CHAPTER II

Determination of Proximate Composition of Wild Edible Fruits

II.1 Introduction

Proximate composition is important in determining the nutritional status and overall composition of food [1]. Proximate analysis usually involves the determination of the major constituents of food such as moisture, ash, crude protein, crude fat, crude fibre, and total carbohydrate contents [2, 3]. Fruits and vegetables are generally high in moisture content which is used to predict the stability of food systems [4]. Carbohydrates are considered as the most abundant constituents in fruits comprising 50–80% of dry weight. The structural framework, taste and nutritive value of plant food are related to its carbohydrate content. Like proteins, carbohydrates also yield 4 kcal of energy per g while fats yield 9 kcal of energy per g. Glucose and fructose are the most abundant simple sugars found in fruits and the disaccharide sucrose upon hydrolysis yields glucose and fructose. Glucose, fructose and sucrose are soluble sugars and are responsible for the sweetness of fruits and vegetables [5]. Ash content of a food sample refers to total minerals present in the food sample that represents an inorganic residues remaining after the moisture and organic matter have been removed by incineration or complete oxidation of a sample [6]. Excessive intake of crude fats has been recognized as the most important dietary factor contributing to high cholesterol levels. Plant-based foods do not contain significant amounts of cholesterol but do contain steroids or phytosterols which are similar to cholesterol [7]. Fruits constitute less than 1% protein, but tree nuts are a good source of high-quality proteins that contain 9 to 20% protein [8]. Proteins are the most important biomolecules made up of amino acids which are needed for the protein synthesis and other important nitrogen-containing compounds [9]. Dietary fibre is increasingly considered as an essential aspect of healthy diet. Increased intake of dietary fibre plays a beneficial role in weight control and prevention of several diseases, for example, dietary fibre may improves glucose tolerance by delaying the transport of carbohydrates into the small intestine thereby preventing the risk of heart diseases and reduces constipation [10, 11].

Wild fruits are fruits of wild plants and are often exotic less known or underutilized plants. Edible wild fruits are excellent sources of nutrients such as minerals, fibres and vitamins which make an important contribution in the diets of rural communities [12]. Fruits also contain enormous biologically active compounds that contribute health benefits beyond basic nutritional values. Among the biologically active constituents, natural antioxidants have gained much attention due to their safety and potential therapeutic effects [13]. Some epidemiological and nutritional studies have reported that the higher intake of fruits and vegetables reduces the incidence of chronic diseases such as coronary heart problems, cancer, diabetes and Alzheimer's disease [14, 15]. Nutritional analysis of some wild food plants have been demonstrated that in many cases the nutritional quality of edible wild plants are comparable and in some cases even superior to cultivated varieties [16, 17]. The study of the traditional uses of plant species is significant to farming, contemporary medicine and even the manufacturing industrial sectors of a society [18]. Wild foods make an important contribution in the diet of rural communities by facilitating meaningful amounts of essential nutrients in normal times as well as in times of food scarcity [19].

In this study, five wild edible fruits were selected which have been consumed by the rural people in Assam, North East India and they are *Grewia sapida* Roxb. ex DC (**Fig. II.1a** and **II.1b**), *Eugenia operculata* Roxb. (**Fig. II.2a** and **II.2b**), *Aporosa dioica* (Roxb.) Muell.-Arg (**Fig. II.3a** and **II.3b**), *Antidesma bunius* (L.) Spreng. (**Fig. II.4a** and **II.4b**) and *Ottelia alismoides* (L.) Pers (**Fig. II.5a** and **II.5b**). *Grewia sapida* belongs to the family Malvaceae and is locally known as *Kusra pitai* in Bodo in Assam, India. It is a small perennial shrub with a woody root stock that grows to a height of 1–1.5 m. *G. sapida* is native to South Asia and mainly occurs in India, China, Pakistan and Nepal. In India, it is spread over West Bengal, Assam, Bihar, Haryana, Gujarat, and Karnataka [20]. *Eugenia operculata* is a perennial tree widespread throughout Vietnam, China and some other tropical countries [21]. The plant is found growing in Orissa, Bihar and Northeast region of India [22]. Fruit is ovoid, turns purple to black when ripe. The plant produces fruits which are consumed by local communities in Assam. The leaves, bark and buds of *E. operculata* were studied and showed food and medicinal properties [21, 23, 24]. Devi *et al*. [22] also studied ethanol and aqueous leaf extracts of *E. operculata* and reported that the leaf contains different phytochemicals and possess antioxidant property. However, available information regarding nutrient composition and antioxidant properties from the fruits of *E. operculata* has not been reported.

Fig. II.1a: *Grewia sapida* plant.

Fig. II.1b: *Grewia sapida* fruit.

Fig. II.2a: *Eugenia operculata* plant.

Fig. II.2b: *Eugenia operculata* fruit.

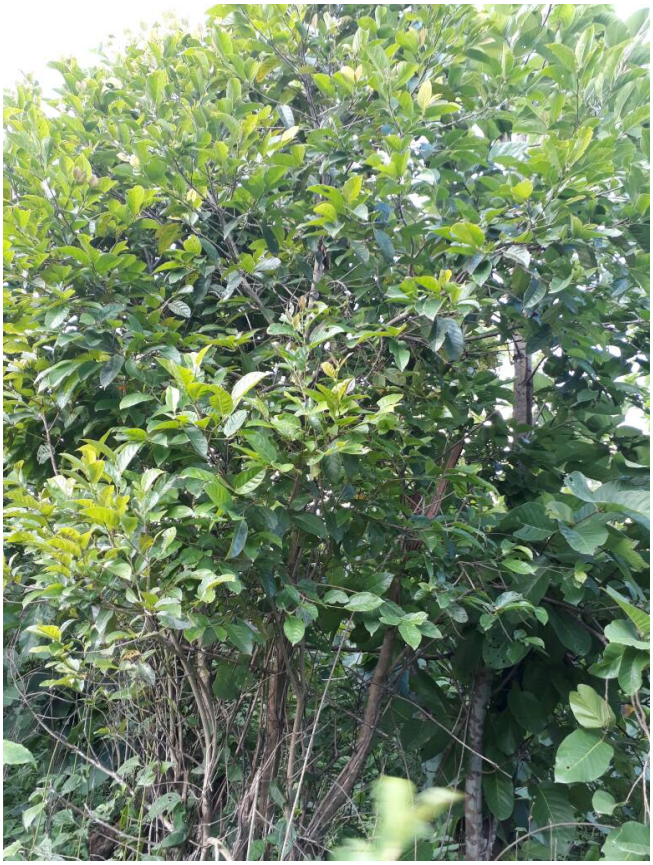

Fig. II.3a: *Aporosa dioica* plant.

Fig. II.3b: *Aporosa dioica* fruit.

Fig. II.4a: *Antidesma bunius* plant.

Fig. II.4b: *Antidesma bunius* fruit.

Fig. II.5a: *Ottelia alismoides* plant.

Fig. II.5b: *Ottelia alismoides* fruit.

Aporosa dioica belongs to the family Euphorbiaceae and is locally known as *Bergao* in Bodo. It is distributed through the eastern Himalayas and North-East India. Fruit is ellipsoid and ranging in size from 1–1.3 cm long. The fruit turns yellow as it ripens, softens up and becomes sweet-sour in tastes [25]. *Ottelia alismoides* belongs to the family Hydrocharitaceae and is locally known as *Khar*. It is widely distributed throughout India, South-East Asia and North eastern Africa. Fruit is ellipsoid to ovoid, cylindrical and is ranging from 1.5–4 cm long, 1–2 cm wide and is opened by decay of the pericarp [25]. *Antidesma bunius* belongs to the family Euphorbiaceae and is distributed widely in Philippines, Thailand and Southeast Asia [26]. Fruit size is about 8 mm long, ovoid in shape and transforms its colour from green to blackish as it ripens.

Herein, we are reporting the proximate composition of these five wild edible fruits found in Assam of Northeast region of India.

II.2 Materials and Methods

II.2.1 Chemicals

Boric acid (H_3BO_3), sodium hydroxide (NaOH), sulphuric acid (H_2SO_4), copper sulphate ($CuSO_4$), potassium sulfate (K_2SO_4) for determination of protein were obtained from Hi-Media Laboratories Mumbai, India and petroleum ether from Merck, Mumbai, India. All other chemicals used in the study were of analytical grade.

II.2.2 Collection and authentication of plant materials

Fresh and mature fruits of *Grewia sapida, Antidesma bunius, Eugenia operculata, Aporosa dioic*a and *Ottelia alismoides* (**Table II.1**) were collected from Chirang (26°30' N latitude and 90°60' E longitude) and Kokrajhar (26°25' N Longitude and 99°16' 38" E Latitude) of Bodoland Territorial Area Districts (BTAD) of Assam, North-East India. *G. sapida*, *A. bunius* and *E. operculata* fruits were collected during April-May, 2015 from Chirang district and the fruits of *A. dioica* and *O. alismoides* were obtained during the month of April and October, 2015, respectively from Kokrajhar district. Approximately, 1 kg of each fruits were collected in tight plastic bags and carried to the laboratories. All the selected fruits appeared healthy in their external appearance. Three herbarium sheets of plants were submitted at Herbarium of Botanical Survey of India (Meghalaya) and authenticated (Ref. No. BSI/ERC/Tech./Plant Iden./2015/211, Dated 25-06-2015) as *Grewia sapida, Eugenia operculata* and *Aporosa dioica*. The voucher specimens of *Antidesma bunius* (Collection No,

501) and *Ottelia alismoides* (Collection No. 502) are deposited in the Herbarium of Botany Department, Bodoland University (Kokrajhar).

Table II.1: Wild edible fruits collected for the study

Botanical name (Family)	Local name (*Bodo)*	Parts used, Test	Availability	Uses
Grewia sapida Roxb. ex DC. (Malvaceae)	Kusra pitai	Whole fruit, Sour and slight sweet	March-May	Fruits are eaten raw when ripe
Eugenia operculata Roxb. (Myrtaceae)	Khorjam	Whole fruit, sweet-sour	June-August	Fruits are eaten raw when ripe
Aporosa dioica (Roxb.) Muell.-Arg. (Euphorbiaceae)	Bergao pitai	Whole fruit, sweet-sour	April-July	Fruits are eaten raw when ripe
Antidesma bunius (L.) Spreng. (Euphorbiaceae)	Pagli tenga	Whole fruit, sweet-sour	June-August	Fruits are eaten raw when ripe
Ottelia alismoides (L.) Pers. (Hydrocharitaceae)	Khar	Whole fruit, Slightly salty	September-December	Fruits are eaten raw when ripe

II.2.3 Sample preparation

The collected wild edible fruits were washed properly with distilled water and processed for determination of moisture content on the same day. For other parameters, the fruits were freeze dried for 72 h, ground to a fine powder using a grinder and kept in the air-tight container prior to use for analysis [17].

II.2.4 Determination of moisture content

The moisture content was determined following AOAC method [27]. Briefly, 5 g of the fresh sample was accurately weighed and heated in a hot air oven (MSW-211, Macro Scientific Works Pvt. Ltd.) at 105°C for 2 h, kept in a desiccator for cooling, weighed and the moisture content was calculated by using the following formula.

$$\text{Moisture (\%)} = \frac{(\text{Weight of sample} - \text{Dry weight})}{\text{Weight of the sample taken}} \times 100$$

II.2.5 Determination of ash content

Ash content was determined by the AOAC method [27]. Silica crucible was first heated in a muffle furnace (NSW-101, Narang Scientific Works Pvt. Ltd), cooled in a desiccator and the initial weight was taken. 5 g of the sample was taken in a weighed silica crucible and then heated in a muffle furnace at 550°C for 6 h, cooled in desiccator to normal temperature, weight of the ash was taken and ash content calculated using the following formula.

$$\text{Ash (\%)} = \frac{\text{(Weight of ash)}}{\text{Weight of the sample}} \times 100$$

II.2.6 Determination of crude fat

The crude fat content was determined following AOAC method [27]. The initial weight of the flask was taken by heating in a hot air oven for overnight at 105°C and cooled in a desiccator. 3–5 g of the freeze dried sample was extracted with petroleum ether (boiling point 60–80°C) using a Soxhlet apparatus (B-811, Buchi) for about 6 h. The extracted fat residue was dried in a rotary evaporator and the weight was measured.

$$\text{Crude fat (\%)} = \frac{\text{(Weight of fat)}}{\text{Weight of the sample}} \times 100$$

II.2.7 Determination of crude protein

Crude protein was determined using Kjeldhal method following the AOAC method [27]. Briefly, 1 g of the powdered sample was digested with 20 mL concentrated H_2SO_4 and Kjeldhal catalyst (9 parts of K_2SO_4 and one part of $CuSO_4$) in a digestion chamber until fuming ceased. A reagent blank was performed without the sample. After digestion, it was distilled in Kjeldhal distillation chamber (Buchi Kjelflex K-360). The evaporated ammonia was condensed and then titrated against the known concentration (0.1 N) of HCl. The concentration of nitrogen was calculated by the following formula.

$$\text{Nitrogen (\%)} = \frac{\text{(A – B)} \times \text{N of HCl} \times 14)}{\text{Weight of the sample}} \times 1000$$

Where, A = Volume (mL) of (0.1 N) HCl used in sample titration.

B = Volume (mL) of (0.1 N) HCl used in blank titration.

14 = Atomic weight of Nitrogen.

The nitrogen content thus obtained was multiplied by a protein conversion factor of 6.25 to get the crude protein content.

Protein (%) = Nitrogen (%) × 6.25

II.2.8 Determination of crude fibre

Crude fibre was also determined following AOAC method [27]. Briefly, 1 g of the dried sample was boiled with 0.25 N H_2SO_4 for 30 minutes. Then it was followed by filtration with muslin cloth, washed with hot water until washings were free of acid. Then the residue was boiled with 0.313 N NaOH. Again it was filtered and washed with hot water followed by 0.5 N H_2SO_4 and 50% ethanol. The residue was dried in an oven at 130°C for 2 h and cooled in a desiccator, then weighed and incinerated in a muffle furnace at 600°C for 30 min. The ash were removed, cooled in a desiccator and weight of the ash was measured. The percentage of crude fibre content was calculated based on 100 g of the freeze dried sample by using following formula.

$$\text{Crude fibre (\%)} = \frac{(\text{Dry weight of digested sample} - \text{Weight of ash})}{\text{Weight of the sample}} \times 100$$

II.2.9 Determination of total carbohydrate

The total carbohydrate content was determined by the difference method [28] with the help of following equation.

Total carbohydrate (%) = 100 – [Moisture (%) + Ash (%) + Crude protein (%) + Crude fat (%)]

II.2.10 Calorific value of fruits

Calorific value or the total energy value of fruits in kcal/100 g were determined by multiplying the values obtained for protein, fat and available carbohydrate [29] with the help of following equation.

Calorific value (kcal/100 g) = 4 × Protein (%) + 9 × Fat (%) + 4 × Carbohydrate (%)

II.2.11 Statistical analysis

All the experiments were carried out for three independent replicates and the data were represented in terms of mean ± standard deviation per 100 g of the dry weight basis. OriginPro 8.5 software (MA 01060, OriginLab Corporation, USA) was used for statistical analysis and executed by the one-way ANOVA and *t*-test at $p < 0.05$.

Table II.2: Proximate composition of wild edible fruits per 100 g of DW

Plants	Moisture (g)	Ash (g)	Crude fat (g)	Crude fibre (g)	Crude protein (g)	Carbohy-drate (g)	Calorific value (kcal)
G. sapida	16.25±0.02[a] 81.06±0.75[*]	0.29±0.03[a]	2.50±0.26[a]	1.71±0.03[a]	0.78±0.02[a]	80.18±0.02[a]	346.34±0.04[a]
E. operculata	3.34±0.04[b] 52.53±0.41[*]	0.34±0.04[a]	1.86±0.02[b]	17.57±0.35[b]	1.32±0.04[b]	93.12±0.08[b]	394.58±0.03[b]
A. dioica	13.32±0.04[c] 80.07±3.55[*]	0.36±0.04[a]	2.63±0.25[a]	28.46±0.71[c]	1.15±0.03[b]	82.57±0.28[c]	358.42±1.11[c]
A. bunius	4.53±0.35[d] 64.47±0.25[*]	0.52±0.03[a]	0.97±0.03[c]	9.43±0.31[d]	1.23±0.05[b,c]	92.74±0.42[d]	384.65±1.29[d]
O. alismoides	4.17±0.02[d] 90.93±1.48[*]	0.28±0.04[a]	1.27±0.21[c]	17.51±0.31[b]	0.86±0.04[a,c]	93.42±0.22[b]	388.51±1.12[e]

**, Moisture content of fresh fruit; DW, dry weight; Values were expressed as mean of three replicates ± standard deviation; The data with different letters in a column are significantly different from each other at p < 0.05.*

II.3 Results and Discussion

In this study, a total of five wild edible fruits were taken and they are listed in **Table II.1** along with their local name, parts used, availability and uses. The results of proximate composition and energy values of the five wild edible fruits studied are presented in **Table II.2**. The results reported here are based on 100 g of freeze dried sample. The **Fig. II.6** and **Fig. II.7** shows the proximate composition and total carbohydrate content of five wild edible fruits per 100 g of dry weight (DW), respectively. Moisture content of the five wild edible fruits ranges from 3.34 ± 0.04 g/100 g to 16.25 ± 0.02 g/100 g of freeze dried sample and it ranges from 52.53 ± 0.41 g/100 g to 90.93 ± 1.48 g/100 g of fresh weight. Highest moisture content was observed in *O. alismoides* (90.93 ± 1.48 g/100 g of fresh weight) and the lowest was found in *E. operculata* (52.53 ± 0.41 g/100 g of fresh weight). The moisture content of *E. operculata* is comparable to that of *Melastoma malabathricum* (56.6 ± 0.71 g/100 g) reported by Nayak and Basak [30] and the moisture content of *A. bunius* (64.466 ± 0.25 g/100 g fresh fruit) is similar to that of *Ficus palmate* (67.82 ± 2.07 g/100 g) and *Arbutus pavarii* (68.06 ± 1.65 g/100 g) reported by Hegazy *et al.* [31]. The moisture contents found in *A. dioica, G. sapida* and *O. alismoides* were close to that of the some conventional fruits reported by Ruiz-Rodriguez *et al*. [32].

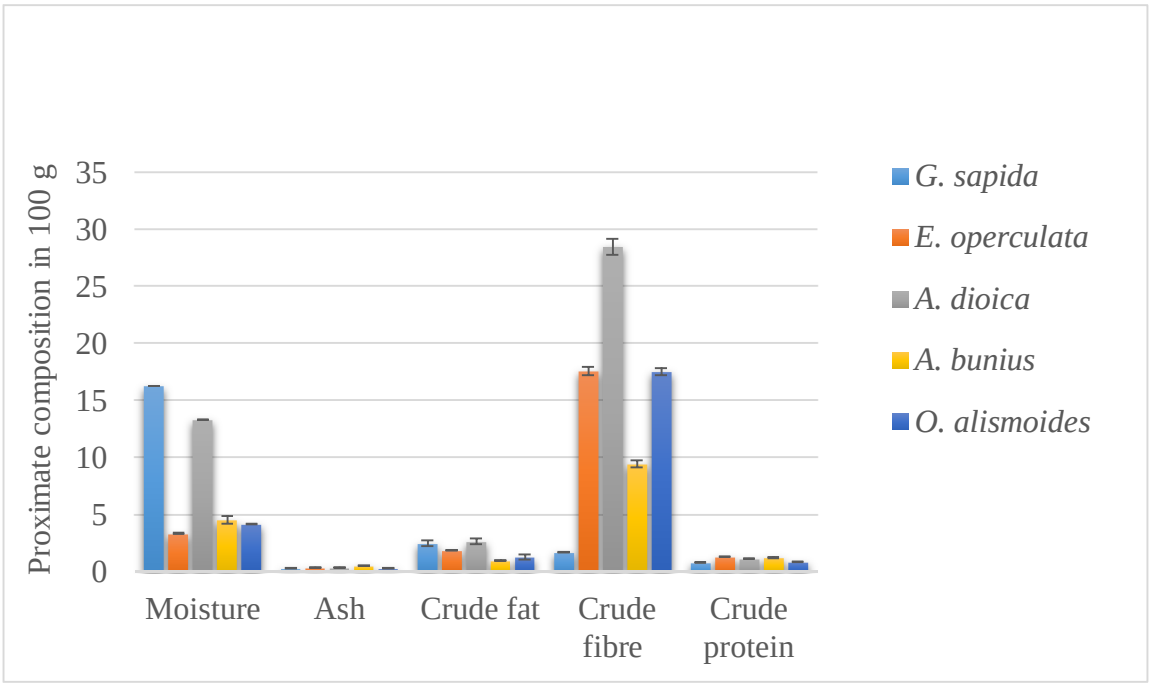

Fig. II.6: Proximate composition in five wild edible fruits per 100 g of DW

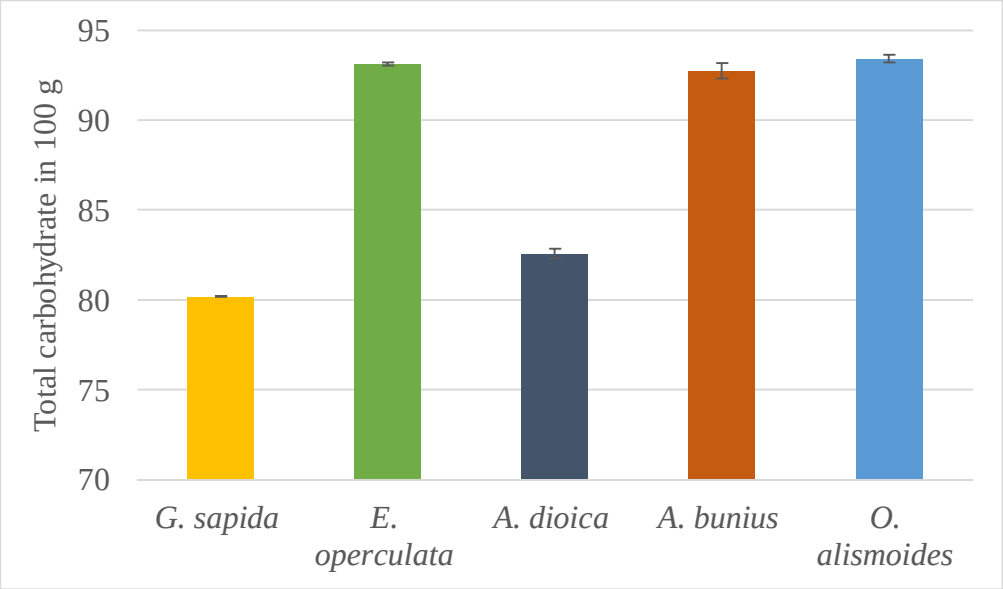

Fig. II.7: Total carbohydrate content in five wild edible fruits per 100 g of DW

Ash is the inorganic residue remaining after all the moisture and the organic materials like carbohydrates, protein, fat, organic acid have been removed [33]. The ash content which is an index of mineral content ranged from 0.29 ± 0.03 g (*G. sapida*) to 0.52 ± 0.03 g (*A. bunius*). The ash content in all the fruits are generally very low and these values are similar with the values of pineapple, orange and pawpaw reported by Ogoloma *et al*. [34].

Crude fat contents of the wild edible plants investigated varied from 0.97 ± 0.03 g (*A. bunius*) and 2.63 ± 0.25 g (*A. dioica*) which is comparable to that of some wild and commercial fruits such as wood apple (3.7%), apple (0.3%), *Baccaurea sapida* (0.73%), *Terminalia chebula* (3.90%), *Eleagnus latifolia* (0.52%) [35, 36]. The crude fat value of *G. sapida* (2.50 ± 0.26 g) is almost similar to the values of tamarind and jujube fruits reported by Salih *et al*. [37]. The crude fat content of *A. bunius* (0.97 ± 0.03 g) and *E. operculata* (1.86 ± 0.02 g) are comparable to the values of *Rosa pulverulenta*, *Rosa dumalis* and *Rosa canina* reported by Ercisli [38].

A. dioica fruit contained the highest amount of crude fibre (28.46 ± 0.71 g) followed by *E. operculata* fruit (17.57 ± 0.35 g/100 g), *O. alismoides* (17.51 ± 0.31 g), *A. bunius* (9.43 ± 0.31 g/100 g) and *G. sapida* (1.71 ± 0.03 g/100 g). The crude fibre obtained in *G. sapida* fruit was comparable to the values of fruits of milk apple, water apple and malay apple reported by Lim *et al*. [39] and it is higher than raspberries (0.85 ± 0.05%) and blueberries (1.48 ±

0.12%) reported by Jeong *et al*. [40]. There was a variation of nutrient content among the fruits studied as the nutritional value of the fruit is strongly affected by the genotype of the fruit [41]. The relatively high fibre content in a food is an indication that can aid digestion and absorption processes in large intestine and thus prevent constipation [42]. High fibre intake could reduce the risk of certain diseases like coronary heart diseases, cancer, high blood pressure, obesity, diabetes and digestive disorders [43, 44]. The recommended dietary allowance (RDA) of fibre for adults, children, pregnant and lactating mothers are 21–38, 19–25, 28 and 29 g/day respectively [45].

The crude protein contents ranged from 0.78 ± 0.02 g in *G. sapida* to 1.32 ± 0.04 g in *E. operculata* per 100 g of dry weight. The crude protein content in *A. bunius* (1.23 ± 0.05 g), and *E. operculata* (1.32 ± 0.04 g) are similar to the values of *Ziziphus spinachristi* fruit reported by Feyssa *et al*. [46] and *Elaeagnus conferta* fruit reported by Rai *et al*. [47]. The crude protein content in *A. dioica* fruit (1.15 ± 0.03 g), *O. alismoides* fruit (0.86 ± 0.04 g) are found similar in some fruits reported by French [48] and the crude protein content in *G. sapida* was also comparable to jujube fruit (0.8 g) reported by Pareek [49].

All the five fruits showed high content of total carbohydrates which varied from 80.18 ± 0.02 g (*G. sapida*) to 93.42 ± 0.22 g (*O. alismoides*). These values are comparable to the values reported for some wild edible plants such as *Eleagnus latifolia* (74.06%), *Baccaurea sapida* (51.90%), *Prunus cerasoides* (84.07%), *Terminalia chebula* (80.61%) and *Morus alba* (87.55%) [36] and some fruits of Meghalaya reported by Seal [50]. The total carbohydrate content in *A. bunius, O. alismoides, A. dioica* and *E. operculata* were higher than the values reported by Gnansounou *et al*. [51]. The fruits rich in carbohydrates are very nutritious for health products and responsible for their high calorific value [52] and these could be a potential supplements for feed formulations.

The calorific value of *E. operculata* (394.58 ± 0.03 kcal/100 g) was found maximum among the five wild edible fruits followed by *O. alismoides* (388.51 ± 1.12 kcal/100 g), *A. bunius* (384.65 ± 1.29 kcal/100 g), *A. dioica* (358.42 ± 1.11 kcal/100 g) and *G. sapida* (346.34 ± 0.04 kcal/100 g). The calorific value of the wild edible fruits of this study were higher in contrast to that of *Carallia brachiata* fruit (310.25 kcal/100 g) reported by Patil *et al*. [53] and are well compared to *Myrica esculenta* (395.04 ± 0.54 kcal/100 g), *Morus indica* (386.00 ± 0.30 kcal/100 g), *Myrica nagi* (386.88 ± 1.25 kcal/100 g) and *Terminalia bellirica* (381.87 ± 0.23 kcal/100 g) [50]. Similarly, calorific value of chempedak and jackfruit reported by Tang *et al*. [54] were 490 kcal/100 g and 301 kcal/100 g respectively. Seal *et al*. [55] also reported the energy value of some wild edible fruits of Meghalaya, India and found

to be in between 342.15 ± 0.13 to 419.09 ± 0.06 kcal/100 g which indicated that these wild edible fruits could be an important source of dietary calorie. High calorific content of the fruits could be attributed to high protein and carbohydrate contents.

II.4 Conclusion

The present study shows that the wild edible fruits have variable amounts of proximate parameters. The calorific value was found the highest in *E. operculata* fruit (394.58 ± 0.03 kcal/100 g) among the five wild fruits followed by *O. alismoides* (388.51 ± 1.12 kcal/100 g) and it was found the lowest in *G. sapida* fruit (346.34 ± 0.04 kcal/100 g). Higher calorific value of the fruits could be attributed to high carbohydrate and protein content. The study showed that these wild fruits have good nutritional qualities and consumption of these fruits in sufficient amount could contribute greatly towards meeting nutritional requirements for normal growth and protection against diseases arising from malnutrition. This study could also provide information to the rural population in knowing the nutritional importance of these fruits. Further research could be conducted for domestication, processing of these fruits to various food products and to improve the shelf-life reducing perishability.

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