for H_2O_2 assay	
5	13.243 ± 0.095^a	5.936 ± 0.145^b	10.410 ± 0.307^c
10	27.631 ± 0.645^a	18.006 ± 0.225^b	27.890 ± 0.160^a
15	36.073 ± 0.315^a	27.203 ± 0.155^b	41.940 ± 0.232^c
20	47.566 ± 0.120^a	32.473 ± 0.110^b	51.451 ± 0.122^c
25	62.196 ± 0.245^a	55.013 ± 0.066^b	60.523 ± 0.281^c
IC_{50}	20.566 ± 0.208^a	24.366 ± 0.057^b	19.766 ± 0.152^c

Conc., concentration; IC_{50} value in μg/mL; **Ascorbic acid was used as standard for DPPH, ABTS, H_2O_2 assays; Results are expressed as mean of 3 replicates ± standard deviation; The results with different letters along a row are significantly different from each other at $p < 0.05$.

bunius fruit extract (61.413 ± 0.398%). While standard ascorbic acid showed 95.066 ± 0.45% inhibition at the same concentration. The IC_{50} values of *A. bunius*, *E. operculata* and ascorbic acid obtained in DPPH assay were 395.002 ± 3.605 μg/mL, 92.330 ± 4.163 μg/mL, and 16.666 ± 2.516 μg/mL respectively. A lower IC_{50} value of sample exhibits higher antioxidant capacity. The IC_{50} value of *M. calabura* fruit reported by Preethi et al. [48] was 90 ± 0.04 μg/mL which is close to that of *E. operculata* fruit. *A. bunius* fruit extract exhibited similar IC_{50} value to that of *Punica granatum* fruit (398.54 ± 47.6 μg/mL) reported by Khomdram et al. [47]. ABTS free radical scavenging activities in methanol extracts of *E. operculata* and *A. bunius* fruits and standard ascorbic acid are shown in Table 5 and this assay also showed antioxidant activities in a concentration-dependent manner. *E. operculata* displayed 90.17 ± 0.655% inhibition at concentration of 250 μg/mL with an IC_{50} value of 52.66 ± 1.154 μg/mL and *A. bunius* showed 80.743 ± 0.895% inhibition at the same concentration with IC_{50} value of 105.331 ± 3.055 μg/mL which indicated that the fruit extract of *E. operculata* had better antioxidant capacity than *A. bunius.* In our previous study, a wild edible fruit (*Grewia sapida*) reported from Assam of North-East India exhibited an ABTS IC_{50} value of 134.33 ± 4.041 μg/mL [26]. H_2O_2 scavenging activities in methanol extracts of *E. operculata* and *A. bunius* fruits are presented in Table 5. The H_2O_2 IC_{50} value for methanolic extract of *E. operculata* fruit was found to be 20.566 ± 0.208 μg/mL and that of *A. bunius* was 24.366 ± 0.057 μg/mL, while the standard ascorbic acid showed an IC_{50} value of 19.766 ± 0.152 μg/mL. Table 6

Table 6
Ferric reducing antioxidant power (FRAP), TPC, TFC and vitamin C content of the fruits

Parameters	*E. operculata*	*A. bunius*
FRAP value (μM TE/g DE)	281.583 ± 8.799^{a}	61.583 ± 3.818^{b}
TPC (mg GAE/g DE)	226.741 ± 2.099^{a}	119.356 ± 1.395^{b}
TFC (mg QE/g DE)	108.761 ± 7.015^{a}	64.323 ± 8.828^{b}
Vitamin C (mg/100 g FW)	6.60 ± 1.123^{a}	7.30 ± 1.452^{b}

DE, dry extract; FW, fresh weight; The results followed by different letters along a row are significantly different from each other at $p < 0.05$.

Table 7
Pearson's correlation coefficients of antioxidant activity (DPPH, ABTS, H_2O_2, FRAP), TPC, TFC and vitamin C in the fruits

	DPPH	ABTS	H_2O_2	FRAP	TPC	TFC	Vitamin C
DPPH	1						
ABTS	1.000^{a}	1					
H_2O_2	1.000^{a}	1.000^{a}	1				
FRAP	-1.000^{a}	-1.000^{a}	-1.000^{a}	1			
TPC	-1.000^{a}	-1.000^{a}	-1.000^{a}	1.000^{a}	1		
TFC	-1.000^{a}	-1.000^{a}	-1.000^{a}	1.000^{a}	1.000^{a}	1	
Vitamin C	1.000^{a}	1.000^{a}	1.000^{a}	-1.000^{a}	-1.000^{a}	-1.000^{a}	1

[a]Correlation is significant at $p < 0.01$.

shows that *E. operculata* fruit (281.583 ± 8.799 μM TE/g DE) had stronger ferric reducing power than *A. bunius* fruit (61.583 ± 3.818 μM TE/g DE). It is interesting to note that all the four assays (DPPH, ABTS, H_2O_2 and FRAP) employed for determination of antioxidant properties revealed that the methanol extract of *E. operculata* fruit exhibited better antioxidant activities than *A. bunius* fruit extract.

3.5. TPC, TFC and vitamin C content

The TPC and TFC in methanol extracts of *E. operculata* and *A. bunius* FD fruits are shown in Table 6. The TPC in *E. operculata* and *A. bunius* fruits were 226.741 ± 2.099 mg GAE/g DE and 119.356 ± 1.395 mg GAE/g DE, respectively, while the TFC were found to be 108.761 ± 7.015 mg QE/g DE and 64.323 ± 8.828 mg QE/g DE, respectively. *E. operculata* fruit extract showed higher content of both total phenolic and total flavonoid than that of *A. bunius* fruit which attributed to better antioxidant capacity of the former fruit. Similarly, the TPC and TFC in the methanol extract of *G. sapida* fruit reported in the previous study were 294.353 ± 4.696 mg GAE/g DE and 116.95 ± 10.71 mg QE/g DE, respectively [26]. Prakash et al. [49] studied some wild fruits from Sikkim Himalayan region of India and reported the TPC that varied from 7.3 to 119.2 mg GAE/g. While the total phenolic contents of 56 wild fruits from South China ranged from 0.49 ± 0.04 to 54.8 ± 3.05 mg GAE/g wet weight [7]. Saikia et al. [50] reported phenolic content (4.62–14.74 mg GAE/g dry weight) and flavonoid content (0.65 – 7.72 mg QE/g dry weight) in some leafy vegetables which are lower in comparison to this study. Ascorbic acid also known as vitamin C is a water-soluble vitamin and found in many fresh fruits and vegetables. The fresh fruit of *A. bunius* (7.30 ± 1.452 mg/100 g) showed slightly higher vitamin C content than *E. operculata* (6.60 ± 1.123 mg/100 g) fruit (Table 6). Earlier study showed vitamin C content of 8.6 ± 0.30 mg/100 g fresh *G. sapida* fruit [26]. Khomdram et al. [47] reported the vitamin C content in the wild endemic fruits from Manipur (India) that varied from 6.91 mg/100 g in *P. armeniaca* to 375.68 mg/100 g of fresh weight in *P. emblica*.

The extraction of phenolic and flavonoid contents from plant materials depend on the polarity of solvent used for extract preparation [26]. Fruits are important sources of ascorbic acid, phenolic, flavonoid and many other compounds, and possess beneficial effects on human health as antioxidant and antibacterial agents [18].

3.6. Pearson's correlation

This study of antioxidant capacity in the fruit extracts displayed a strong positive correlation of DPPH assay with ABTS assay, H_2O_2 assay and Vitamin C significantly at $p < 0.01$ (Table 7). The study also showed a strong positive correlation of ABTS with H_2O_2 and Vitamin C, H_2O_2 with Vitamin C, FRAP with TPC and TFC, and TPC with TFC. Similar type of study on some wild and cultivated blueberries from Romania was reported by Bunea et al. [51] which is in agreement with this report. Ku et al. [52] also reported a positive correlation of FRAP assay with phenolic and flavonoid contents. A very well-correlation of phenolic and flavonoid compounds with antioxidant capacity of plant extract was established and the involvement of these compounds to the overall antioxidant activity is mainly because of their redox properties and proton donating abilities [18, 51–55].

4. Conclusion

The two wild edible fruits have appreciable proximate and mineral compositions. Phytochemical screening exhibited the presence of various phytochemical constituents which are of biologically and pharmaceutically important. The studies revealed that *E. operculata* fruit had stronger antioxidant capacity than *A. bunius* fruit showing better DPPH, ABTS and H_2O_2 scavenging activities, and higher FRAP value, TPC and TFC. TPC, TFC and vitamin C content of fruits established the food properties which are linked to free radical scavenging activities. TPC and TFC showed a strong positive correlation with antioxidant activity assayed by FRAP and a strong positive correlation of antioxidant activity (DPPH, ABTS and H_2O_2) was also observed with vitamin C suggesting that these compounds are the main compounds responsible for the antioxidant property. Hence, the fruits could play a role against the diseases caused by oxidative stress inhibiting the development of various human diseases and further, isolation and identification of bioactive compounds responsible for antioxidant activity is encouraged.

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