



Nutritional and functional properties of *Moringa oleifera*

Salma Sultana

Department of Biochemistry and Molecular Biology, Bangladesh Agriculture University, Bangladesh



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ABSTRACT

This study was carried out to evaluate the nutrient composition and functional properties of dried *Moringa oleifera* leaves collected from two different ecological zones in Bangladesh, Joypurhat and Mymensingh. The proximate analysis revealed that *M. oleifera* leaves were rich in protein content, ranging from (22.99–29.36%), and low in fat, from (4.03–9.51%), fiber, from (6.00–9.60%), and ash, from (8.05–10.38%). The vitamin C content of fresh *M. oleifera* leaves ranged from (187.96–278.50 mg/100 g), Ca ranged from (1.322–2.645%), P ranged from (0.152–0.304 g/100 g), and K ranged from (1.317–2.025 g/100 g). The functional properties included WAC (158.00–415.00%), FC (28.30–117.65 mL/l) and FS (333.33–1000 mL/l). Together, these findings indicate that *M. oleifera* leaves are rich in vital phytonutrients, suggesting a promising balance of food ingredients for human and animal diets.

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1. Introduction

The *Moringa oleifera* plant can grow well in the humid tropics or hot dry lands, can survive in less fertile soils, and is also little affected by drought. Studies have indicated that the leaves have immense nutritional value [1]. As such, the leaves have been used to combat malnutrition, especially among infants and nursing mothers [2]. Traditionally, *M. oleifera* leaves have been used to treat many ailments, such as nervous debility, paralysis, asthma, diabetes, blood pressure, diarrhea, fever, cough, cholera, spasms, enlarged liver and spleen, infection and ulcer, and inflammation, and to promote wound healing [3,4]. Over the past two and a half decades, many reports have appeared in mainstream scientific publications describing the leaves' nutritional, medicinal and other properties [5] (see Figs. 1 and 2).

However, there are considerable variations in the nutritional value of *Moringa* species, and this nutritional value depends on factors such as genetic background, environment and cultivation methods [6].

Therefore, the current research was designed to elucidate the following objectives:

- i) To analyze the proximate composition of dried *Moringa* leaves,
- ii) To determine the vitamin C contents and mineral compositions, including Ca, P and K, of dried *Moringa* leaves, and

- iii) To evaluate the functional properties, including the water absorption capacity, foaming capacity and foaming stability, of dried *Moringa* leaf powder.

2. Materials and methods

This study was carried out at the postgraduate laboratory of the Department of Biochemistry and Molecular Biology, Bangladesh Agricultural University (BAU), Mymensingh.

2.1. Plant materials

2.1.1. Collection of *M. oleifera* leaves

Fresh leaves of local cultivars of *M. oleifera*, three from Joypurhat and two from the BAU campus, Mymensingh, were collected. Each plant sample was given a code for blind analysis. Voucher specimens were deposited in the postgraduate laboratory of the Department of Biochemistry and Molecular Biology, BAU, Mymensingh.

Taxonomy.

Kingdom: Plantae

Order: Brassicales

Family: Moringaceae

Genus: *Moringa*

Species: *M. oleifera* Lam.

E-mail address: shayonsalma@gmail.com.

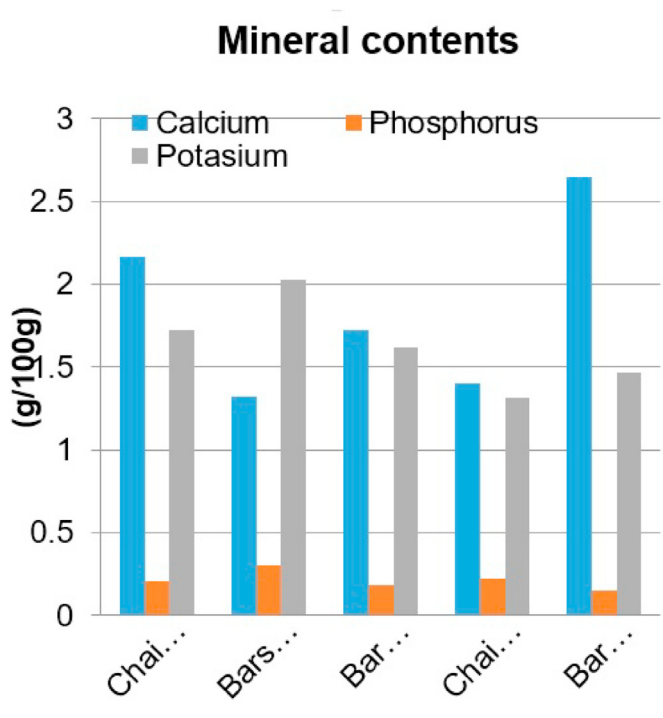


Fig. 1. Mineral contents of *Moringa oleifera* leaves.

2.1.2. Preparation of leaf powder

The leaves of *M. oleifera* were shade-dried for three days and subsequently pulverized using a grinder. The powdered samples were then sieved to remove the debris. The sieved powder was stored in sealed sample bags in the dark at -20°C for further experiments.

Proximate analysis of *M. oleifera* leaves.

2.1.3. Moisture

The moisture content of the sample was determined using the method described by Ref. [7]. The samples were collected in weighed, tared porcelain crucibles. The crucibles containing the samples were weighed and then placed in an oven maintained at 105°C for 24 h. The crucibles were then transferred to desiccators and allowed to cool to room temperature. The final weights of the crucibles with the dried samples were then determined. The percent weight loss was reported as the percent moisture content. The moisture was determined from the following formula:

$$\% \text{ Moisture} = \frac{\text{Initial weight (g)} - \text{Final weight (g)}}{\text{Weight of the sample (g)}} \times 100$$

where Initial weight = sample weight + crucible weight (before heating).

Final weight = dry sample weight + crucible weight (after heating).

2.1.4. Protein content

The principle of protein estimation is based on estimating the nitrogen content of the material and then multiplying the nitrogen value by 5.58. This value is referred to as the crude protein content, since the nonprotein nitrogen (NPN) present in the material was taken into consideration in the protein investigation. The

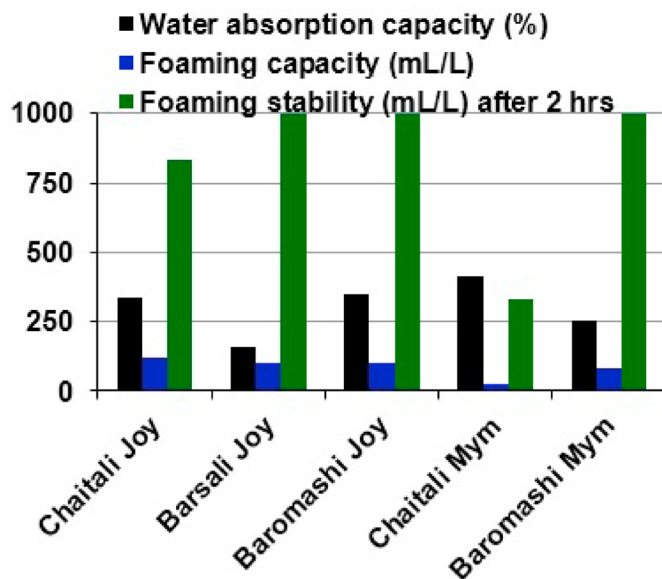


Fig. 2. Functional properties of *Moringa oleifera*.

estimation of nitrogen was made by the modified micro-Kjeldahl method, which depends on the fact that organic nitrogen, when digested with concentrated sulfuric acid, is converted into ammonium sulfate. Ammonia liberated by making the solution alkaline is then distilled into a known volume of standard boric acid, which is then back-titrated.

Reagents:

Catalyst mixture (Copper sulfate: Potassium sulfate = 1:7).

Concentrated sulfuric acid (H_2SO_4) solution.

2% Boric acid (H_3BO_3) solution.

Hydrochloric acid (0.2 N HCL) solution.

40% sodium hydroxide solution (NaOH).

Mixed indicator (methyl red and methylene blue).

Working procedure:

A 0.5 g sample of dried ground leaves was collected in weighing paper and accurately measured. The sample was then poured into a 75-ml clean and dried Kjeldahl flask, to which 5 ml of concentrated H_2SO_4 and 4 g of catalyst mixture were added. The sample mixture was then heated at 370°C for 1 h on a preheated heater. When the sample became colorless (white), the digestion of the sample was completed. The digested sample was cooled to room temperature (25°C) and diluted to 60 ml. Ten milliliters (100 ml) of the digested, diluted sample solution was placed in a distillation apparatus with 25 ml of 40% NaOH. The distillate (approximately 60 ml) was collected in a conical flask containing 0.25 ml of 2% boric acid solution and 2 drops of mixed indicator. The total distillate was collected and titrated with standardized HCL solution (0.1 N HCL).

Calculation:

The amount of nitrogen was calculated according to the following equation:

$$\% \text{ Nitrogen} = \frac{(TS - TB) \times \text{Strength of HCL} \times 0.014}{\text{Weight of the sample (g)}} \times 100$$

where TS = Titer value of the sample in ml.

TB = Titer value of the blank

Strength of HCL acid = 0.1 N

Then, the percentage of the nitrogen of the sample was

multiplied by 5.58 to obtain the total crude protein according to the following equation:

$$\% \text{ Crude protein} = \% \text{ Nitrogen} \times 5.58$$

2.1.5. Determination of crude fat

Reagent.

Acetone solution:

Working procedure:

Crude fat was determined with the help of a Soxhlet apparatus. Three grams of a ground leaf sample was collected in a thimble and continuously extracted with 60 mL of n-acetone for a period of nearly 20 h. After evaporation of the flask containing n-acetone, the difference in the weight of the tared beaker and the weight of the oil beaker was used to calculate the crude fat.

Calculation:

The fat content was calculated according to the following equation

$$\% \text{ Crude fat} = \frac{(\text{Wt. of crude fat} + \text{beaker}) - \text{wt. of tared beaker}}{\text{Wt. of the grounded leaf sample}} \times 100$$

2.1.6. Determination of crude fiber

Reagents:

25% H₂SO₄ solution.

1.25% NaOH solution.

Working procedure:

The crude fiber content of the *M. oleifera* leaf samples was determined by sequential acid and alkali hydrolysis followed by ignition of the hydrolysate as described in the [8]. One gram (1 g) of a grounded sample (oven-dried) was collected in a 1-L beaker covered with a round-bottom flask containing cold water. The system maintains constant volume during boiling. The content of the beaker was refluxed to the boiling point with 250 ml of 1.25% H₂SO₄. The beaker was then sealed with cotton, and the mixture was boiled on a heater for 30 min. Then, the content was filtered quickly in a Buchner funnel through Whatman No. 42 filter paper and washed to remove the acid. The acid-free residue was refluxed with 200 ml of 1.25% NaOH solution at boiling point for exactly 30 min, maintaining the constant volume as before. Then, the mixture was filtered as before and washed with water to make the residue alkali-free. The residue was transferred to a crucible and dried in an oven at 100± 5 °C until a constant weight was attained. The sample was then cooled in a desiccator and weighed (the residue contained ash and crude fiber). The sample was then ignited in a muffle furnace at 600 °C for 4 h, after which it was cooled again in a desiccator. The weight of the ignited sample was recorded after subsequent cooling. The weight loss of the samples after ignition indicated the amount of crude fiber.

Calculation:

The percent of crude fiber calculated using the following formula

$$\text{Weight of crude fiber} = (\text{Weight of crucible} + \text{crude fiber} + \text{ash}) - (\text{Weight of crucible} + \text{ash})$$

2.1.7. Determination of ash

Working procedure:

A 3.0-g sample of powdered leaf was collected in the crucible, and the preashing of the sample was performed by placing the crucible in a muffle furnace maintained 300 °C for 3 h. The temperature of the ashing was increased to 600 °C for 9 h. The crucible was then cooled and kept in a desiccator for some time and weighed. The percent of ash was the calculated based on the recorded weights.

Calculation:

The percent of ash was calculated using the following formula:

$$\text{Weight of ash} = (\text{Weight of crucible} + \text{ash}) - \text{weight of crucible}$$

$$\% \text{ Ash} = \frac{\text{Wt. of ash (g)}}{\text{Wt. of sample (g)}} \times 100$$

2.1.8. Carbohydrates

The carbohydrate content (nitrogen free extract or NFE) was determined by the difference; that is, the sum of all the percentages of moisture, fat, crude protein, ash, and crude fiber was subtracted from 100%. This calculation provided the amount of nitrogen-free extract, otherwise known as carbohydrate [9].

$$\% \text{ Carbohydrate} = 100$$

$$- (\% \text{ moisture} + \% \text{ fat} + \% \text{ ash} + \% \text{ crude fibre} + \% \text{ crude protein})$$

2.1.9. Organic matter

The organic matter content was estimated by subtracting the percentages of moisture and ash content from one hundred [9].

$$\% \text{ Organic matter} = 100 - (\% \text{ moisture} + \% \text{ ash})$$

2.1.10. Energy value

The caloric value of the sample was calculated using the "Atwater factor" by multiplying the values of the crude protein, lipid and carbohydrate by 3.99, 9.1, 3.99, respectively, and taking the sum of the product [9].

2.2. Determination of vitamin C contents

Ascorbic acid was determined following a procedure that was previously described by the authors [10] by using 2,6-dichloroindophenyl and measuring the content by a titrimetric method. The results are expressed as mg of ascorbic acid per 100 g of fresh weight.

2.3. Determination of mineral contents

To determine the calcium, phosphorus and potassium contents of *M. oleifera* leaves, a protocol developed by Ref. [11] was followed. Reagents:

Nitric acid solution
Perchloric acid solution
Double-distilled water

Digestion of sample:

- A 0.1-g sample of ground powder was collected into a measuring cylinder, dissolved in nitric acid and perchloric acid (2:1 = 10 ml: 5 ml) solution and maintained for 1–1.5 h in a digestion block at 200 °C.
- After proper digestion, when the colors had disappeared, the samples were transferred into a volumetric flask after filtration, and distilled water was added to ensure a final volume of 100 ml.
- Next, the samples were transferred into plastic bottles and preserved until analysis.

2.3.1. Quantitative measurement of potassium (K)

- The samples were diluted using double-distilled water.
- The K content was measured using a flame photometer.
- The final reading was calculated using the following formula:

2.3.2. Determination of phosphorus (P) content

The content of P in the plant samples was colorimetrically determined by developing a blue color with stannous chloride (SnCl₂: 2H₂O) via the reduction of the phosphomolybdate blue complex and measuring the color using a spectrophotometer at a wavelength of 600 nm [12].

2.3.3. Determination of calcium content

The calcium concentration of plant samples was analyzed by a complexometric titration method using Na₂EDTA as a chelating agent at pH 12 in the presence of a calcon indicator. This analytical technique was outlined by Ref. [13].

2.4. Functional properties

2.4.1. Water absorption capacity (WAC)

Water absorption was measured using a modification of the centrifugation technique of [14]. Two grams of *Moringa oleifera* flour was weighed into a 50-ml centrifuge tube and mixed with 25 ml of distilled water with continuous stirring for 1 min. The resulting slurry was centrifuged at 3075×g for 30 min. The supernatant was then decanted, and the water retained per gram sample was calculated.

$$\text{Water absorption capacity} = \frac{\text{Wt. of bound water (g)}}{\text{Wt. of sample (g)}} \times 100$$

2.4.2. Foaming capacity and foaming stability (FC and FS)

Foaming capacity and stability were studied according to the method of [15]. Two grams (2 g) of a sample was whipped with 100 ml of distilled water for 5 min in a Waring blender and then poured into a 250-ml graduated cylinder. The foaming capacity and stability were calculated according to the following equations:

$$\text{Foaming capacity} = \frac{\text{Foam volume immediately after mixing (ml)}}{\text{Starting volume of liquid phase (ml)}} \times 1000$$

$$\text{Foaming stability} = \frac{\text{Foam volume after 2 hr of mixing (ml)}}{\text{foam volume immediately after mixing (ml)}} \times 1000$$

2.5. Data analysis

All the data are expressed as the mean ± SD with a minimum of triplicate analysis. Statistical comparisons were made by one-way analysis of variance (ANOVA) with post hoc Duncan multiple comparisons (SPSS software, version 16.0). Predetermined *p* values ≤ 0.05 were considered statistically significant.

3. Results

A total of five local cultivars of *Moringa oleifera* (*Sajna*) collected from the Joypurhat and Mymensingh zones were analyzed for nutrient composition and functional properties (see Table 1).

3.1. Proximate composition

The proximate compositions of *Sajna* leaves was determined by the AOAC methods. These compositions varied among the cultivars (Table 2). The crude protein contents varied significantly among the cultivars, ranging from 22.99 to 29.36%. The highest protein content was found in Barsali Joy, whereas the lowest was found in Chaitali Myn. Chaitali Joy contained the highest crude fat content, whereas Baromashi Myn contained the lowest content. Similarly, the crude fiber content was highest in Chaitali Joy, whereas the lowest was found in Baromashi Myn. The carbohydrate contents were calculated by the subtraction method on a moisture-free basis. Baromashi Joy possessed the highest carbohydrate contents among the cultivars, although there was no significant variation in this proximate content. The energy contents were also determined by a calculative value. This value was found to be highest in Baromashi Joy and Chaitali Joy and lowest in Barsali Joy and Baromashi Myn.

3.2. Vitamin C content and mineral compositions

The vitamin C content of fresh *Sajna* leaves was determined by a titrimetric method. The content varied widely among the cultivars (Table 3). *Chaitali* Myn contained the highest contents of vitamin C (278 mg/100 g), and the vitamin C contents of other cultivars ranged from 187.96 to 212.50. The mineral contents of *Sajna* leaves also varied among the cultivars (Table 3). The calcium content was determined by a titrimetric method. The highest Ca content (2.645 g/100 g) was found in *Baromashi* Myn, followed by *Chaitali* Joy (2.164 g/100 g). The phosphorus content was determined by a spectrophotometric method. *Barsali* Joy contained the highest P content (0.304 g/100 g DM). The potassium contents were determined by a flame photometric method. *Barsali* Joy also contained the highest K content (2.025 g/100 g DM).

Table 1
Cultivars of *M. oleifera* used for the experiments.

Ecological zone	Cultivars	Code name
Joypurhat	Chaitali	Chaitali Joy
	Barsali	Barsali Joy
	Baromashi	Baromashi Joy
Mymensingh	Chaitali	Chaitali Mym
	Baromashi	Baromashi Mym

Table 2
Proximate composition (%) and energy value (kcal/100 g DM) of powdered leaves of five local cultivars of *Sajna*.

<i>Sajna</i> cultivars	Moisture	Ash	Crude protein	Crude fat	Crude fiber	Carbohydrates (NFE)	Organic matter	Energy
Chaitali Joy	8.29 ± 0.21ab	10.38 ± 0.45a	23.26 ± 0.43d	9.51 ± 0.12a	9.60 ± 0.29a	47.25 ± 0.39d	81.33 ± 0.67c	367.88 ± 1.28a
Barsali Joy	7.55 ± 0.25c	9.22 ± 0.30b	29.36 ± 0.54a	4.45 ± 0.21c	8.00 ± 0.25b	48.97 ± 0.88c	83.23 ± 0.04a	353.03 ± 3.30b
Baromashi Joy	8.22 ± 0.33ab	8.05 ± 0.39c	24.05 ± 0.26c	5.25 ± 0.19b	6.40 ± 0.12c	56.25 ± 0.97a	83.73 ± 0.11a	368.17 ± 1.09a
Chaitali Mym	8.65 ± 0.37a	8.79 ± 0.19b	22.99 ± 0.24d	5.52 ± 0.25b	6.70 ± 0.19c	56.00 ± 0.38a	82.56 ± 0.20b	365.40 ± 2.87a
Baromashi Mym	7.94 ± 0.31bc	10.29 ± 0.23a	25.56 ± 0.46b	4.03 ± 0.14d	6.00 ± 0.40c	54.12 ± 0.86b	81.77 ± 0.07c	354.60 ± 2.90b

NFE, nitrogen-free extract.

DM, dry matter.

Values are expressed as the mean ± SD (minimum of triplicate analysis).

Means in a column not sharing the same letter are significantly different ($P < 0.05$).

3.3. Functional properties

The water absorption capacity (WAC) of *Sajna* leaves varied widely among the cultivars (Table 4), ranging from 158.00 to 415.50%. The foaming capacity (FC) and foaming stability (FS) also varied widely among the cultivars (Table 4). The FC ranged from 28.30 to 117.65 mL/L, and the FS, recorded after 2 h, ranged from 333.33 to 1000 mL/L.

4. Discussion

This study demonstrates the nutritional and functional attributes of *Moringa oleifera* leaves. The crude protein content, which was one-fourth of the total nutrient contents based on dry matter, is noteworthy. Other studies have reported variable protein contents of 16, 22.42, 23.27, 27.4–40% [16]; Sarwatt et al., 2004; [17].

The *Moringa* leaves contain a low fat content (4.03–9.51%), which is desirable. Moreover, *Moringa* contains more dietary polyunsaturated fatty acids (PUFAs) than saturated fatty acids (SFAs). A higher content of PUFAs and a lower amount of SFAs is desirable [18]; as such, the inclusion PUFAs in the diet is recommended, as they can prevent the occurrence of diseases, thereby promoting good health.

The *Moringa* leaf powder is rich in carbohydrates and has great caloric value that can contribute to the caloric requirements of the body. Carbohydrates are an essential part of a healthy diet and should make up 50% of our daily calorie intake.

The low moisture content of the leaf powder is an attribute of a very high shelf life. Hence, long storage of the leaf powder would not lead to spoilage due to microbial attack, which supports the practice of storage in dry form by users. In addition, moisture content is among the most vital and most commonly used measurements in the processing, preservation and storage of food [9].

Ash in food contributes to the residue remaining after all the moisture has been removed and after the organic materials (fat, protein, carbohydrates, vitamins, organic acid etc.) have been incinerated [9]. Thus, the ash content of the dried leaf powder is considered to be a measure of the mineral content. The results

indicate that the dried *Moringa* leaves have high deposits of mineral elements, which is in agreement with the previous findings [2]. Calcium is required for the formation and maintenance of bones and teeth, thus preventing osteoporosis. Calcium is also needed for normal blood clotting and nervous function.

Crude fiber is largely composed of cellulose with small amounts of lignin, which is indigestible for humans. The low fiber content in the study compared with most forage plants is of interest because the fiber fraction defines the extent and rate of feed digestibility [19]. Although appropriate crude fiber enhances digestibility and aids in the absorption of microelements, glucose and fat, its presence in high levels can cause intestinal irritation, lower digestibility and decreased nutrient usage [20]. The crude fiber content of *Moringa* leaves (6.00–9.60%) obtained in this study was considered to be at the acceptable level, making *Moringa* leaves a promising ingredient for human and animal diets.

Moringa leaves are rich in vitamins, including water-soluble vitamins. In particular, *Moringa* is reported to be rich in vitamin C, which increases iron absorption in the body [1]. The vitamin C content was found to be high compared to other vitamin C-rich plant sources.

The WAC values of *Moringa* leaves ranged from 158.00 to 415.50% and were comparatively high due to the leaves' high protein contents. The WAC is considered an essential attribute of food ingredients for the formulation of various value-added food products, including bakery products. The foaming capacity (FC) ranged from 28.30 to 117.65 mL/L, and the foaming stability (FS) ranged from 28.30 to 117.65 mL/L. The FS recorded after 2 h ranged from 333.33 to 1000 mL/L. The FC and FS are important quality indicators in food products, such as breads, cakes, crackers, meringues, ice creams and several other bakery items, to maintain their texture and structure during or after processing [21].

The mode of storage between the collection and analysis (i.e., drying, refrigeration, freezing) might influence the leaves' nutritional composition. *Moringa* has been reported to possess some medicinal properties; thus, its inclusion in humans' and animals' diets could function as curative and therapeutic therapy. However numerous studies have shown that *Moringa oleifera* leaves offer

Table 3
Vitamin and mineral contents of powdered leaves of five local cultivars of *Sajna*.

<i>Sajna</i> cultivars	Vitamin C (mg/100 g)	Minerals (g/100 g DM)		
		Ca	P	K
Chaitali Joy	212.50 ± 6.45b	2.164 ± 0.07b	0.208 ± 0.03b	1.722 ± 0.01b
Barsali Joy	193.27 ± 5.98c	1.322 ± 0.02e	0.304 ± 0.02a	2.025 ± 0.02a
Baromashi Joy	187.96 ± 6.24c	1.723 ± 0.03c	0.185 ± 0.01bc	1.621 ± 0.00c
Chaitali Mym	278.50 ± 8.11a	1.402 ± 0.2d	0.219 ± 0.01b	1.317 ± 0.00e
Baromashi Mym	209.50 ± 3.97b	2.645 ± 0.4a	0.152 ± 0.01c	1.469 ± 0.01d

DM, dry matter.

Values are expressed as the mean ± SD (minimum of triplicate analysis).

Means in a column not sharing the same letter are significantly different ($P < 0.05$).

Table 4Water absorption capacity (WAC), foaming capacity (FC) and foaming stability (FS) of powdered leaves of five local cultivars of *Sajna*.

<i>Sajna</i> cultivars	Water absorption capacity (%)	Foaming capacity (mL/L)	Foaming stability (mL/L) after 2 h
Chaitali Joy	335.50 ± 21.68b	117.65 ± 8.00a	833.33 ± 26.21b
Barsali Joy	158.00 ± 12.49d	100.00 ± 7.02b	1000.00 ± 36.34a
Baromashi Joy	350.50 ± 18.54b	102.04 ± 7.99b	1000.00 ± 46.48a
Chaitali Mym	415.50 ± 13.20a	28.30 ± 4.32d	333.33 ± 25.18c
Baromashi Mym	255.50 ± 9.96c	80.00 ± 4.50c	1000.00 ± 26.85a

Values are expressed as the mean ± SD (minimum of triplicate analysis).

Means in a column not sharing the same letter are significantly different ($P < 0.05$).

high protein content; contrary to these findings, our study did not find a high level of protein. This variation may be explained by several factors, such as climate and the geography of development of the crop [22]. In my study protein level varied in Joypurhat and Mymensingh districts due to above mention factors. It is important to bear in mind that the mineral and phenolics contents present in leaves depend on several factors such as geographical area where the plant is cultivated, type of soil, water and fertilizers, industrialization process, and storage conditions [22]. Taking these precedent into consideration it may be stated that the variation in nutritional and functional properties of *Moringa oleifera* in two different districts in Bangladesh are due to above mention factors./

5. Conclusion

In the present study, dried leaves of five *Moringa oleifera* cultivars were investigated for their nutrient compositions and functional properties.

Declaration of competing interest

This is an original article. No part of this work has been borrowed from any other scientific writing.

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