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# Preliminary Phytochemicals Screening and Estimation of Total Nutritional Content in Different Varieties of Mizo Chilli (*Capsicum frutescens*)

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## ABSTRACT

Chilli is considered as an excellent source of bioactive compound. Moreover, an average amount of nutrients are found in chilli extract. Besides, chilli content high amount of vitamin C as compared to other spices and vegetables. Qualitative phytochemical screening and total nutritional content of the three Mizo chilli varieties viz; A, B and C was determined. The principle includes preliminary phytochemical screening and total estimation of ash, proteins, carbohydrates and ascorbic acid content. The output of these analyses indicates that sufficient amount of nutrients are present in these indigenous varieties of *Capsicum frutescens* and nutritional content varies from varieties to variety. Thus, the proposed study will give us information on phytochemical and nutrient application of Mizo chilli plants.

**Key words:** Mizo chilli, *Capsicum frutescens*, Vitamin C, Phytochemicals

*Capsicum sp.* is a major spice crop commonly cultivated in different parts of the world. Besides chilli content important nutrients for human consumption because of its nutritional and vitamins content. It is one of the most common valuable crops of India which belongs to family Solanaceae and cultivated throughout the country. Moreover, the extracts of Chilli are used in pharmaceuticals, cosmetic products, paints and chilli sprays. In North eastern parts of India, Jhum cultivation or shifting cultivation is the main agriculture practice where most of the farmers still rely on. The Chilli variety is indigenous to the Northeast region of India and it is an important crop of the North-East India but scientifically it has not been explored to its fullest. According to Dhaliwal [1], chilli was introduced directly by the Christian missionaries to north eastern India from South America.

*Capsicum frutescens* which is exclusively grown in Mizoram is distinctly different from other varieties grown in different parts of the country. It is cultivated on hill slopes under shifting cultivation system or *jhuming* system which are grown completely organically in the *jhum* lands. In Mizoram, Bird's eye chilli is locally known as *Hmarchate* or Mizo chilli or *Mizoram Bird's eye chilli* (MZBEC) which belongs to species *Capsicum frutescens* and is widely grown in the state of Mizoram. In Mizoram itself three different varieties of Mizoram Bird's eye chilli (MZBEC) viz; A, B and C are

cultivated in different parts of the state. The government of Mizoram claimed that Mizoram is the native place of Bird's eye chilli locally called *Hmarchate* or Mizo chilli. The ethnic communities of the north-eastern India also used in treating various ailments [2]. However, systematic and more extensive ethnomedicinal investigations are not carried out to provide new insights into the other traditional uses of this important plant. Moreover, no scientific study has been made on nutrient content and phytochemical screening of *Capsicum frutescens* in Mizoram and no literatures are available in this aspect.

Plants are the reservoirs of biological active compounds to combat various pathogens [3]. Phytonutrients provides crucial linked between health and nutrition. Chilli is a rich source of ascorbic acid and an average amount of carbohydrates and proteins are present. It is found in the literature that 100 grams of the edible portion of chilli provides 24 k Cal of energy, 1.3 grams of protein 4.3 grams of carbohydrate and 0.3 grams of fat [4]. Vitamin C cannot be endogenously synthesized by human body, so it is very essential for our dietary component [5]. Vitamin C is instrumental in neutralizing free radicals, which are harmful to the body, assimilation of iron, healing of wounds, helps to build collagen which aids the skin, and also defence against bacteria and virus infection [6]. Vitamin C has been considered greatly as an important ingredient for boosting immune system during this Covid-19 pandemic. It is considered as most effective biochemical for immune response against Covid-19 virus recently with the outbreak of Covid-19 pandemic. Vitamin C or Ascorbic acid is the biologically active form of dehydroascorbic acid and it also prevents some important illness such as cancer, anaemia, diabetics, and cardiovascular diseases. It is fundamentally provided by fruits and vegetables [7]. Higher amount of ascorbic acid is found in

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chilli than other fruits and vegetables [8]. In addition, ascorbic acid content of the fresh peppers increases during the ripening and ascorbic acid content decreases during the post-harvesting period. Also, this value might be changed with the cultivar, production practices, maturity at harvest and storage conditions [9]. The objective of this study was therefore to find out the qualitative phytochemical properties and total nutritional contents of Mizo chilli (*Capsicum frutescens*) viz. Grade A, B and C grown in Mizoram.

## MATERIALS AND METHODS

### Collection of sample material and extraction

Fresh varieties of Mizo chilli were air-dried and used as the samples in these experiments. The samples were collected from Horticulture Centre of Excellence, Thiak village, about 45 kilometres South of Aizawl, Mizoram. Air-dried ripe fruits of Mizo chilli were powdered and extracted by Soxhlet extraction method.

### Preliminary phytochemical screening

Phytochemical screening of the chilli extract was done using the standard protocol described below.

#### Test for alkaloids (Mayer's reagent test)

2 ml of the extract was dissolved in 2 ml of 2N Hydrochloric acid and two to three drops of Mayer's reagent was added. The creamy white precipitate formation indicated the presence of alkaloids [10-11].

#### Test for terpenoids (Salkowski test)

In 5 ml of each extract 2 ml of chloroform was added, 3 ml of sulphuric acid was added with a careful overlaying. Reddish brown precipitate formation at the interface indicated the presence of Terpenoids [10].

#### Test for saponins (Foam test)

The extract of 3 ml was diluted with distilled water upto 10 ml and was shaken vigorously for two minutes. White froth formation indicated the presence of Saponins.

#### Test for flavonoids ( $H_2SO_4$ test)

A few drops of concentrated sulphuric acid were treated with 5 ml of the extract. Orange-yellow colour appearance indicated the presence of flavonoids [10].

#### Test for tannins (Ferric chloride test)

0.5g or 500mg of the plant extract was boiled in 20ml of distilled water and filtered. To the filtrate a few drops of 0.1% ferric chloride was added. Blue-black or brownish green colour formation indicated the presence of tannins [11].

### Determination of total nutritional content

#### Estimation of total ash content

Dry ashing method was used for the estimation of total ash content in the sample. In this method, 1g of the oven dried sample was ignited in a muffle furnace with a temperature of 550°C for three hours. The process was repeated until a constant weight of the dried crucibles was observed.

$$\text{Ash (\%)} = \frac{\text{weight of Ash}}{\text{weight of sample}} \times 100$$

#### Determination of Ascorbic acid is done by 2, 4-dinitrophenylhydrazine method

5 grams of chilli pepper fruits were washed with tap water and cut into small pieces and homogenized with the help

of mortar and pestle by adding 5 ml of 4% oxalic acid. The homogenates were centrifuged at 5,000 rpm for 10 minutes then the supernatants were filtered with 540 Whatmann filter paper. The obtained residues were made up to 25 ml with 4% oxalic acid. The ascorbic acid content was estimated by using 2, 4 dinitrophenylhydrazine (DNPH) reagents in conjunction with spectrophotometer at 540 nm [12]. Pure Ascorbic acid reagent is used as standard reagent for preparing the standard curve (Fig 1) against which the unknown concentration of Vitamin-C was expressed in the milligrams per 100 milligrams of the sample.

#### Determination of total protein content

Protein estimation was done by Lowry [13] method. Reagent A: 2gm NaOH +500ml. double distilled water + 1gm Sodium carbonate. Reagent B: 1gm potassium sodium tartrate + 50ml double distilled water +0.25gm  $CuSO_4$  Reagent C: 1ml of Reagent B + 50ml of reagent A Reagent D: 1ml of foline + 1ml double distilled water Phosphate Buffer: Solution A: 1.36gm potassium dihydrogen orthophosphate + 100ml double distilled water Solution B: 1.74 g dipotassium orthophosphate + 100ml double distilled water Solution C: 39ml of solution of A + 61ml of solution B. Normal NaOH; 4gm NaOH+100ml double distilled water 0.5 gm fruit weight and crushed in 5 ml of Phosphate buffer and centrifuge at 3000 rpm for 5 minutes. 0.5 ml of the Supernatant was collected in the test tube and 0.5 ml of 1N NaOH, 2.5 ml reagent C and 0.5ml reagent D was added to it after adding the reagent D blue colour was appeared and absorbance was read at 660 nanometers. Bovine Serum Albumin (BSA) is used as standard reagent for preparing the standard curve (Fig 2) against which the unknown concentration of proteins was expressed in the milligrams per 100 milligrams of the sample.

#### Quantification of total carbohydrates content

The total carbohydrates content was determined by Anthrone method [14]. Anthrone reagent: Dissolved 2grams of Anthrone in 1 litre of concentrated  $H_2SO_4$ . The freshly prepared reagent is used for the assay. Glucose stock solution: 200µg glucose per mL distilled water. It was pipette out into a series of test tubes different volumes of glucose solution from the supplied stock solution (200µg /ml) and the volume is made upto 1 mL with distilled water. Then 4 mL of the Anthrone reagent (supplied) were added to each tube and mixed well by vortexing. After the tubes were cooled, the tubes were covered with aluminium on top and incubate at 90°C for 17 minutes. Then the tubes were cool to room temperature and the optical density was measured at 620nm against the blank. D-glucose is used as a reagent for preparing a standard curve (Fig 3) against which the unknown concentration of total carbohydrates was expressed in the milligrams per 100 milligrams of the sample.

## RESULTS AND DISCUSSION

### Preliminary phytochemical screening

From the result (Table 1) all the varieties of Mizo chilli showed the presence of alkaloids, Terpenoids, Saponins, Flavonoids and Tannins. It indicates that most of the bioactive compounds are present in these varieties of chilli. So, the presence of phytochemical constituents plays a significant role in maintaining the vital health condition of the human body. The phytochemical can have complementary and overlapping mechanisms of oxidative agents, stimulating the immune system, regulator of gene expression in cell proliferation, and apoptosis [15]. The alkaloids show strong antioxidant activity. It has a wide range of pharmacological activities including antimalarial, antiasthma, anticancer, and antibacterial properties [16].



Table 1 Varieties of Mizo chilli showed the presence of preliminary phytochemicals

Mizo chilli varieties	Alkaloids	Terpenoids	Saponins	Flavonoids	Tannins
MZBEC-A	++	++	+	++	+
MZBEC-B	++	+	+	+	+
MZBEC-C	+	++	+	++	+

Table 2 Nutritional estimation of Mizo chilli varieties

Mizo chilli varieties	Ascorbic acid (mg/100mg±SD)	Proteins (mg/100mg±SD)	Carbohydrates (mg/100mg±SD)	Ash content (%±SD)
MZBEC-A	2.18±0.013	1.75±0.025	1.15±0.009	6.02±0.022
MZBEC-B	0.79±0.002	0.9±0.066	1.22±0.21	5.74±0.068
MZBEC-C	1.68±6.103	1.1±0.133	1.18±0.040	4.4±0.208

### Nutritional analysis

The nutritional estimation has demonstrated the potential benefits of Mizo chilli varieties. The proximal values were calculated against the standard curve and depicted in the (Table 2).

From (Table 2), it indicates that high amount of vitamin C is present in the fruits of Mizo chilli that varies from 0.79 mg/100mg of the sample to 2.18 mg/100mg of dry weight of the sample. From the result, MZBEC-A variety has the highest Vitamin-C content (2.18mg/100mg) followed by MZBEC-C (1.68mg/100mg) and MZBEC-B has the lowest Vitamin-C content (0.79mg/100mg). With conformity with the present finding Teodoro *et al.* [17] also reported the vitamin C contents in the range of 54.1mg/100g to 129.8 mg/100 g in Habanero pepper accessions (*Capsicum chinense*). Similarly, Orobiyi *et al.* [18] reported 84.64 -192.64 mg/100g ascorbic acid content in the fresh fruits of chilli pepper. From the result, the Mizo chilli varieties has high amount of Vitamin- C as compared to other chilli varieties. Proteins content was found far highest in MZBEC-A (4.22mg/100mg) as compared to MZBEC-B

(0.9mg/100mg) and MZBEC-C (1.1mg/100mg). But carbohydrates content was found highest in MZBEC-B (1.18mg/100mg) followed by MZBEC-C (1.18mg/100mg) and lowest in MZBEC-A (1.15mg/100mg). The Total Ash content of the sample was found to be highest in MZBEC-A variety (6.02%) followed by MZBEC-B (5.74%) and lowest in MZBEC-C (4.4%) in one gram of each sample. The total estimation of ash content indicates the presence of inorganic minerals by burning the organic substances in the samples. It is important because the amount of minerals can determine physiochemical properties of the chilli. From the experimental result, it collaborates with those of which state that plant morphological variation and fruit shape, as well as fruit colour, strongly depend on the cultivated chilli varieties [19]. These results also confirmed that nutrients vary from one stage to another because of climatic conditions, variety of cultivars and differences in analytical procedures [20]. The significant differences among the varieties were probably due to the different factors like soil texture, climate, growing conditions, cultivars, postharvest handlings and storage conditions [21].

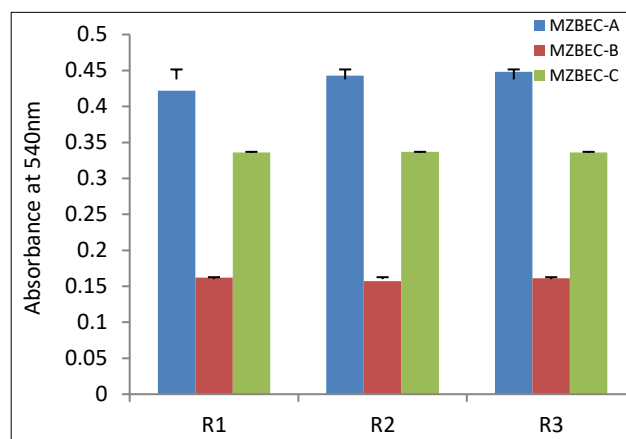
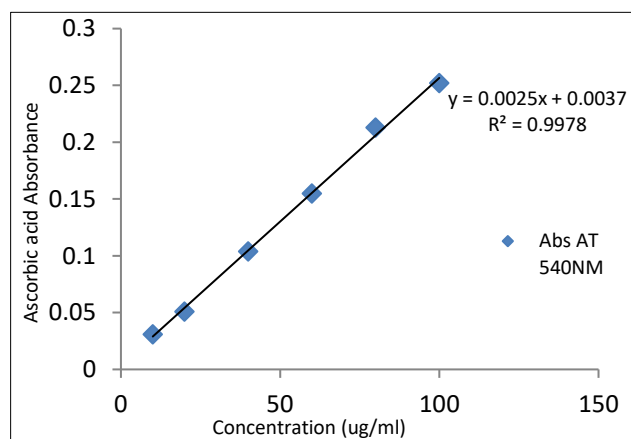


Fig 1 (a) Standard curve for Vitamin-C using Ascorbic acid (b) Total Vitamin-C content (mg/100mg of sample) in Mizo chilli varieties

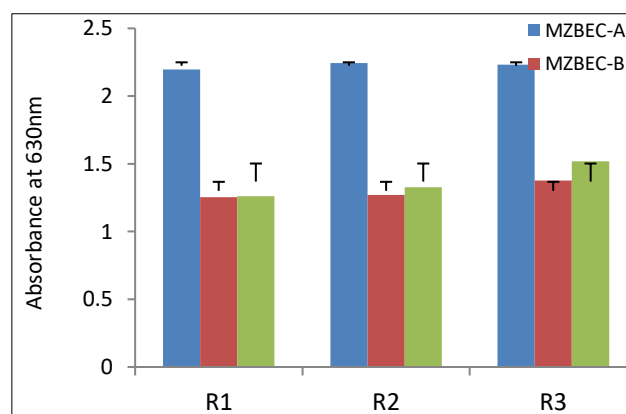
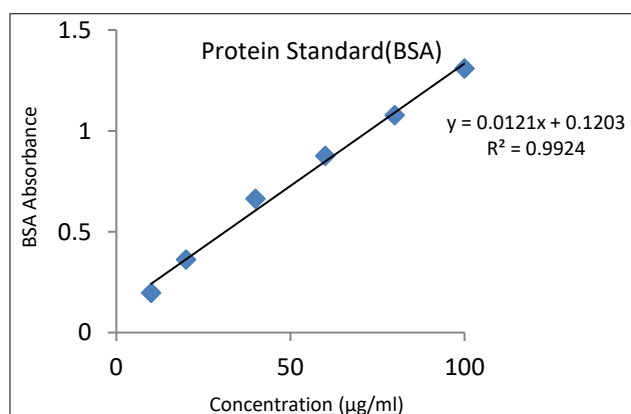


Fig 2 (a) Standard curve for proteins using Bovine Serum Albumin (BSA) (b) Total proteins content (mg/100mg of sample) in Mizo chilli



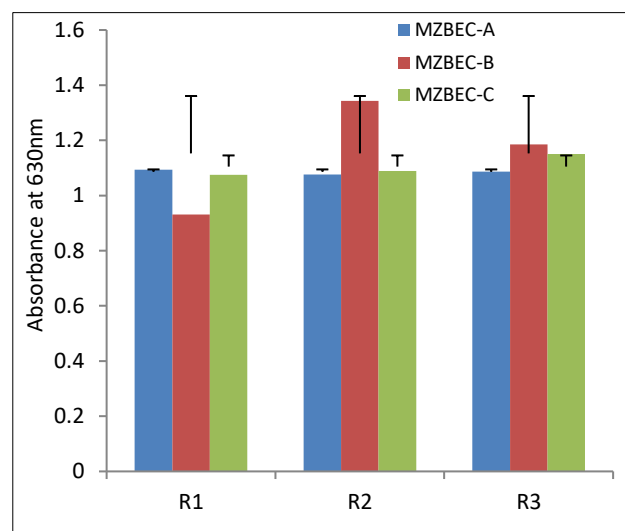
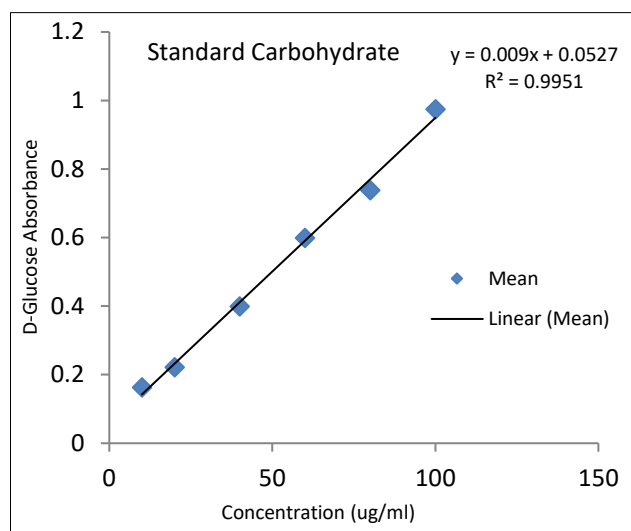


Fig 3 (a)Standard curve for Carbohydrates using D-Glucose (b) Total carbohydrates content (mg/100mg of sample) in Mizo chilli

## CONCLUSION

The fruits of Mizo chilli varieties were found to be rich in Vitamin-C, Proteins, Carbohydrates and Ash content and average amount of phytochemicals. There is a great diversity within the *Capsicum* species consumed in India. From the result, it indicates that Mizo chilli varieties can be considered to be a good source of nutrients and bioactive compounds. The variability presented in Mizo chilli's varieties germplasm for nutrition and bioactive substances can be exploited for breeding of new varieties with improved nutritional qualities. Considering the significance of Vitamin-C in human health and diseases, and its application in pharmaceutical product and commercial implication in food industry and current Covid-19 pandemic, Mizo chilli's offers great potential for future exploitation.

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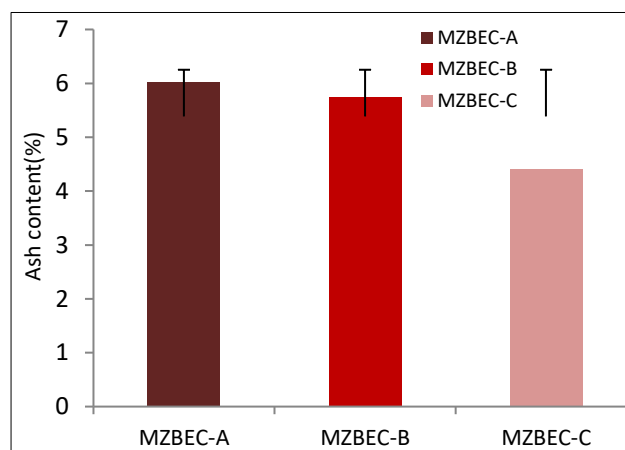


Fig 4 Total ash content (%) in Mizo chilli

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