

EVALUATION OF PHYSICOCHEMICAL CHARACTERISTICS AND PHYTOCHEMICAL PROPERTIES OF TOMATO (*Lycopersicon esculentum* L.) DURING DIFFERENT MATURITY STAGES

(Penilaian Ciri Fizikokimia dan Sifat Fitokimia Tomato (*Lycopersicon esculentum* L.) pada Tahap Kematangan yang Berbeza)

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Abstract

Tomato is an indispensable constituent of the daily food consumed in Bangladesh. The present study evaluated the physicochemical characteristics and phytochemicals present in tomato during different maturity stages, showing that pH and total soluble solids gradually increased with the decreased titratable acidity during different maturity stages of tomato. Chlorophyll content also decreased with the maturation of the tomato. Also, the anti-nutritional factors, tannin, phytate, and cyanogenic glycoside as well as antioxidants, such as vitamin C, were quantified, showing the highest amount of tannin, (2.61 ± 0.02 mg/100 g), phytate (4.28 ± 0.02 mg/100 g), and cyanogenic glycoside (7.81 ± 0.02 mg/100 g) in the green mature (unripe) stage, with the lowest values (1.16 ± 0.02 mg/100 g, 3.08 ± 0.01 mg/100 and 5.91 ± 0.02 mg/100 g, respectively) in the red (ripen) stage. In contrast, the maximum amount of vitamin C (25.07 ± 0.05 mg/100 g) was found in the red (ripen) stage, with the lowest amount (22.15 ± 0.22 mg/100 g) in the green mature stage. Overconsumption, that is, more than the daily recommended intake, of tomatoes may harm health but the concentration of anti-nutritional factors present in tomato was found to below toxic levels. Furthermore, vitamin C is a positive indicator of health and was in a high concentration in all maturity stages of tomato. In conclusion, this study revealed that the red ripened tomato is the best for human consumption compared to those green-yellow and green mature stages in terms of the physicochemical characteristics and phytochemical properties.

Keywords: tomato, maturity, tannin, phytate, cyanogenic glycoside

Abstrak

Tomato merupakan bahan utama yang diguna secara harian di Bangladesh. Kajian ini menilai ciri fiziko-kimia dan fitokimia yang hadir di dalam tomato semasa peringkat kematangan yang berbeza, menunjukkan nilai pH dan jumlah pepejal terlarut meningkat dengan pengurangan keasidan yang boleh dititrat semasa peringkat kematangan tomato yang berbeza. Kandungan klorofil juga berkurangan pada kematangan tomato. Juga, faktor anti-nutrisi, tannin, fitat, dan glikosida sinogenik bersama antioksidan, seperti vitamin C, telah dikuantifikasi, menunjukkan kandungan yang tinggi bagi tannin (2.61 ± 0.02 mg/100 g), fitat (4.28 ± 0.02 mg/100

g), dan glikosida sinogenik (7.81 ± 0.02 mg/100 g) pada peringkat belum matang (hijau) dengan nilai paling rendah telah ditemui (masing-masing 1.16 ± 0.02 mg/100 g, 3.08 ± 0.01 mg/100 g dan 5.91 ± 0.02 mg/100 g) pada peringkat matang (merah). Sebaliknya, jumlah maksimum vitamin C (25.07 ± 0.05 mg/100 g) dijumpai pada peringkat matang (merah) dengan kandungan paling rendah (22.15 ± 0.22 mg/100 g) pada peringkat belum matang, hijau. Pengambilan secara lebih melebihi cadangan harian, tomato mungkin membahayakan kesihatan tetapi kepekatan factor anti-nutrisi hadir di dalam tomato dijumpai lebih rendah dari aras toksik. Selanjutnya, vitamin C ialah indikator positif kesihatan dan ia mempunyai kepekatan yang tinggi pada semua peringkat kematangan tomato. Kesimpulan, kajian ini membuktikan bahawa tomato pada kematangan merah adalah terbaik bagi pengambilan oleh manusia berbanding kematangan hijau kekuningan dan hijau dari aspek ciri fizikokimia dan sifat fitokimia.

Kata kunci: tomato, kematangan, tannin, fitat, glikosida sinogenik

Introduction

Fruits and vegetables are important sources of nutrients for humans but in some lower-middle-income countries like Bangladesh, their production is periodic [1]. The tomato (*Lycopersicon esculentum* L.) is commonly consumed in Bangladesh as well as globally in garden-fresh or processed form [2]. Tomato farming in Bangladesh is of economic importance, accounting for approximately 68,366 acres with the production of 388,725 metric tons [3]. The tomato is a good source of nutrients and phytochemicals such as vitamin C, chlorophyll, phytate, tannins, cyanogenic glycoside that impact health [4]. Indeed, the tomato is a rich source of vitamin C, whose synergistic effects on wound healing are beneficial to health preventing scurvy [5]. Vitamin C is often used as a food additive due to its oxidation-reduction characteristics, reducing harmful agents present in food and to stabilize the color of food [6]. The maturation and superiority of tomato are predominantly based on color, with unripe tomatoes being green due to the high chlorophyll content, which degrades to yellow constituents, such as β -carotene and xanthophyll as the tomato matures, finally becoming red due to lycopene [7, 8]. Consumer and market demands influence the harvesting stage of the tomato [9]. Generally, the tomato is harvested during the mature green stage to full ripened stage until post-harvest damage considering various factors, for instance, cultivation time, growing environment, and market value [8, 10].

Furthermore, phytate, tannins, cyanogenic glycoside are present in the tomato in varying amounts. Excessive consumption of tannin in tomato reduces the digestion of several nutrients and prevents the body from capturing advantageous bioavailable elements [11].

Digestive enzymes, for example, amylase, pepsin, and trypsin are hindered by the consumption of a high amount of phytate in tomato [12]. Mono- and divalent cations K^+ , Mg^{2+} , and Ca^{2+} are phytate salts and stored in tomato seeds at different harvesting times. The high pH favorable for phytate has adverse effects on the bioavailability of divalent and trivalent mineral ions, such as Zn^{2+} , $Fe^{2+/3+}$, Ca^{2+} , Mg^{2+} , Mn^{2+} , and Cu^{2+} , resulting in mineral deficiency when large amounts of phytate in tomato are consumed [13]. Most of the people in Bangladesh are not aware of the presence of anti-nutritional factors in tomatoes and their effect on health due to lack of information. Indeed, to the best of our knowledge, this is the first study of the anti-nutritional factors in tomatoes in Bangladesh. The present study aimed to quantitatively assess the anti-nutritional factors, physicochemical characteristics, and other phytochemicals present in tomato during different maturation stages.

Materials and Methods

Samples and Chemicals

Tomatoes were collected from the field during the three maturity stages, with spotless, uniform tomatoes collected randomly for analysis. The collected samples were washed and stored at 4 °C for analysis. Chemicals used in this research were analytical grade.

Determination of pH

The pH of samples was determined using a pH meter (Model PCE-PH20M) at room temperature. An amount 10 g of sample were placed taken into 250 mL beaker, then homogenized with 100 mL distilled water (pH 7.00) in a blender. Finally, the pH electrode was put into

the suspension and allowed to stand until a constant pH was recorded.

Measurement of total soluble solids

The total soluble solids in the blended samples were measured using a digital refractometer (DR 6000T, Germany).

Estimation of titratable acidity

Titratable acidity was estimated according to the method described by Srivastavata and Sanjeev [14]. An amount

5 g of sample was blended with 15 mL distilled water for 3 minutes, then 3 mL of the sample was taken for titration against 0.1N sodium hydroxide using phenolphthalein as an indicator, with the presence of the pink color indicating the endpoint. The results were expressed as a percentage of citric acid.

Calculation of maturity

The maturity index was calculated using the formula given by Navez et al. [15] and Nielsen et al. [16]. The formula was used as shown in Equation 1:

$$\text{Maturity index} = [(\text{Total soluble solid}/20 \times \text{titratable acidity}) + \text{titratable acidity}] \quad (1)$$

Spectrophotometric measurement of chlorophyll

Chlorophyll a and Chlorophyll b were measured using the method described by Srivastavata and Sanjeev [14]. An amount 20 g of the sample was homogenized in 120 mL of distilled water using GEA lab homogenizer panda plus 1000 for 5 minutes. After filtration, the solution was centrifuged (Model: Rotina 380/380 R) at 6000 rpm for 5 minutes. After extraction of the pigments, 5 mL aliquot was placed in 25 mL of 90% acetone in a centrifuge tube and centrifuged at 2000 rpm for 3 minutes. The supernatant was removed and the optical density was recorded at 645 and 663nm respectively (Model: GENESYS 150 UV-visible spectrophotometer).

Measurement of vitamin C

The vitamin C content was measured using a 2,4-dinitrophenyl hydrazine (DNPH) method as described by Rahman et al. [17]. Briefly, 10 gm of sample was blended (Barum MR 404 plus), homogenized in 60 mL of 5% meta phosphoric acid-10% acetic acid solution, and transferred into a 100-mL volumetric flask and shaken lightly. The suspension was diluted up to the mark using a 5% meta phosphoric acid-10% acetic acid solution and filtered for the estimation of vitamin C. Bromine water was added to the solution until the solution becomes colored, excess bromine was removed by adding thiourea to give a clear solution. The standard solution of ascorbic acid was made from the stock solution as per the required dilution, before the addition

of 1.5 mL of DNPH solution to all standards and samples to oxidize the ascorbic acid. All standards, samples, and blank samples (without sample) were kept in a water bath at 37 °C for 4 hours to complete the reaction, then placed in an ice bath with the addition of 85% sulfuric acid before the optical density was measured at 521 nm.

Determination of phytate

Phytic acid was determined by the method described by Lucas and Markakas [18]. The sample (2 g) was dissolved in 100 mL of 2% concentrated hydrochloric acid for 4 hours and filtrated. Then, 50 mL of the filtrate was placed in a 250-mL beaker and 100 mL of distilled water was added to ensure adequate acidity. As an indicator, 10 mL of 0.3% ammonium thiocyanate solution was added and the solution was titrated using 0.00195 g iron/mL containing standard iron (III) chloride. The yellow-brownish color indicated the endpoint of the titration.

Measurement of cyanogenic glycoside

The alkaline picrate method of Onwuka [19] was used to measure cyanogenic glycoside. An amount 6 g of the samples were dissolved in 50 mL of distilled water and the cyanide was extracted overnight. Then, 4 mL of alkaline picrate was added to 1 mL filtrate sample and standard cyanide solution (different concentration) separately, before incubation in a water bath for 10 minutes and the optical density was recorded at 490 nm.

Measurement of tannin

Two gm of the sample was dissolved in 25 mL distilled water and stirred for 1 hour at room temperature. Then, the sample was centrifuged at 2500 rpm for 4 minutes and 5 mL of the supernatant was transferred to a 50-mL volumetric flask. An amount 5 mL standard tannic acid solution was prepared in another 50-mL flask. An amount 4 mL of saturated sodium carbonate solution and 2 mL folin dennis reagent were added to the sample solution and the standard solution respectively. The solution mixture was diluted to 100 mL and incubated for 90 minutes at room temperature before the optical density was measured at 250 nm. The percentage of tannin was calculated based on the formula of Jaffe [20].

Statistical analysis

The data are presented as the mean and standard deviation of three replicates using Microsoft Excel (2016 version) software.

Results and Discussion

Physicochemical properties play a vital role in the selection of tomatoes by the consumer. Consumer acceptance mostly depends on maturation which determines the physical quality of the tomato. Tomato maturation is influenced by acidity and total soluble substances as presented in Table 1.

The ratio of acidity and the total soluble solids was lowest in mature green tomatoes, indicating a minimum maturity index (6.096), whereas the red (ripen) tomatoes had the highest ratio of acidity and total soluble solids, indicating a maximum maturity index (9.263). The highest pH was found in red (ripen) tomatoes (4.93 ± 0.014), with the lowest pH observed in the green-yellow stage (3.10 ± 0.01). The pH value obtained from mature green (unripe) (3.42 ± 0.41) and green-yellow (half ripen) was below the value (3.7 to 4.1) as previously reported by Fagbohoun and Kiki [21].

A low pH (below 4.00) in tomatoes is unfavorable for the growth of pathogenic organisms as stated by Foolad [22]. Red (ripen) tomatoes was more perishable due to the high pH. The present study indicated that acidity gradually decreases with increased tomato maturation. The acidity ranged from $0.57 \pm 0.02\%$ to $0.36 \pm 0.26\%$

in line with a previous report by Fagbohoun and Kiki [21]. Sensory properties, for example, the essence of tomato is linked to the level of acidity [23], with the lowest acidity of tomato in the red (ripen) stage having the best flavor.

The quantity of soluble solids (Brix) is also a major parameter for evaluating tomato quality. The core soluble part of the tomatoes is the reducing sugar (fructose and glucose) according to Malundo et al. [24]. The present study recorded the lowest amount of soluble solids (5.92 ± 0.04) in mature green (unripe), with the highest amount (9.1 ± 0.17) in the red (ripen) stage. The highest quality tomatoes contained total soluble solids ranging from 4.8 to 8.8 as stated by Duma et al. [8]. The data presented in Table 1 showed that the acidity content decreases with the increase of soluble solids and the maturation stage of tomato. Significantly, low acidity and a high soluble solid content specify a high-quality tomato. The high pH is not a good quality marker due to the initiation of spoilage by pathogenic organisms, therefore, maturation (ripen) of the tomato is better determined in terms of physical quality.

Chlorophyll is responsible for the green color of a tomato, hence, the content of chlorophyll a and chlorophyll b was high in the green tomato, as shown in Figure 1. As expected, the chlorophyll content decreased in the green-yellow (half ripen) tomato and the tomatoes became more yellow as the chlorophyll was reduced as the tomato matured. The enzymes peroxidase and lipoxygenase are responsible for the degradation of chlorophyll, thereby enhancing the yellow color of the tomato [25].

The phytochemical properties of the tomato at different maturity stages are presented in Table 2. The highest amount of tannin was found in the mature green stage (2.61 ± 0.02 mg/100 g), gradually decreasing as the tomato ripened (1.16 ± 0.02 mg/100 g). A previous study revealed that a low amount (0.15 - 0.2 mg/100 g) of tannin present in food is advantageous to health [26] but excessive consumption of tannin acts as an anti-nutrient. The high level of tannins present in tomato may impede the actions of trypsin, chymotrypsin, amylase, and

lipase, thereby making the protein inferior quality as well as delaying iron absorption [27].

Tomato is a source of phytate, which prevents heart disease by reducing blood clots and cholesterol, as well as averting stone development in kidneys by removing heavy metal ions [28]. In the present study, the phytate content of tomatoes ranged from 4.28 ± 0.02 mg/100 g to 3.08 ± 0.01 mg/100 g. The daily intake recommended in developing countries is 2000 - 2600 mg for vegetarians and 150 - 1400 mg for mixed diets [29]. The consumption of excessive phytate may cause mineral deficiency due to a lack of the digestive phytase enzyme that is required for the absorption of iron, zinc, and calcium [30]. The level of cyanogenic glycoside was higher in all mature stages compared to tannin and phytate presented in Table 2. Phytate and tannin are heat resistant, whereas cyanogenic glycoside vanished at an

extreme temperature [31]. Tomato contains a high amount of vitamin C at different maturity stages, assisting in the deactivating effect of tannin on iron absorption [32]. The maximum amount of vitamin C was found in the red (ripen) stage (25.07 ± 0.05 mg/100 g), while the minimum amount was found in the mature green stage (22.15 ± 0.22 mg/100 g), which agrees with the previous observation of Ilahy et al. [33] and is in line with Dumas et al. [34] and Gautier et al. [35], who reported that the vitamin C content increases on ripening. In the ripening stage of tomato, a high amount of oxygen is absorbed by the cell as a consequence of the increased rate of respiration due to the anti-oxidative function of vitamin C.

Table 1. Physicochemical properties during different maturity stages of tomato

	pH	Acidity% (Citric Acid Equivalent)	Total Soluble Solids (Brix)	Maturity Index
Mature green (Unripe)	3.42 ± 0.41	0.57 ± 0.02	5.92 ± 0.04	6.096
Green-yellow (Half ripen)	3.10 ± 0.01	0.50 ± 0.02	7.34 ± 0.03	7.525
Red (Ripen)	4.93 ± 0.014	0.36 ± 0.26	9.1 ± 0.17	9.263

Data presented as mean $\pm$ standard deviation of three replicates

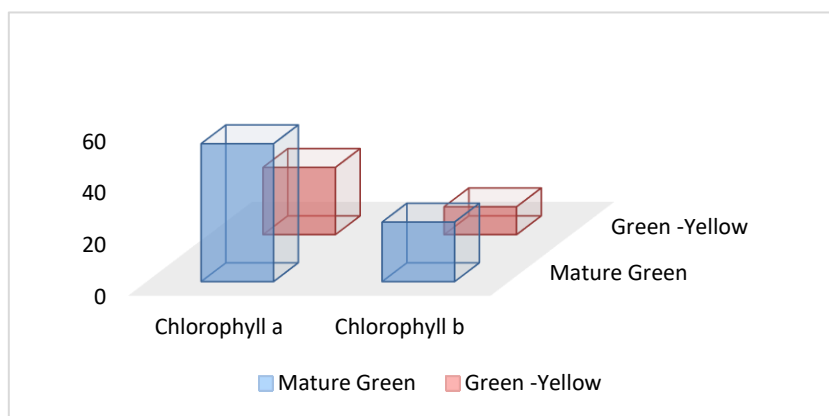


Figure 1. Comparison of chlorophyll a and chlorophyll b at different maturity stages of the tomato

Table 2. Phytochemical properties of tomato at different maturity stages

Sample	Tannin (mg/100 g)	Phytate (mg/100 g)	Cyanogenic Glycoside (mg/100 g)	Vitamin C (mg/100 g)
Mature green (Unripe)	2.61 ± 0.02	4.28 ± 0.02	7.81 ± 0.02	22.15 ± 0.22
Green-yellow (Half ripen)	2.06 ± 0.02	3.86 ± 0.02	6.2 ± 0.05	24.81 ± 0.02
Red (Ripen)	1.16 ± 0.02	3.08 ± 0.01	5.91 ± 0.02	25.07 ± 0.05

Data presented as mean ± standard deviation of three replicates

Conclusion

Tomato quality is highly influenced by the maturity stage, with mature tomatoes having a high soluble solid content and low titratable acidity. Furthermore, the chlorophyll content decreases with maturity, consequently, the tomatoes become less green as the chlorophyll degrades. Tomatoes contain anti-nutritional factors, tannin, phytate, and cyanogenic glycoside, the amount of which decreases as the tomatoes ripen, thereby confirming that consumers should select the best quality tomatoes for consumption. Future studies should identify and quantify other anti-nutritional factors present in tomatoes.

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