Proximate composition and sensory evaluation between artificially ripened and naturally ripened pineapples (*Ananas comosus*)

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Abstract

The number of artificially ripened pineapples is outnumbered than the naturally ripened pineapples. However, there is a lack of understanding between artificially ripened and naturally ripened pineapples. Thus the inquiry was anticipated to explore the physicochemical changes and organoleptic acceptability of the naturally ripened and artificially ripened pineapples. Farmers used different chemicals such as calcium carbide, ethylene, besides growth hormones to reduce production loss. Here we evaluated the content of moisture, ash, protein, fat, crude fiber, reducing sugar, total sugar, titratable acidity, sucrose, and vitamin C in both naturally ripened and artificially ripened pineapples. Artificially ripened pineapples showed a significantly lower vitamin C than naturally ripened ones, but arsenic content was nil in both samples. In the case of color and appearance, there was no significant difference between the two samples, but in case of the other organoleptic properties, such as flavor, sweetness, sourness, the natural one was more acceptable. Thus naturally ripened pineapples are more beneficial to consumers than artificially ripened ones.

Keywords: Pineapples, Ripening, Proximate composition, Sensory evaluation.

1. Introduction

Pineapples produced in the tropical and subtropical regions need a temperate climate and good rainfall. Based on respiration rate, fruits can be classified into climacteric fruits that produce more ethylene and non-climacteric fruits that produce less ethylene [1]. As a non-climacteric fruit, millions of tons of pineapples are produced worldwide. Although pineapple is classified as non-climacteric fruit, the peel color is increased like climacteric fruit, and the 20% yellow color stage is the best for harvesting the pineapple. Both natural and chemical ripening procedures are used for pineapples. However, different ripening procedures may have different nutrient values and sensory profiles, influencing consumers' choices [2]. Pineapples contain different vitamins, antioxidants, and enzymes with calcium-binding ability and anti-inflammatory properties. The final fruit ripening stage involves a sequence of biochemical and physiological occurrences prime to changes in texture, color, aroma, and flavor that shape the fruits more palatable, attractive, and tasty [3]. Moreover, reducing sugars, pH, sucrose, citric acid, alcoholinsoluble solids, and titrable acids in the pulp showed regular patterns of up-gradation during pineapple maturation [4]. Naturally ripened fruits are perishable: therefore, various chemicals like ethephon, ethylene, and calcium carbide that are hazardous to human health are being used to ripen and enhance the fruits' shelf-life. Artificial ripening may contaminate the fruit and may cause toxicity [5]. Consequently, these chemicals are responsible for different kinds of health burdens such as weakness, skin disease. kidney failure, cancer, lung failure, dizziness, ulcer, heart diseases [6]. Treatment with ethylene is less prone to ripening but responsible for the breakdown of the green chlorophyll to yellow color, making the fruit more colorful but less mature [7]. Artificial agents are used to reduce the production loss and attract the customer, while artificial growth hormones are used to produce larger fruit in size, including early harvesting during the off-season for more profit [8]. Alkaline, such as calcium carbide, is an ideal chemical for producing acetylene similar to ethylene, can cause stomach upset [9].

There are many varieties of pineapple available throughout Bangladesh. Honey Queen, Ghorasal, and Giant Kiew are the main three types of pineapple mainly cultivated in Bangladesh, and 'Asshina,' a local pineapple, is cultivated in the Tangail district. Tangail is on top by covering 49% of total pineapple cultivation land and 59% of the country's production. Farming experience, extension media contact, training on agrochemicals use, and knowledge on harmful effects of agrochemicals have significant relationships with their extent of use in pineapple farming, where economic profit is the key to using artificial ripening agents [10]. Different types of hazardous agrochemicals are used for pineapple cultivation in Bangladesh, creating health concerns for both the consumers and farmers [11]. Temperature plays an essential role in the ripening of fruits, and only one ppm ethylene is sufficient in the air for the ripening of the fruits [12].

At present, food safety is a burning question. Food adulteration has become a regular offense in many countries. Using chemicals such as ripening agents and growth hormones in pineapple is one of the most burning issues at the moment [13]. Therefore, it is essential to unmask the actual situation, harmful effects, and constituents of artificial ripening pineapples. The principal aim of the investigation was to increase the responsibilities of the government agencies in the sector of pineapple ripening process to control the use of artificial ripening agents, which are harmful to the human body and cause various types of chronic diseases. Moreover, this study also cast to enhance the

awareness of the fruit cultivators about the danger of chemical agents and imbue them with a sense of moral responsibility to society. The naturally ripened and chemically ripened pineapples used in this study were collected from Madhupur, Tangail. Thus, we investigated the compositional changes in artificially ripened pineapples with naturally ripened ones, including sensory evaluation's acceptance. We showed that naturally ripened pineapples contain more Vitamin C than artificially ripened ones. Nevertheless, there is no decrease in moisture, ash, protein, fat, reducing sugar, total sugar, titratable acidity, sucrose, arsenic, p^H (data not shown) in both artificial ripen and naturally ripen pineapples. Interestingly, crude fiber content was higher in artificially ripened pineapples than naturally ripened ones but not statistically significant. In sensory evaluation, naturally ripened pineapples showed more flavor, sweetness, and sourness than the chemically ripened ones. In a nutshell, the overall acceptance of the naturally ripened pineapples was significantly higher than chemically ripened ones. Thus, consumers' choices, nutrient values, and health concerns are important issues to consider in chemically ripened pineapples.

2. Materials and methods

Naturally and chemically ripened pineapples were collected from Madhupur, Tangail, cleaned with laboratory quality water, and preserved in a refrigerator for further experiment.

2.1. Collection of information

The information required to perform this study was collected from a formal discussion with farmers, observation, previous research, scientific publications, etc. The meeting

with farmers aimed to know the trade, cultivation, and chemical agents used in the production of pineapples. Accumulated information was scanned, classified, analyzed, and coordinated to prepare the working strategy. The physicochemical analysis of pineapple was conducted in the Department of Food Technology and Nutritional Science (FTNS) lab at Mawlana Bhashani Science and Technology University (MBSTU) and IFST, BCSIR. Information about the growth hormones and ripening agents, including practical information about pineapple cultivation, was collected from the farmers.

2.2. Physicochemical analysis

2.2.1. Determine the moisture content

Moisture content was determined as described previously [14]. Weight loss of the pineapples was conducted through evaporation at 105 °C.

2.2.2. Evaluation of ash content

Ash content was determined as described previously [15]. In brief, the sample was heated in Bunsen flame till smoke ceased and finally heated at 600 °C to burnt in a muffle furnace to white ash. After cooling via desiccator, the sample was weighed, and the amount of ash was calculated.

2.2.3. Evaluation of protein amount by Kjeldahl method

The protein amount was resolved as described previously [16].

2.2.4. Evaluation of the fat amount

The fat amount was determined as described previously [17]. In brief, the protein was digested by HCl to enhance fat release, which was extracted with petroleum ether. After evaporating the ether, the residue was weighed to calculate the fat content.

2.2.5. Evaluation of crude fiber content

The crude fiber was estimated as mentioned previously [18]. In brief, the defatted sample was taken and boiled for 30 mins with 200 ml sulfuric acid (1.25%) under reflux. After the rinse, the residue was treated and boiled for 30 min with 200 ml (1.25%) sodium hydroxide. The remnant was dried at 100 °C. After desiccator cooling, and the residue was burnt at 600 °C in the muffle furnace. After cooling, the residue was weighed to calculate the amount of fiber.

2.2.6. Evaluation of reducing sugar

Reducing sugar was determined by Fehling's solution as described previously [19]. A 5 ml pineapple juice was taken in a 100 ml volumetric flask and filled the volume with lab water and mixed efficiently. Fehling's solution (A: B = 5ml: 5ml) was added to the 100 ml volumetric flask. Next, heat and titrate with juice solution from the burette. The volume of the juice solution was noted down.

Reducing sugar (%) = (Dilutions × Fehling's Factor × 100)/(Weight of sample × titer value).

2.2.7. Estimation of total sugar

Total sugar was determined by modified Fehling's solution [20]. In brief, a 5 ml sample was taken into a conical flask and then added 50 ml distilled water. Next, a 5 ml

concentrated HCI was added and incubated for 24 hours in a cool, dry place. After 24 hours, it was made neutral by adding KOH and made 100 ml volume with distilled water. Next, it was titrated with Fehling's solution (A: B = 5ml: 5ml), including methyl blue indicator at the boiling temperature.

Total sugar (%) = (Fehling's Factor × Total volume × 100)/(Weight of sample × titer value).

2.2.8. Evaluation of titratable acidity

Titratable acidity was measured as described previously [21]. In brief, the sample was poured off into a conical flask, and then five drops of Phenopthaline were put in. Next, titrated with 0.1M NaOH and took the reading.

Reducing sugar (%) = (Number of ml of NaOH)/(Number of ml of Juice) \times 0.75

2.2.9. Determine the sucrose content

Sucrose content was estimated as described previously [20]. In brief, sugar was extracted by using 50% alcohol with lead acetate and dipotassium oxalate as the clearing agents. HCI inversion, and then warmed in the water bath were carried out. After dilution and filtration, the sucrose content was determined by Lane and Eynon's method with the standard Fehling's solution.

Sucrose (%) =
$$(TI-BI) \times 0.95$$
.

[TI = % of reducing sugars after inversion; BI = % of reducing sugars before inversion].

2.2.10. Evaluation of arsenic content

Arsenic content was determined using A.A.S. (Thermo-scientific Ice 3000 series Atomic Absorption Spectrometer) as described previously [22, 23].

2.2.11. Estimation of vitamin C content

Vitamin C was determined by Bessey's titrimetric method as described previously [24, 25]. In brief, standard vitamin C solution (10 ml) was added into a conical flask and titrated with the dye solution. Next, pineapple juice (4-6 g) was added and mixed well with metaphosphoric acids (3%) and filtered through a clean muslin cloth (double layered). Next, 10 minutes centrifugation of the filtrate at 3,000 r.p.m, and 2, 6 dichlorophenol indophenol solution was used to titrate the supernatant. By comparing with the titration results of the standard vitamin C solution, the amount of vitamin C present in the samples was determined.

Vitamin C (%) = (Vitamin C obtained × 100) / (Pineapple juice).

2.3. Sensory analysis of pineapples

Sensory evaluation was conducted in the department of FTNS conference room, MBSTU, as described previously [26]. Twenty semitrained undergraduate students of the department were asked to be the panelists in the study. To evaluate fruit preferences, the applied method was the questionnaire about the naturally ripened and artificially ripened pineapples, where a hedonic scale was executed to rate the degree of choice. Hedonic scale is an organoleptic quality rating scale where the inspectors convey the degree of choice. A 1 to 9 point balanced scale was applied. Overall inspections were performed by using a six-point hedonic scale. The organoleptic hedonic scale was used in the following order: 1 = dislike extremely, 2 = dislike very

much, 3 = dislike moderately, 4 = dislike slightly, 5 = neither like nor dislike, 6 = like slightly, 7 = like moderate, 8 = like very much, 9 = like extremely.

2.4. Statistical analysis

Data were analyzed using Graph Pad Prism and Microsoft Excel. The data were represented as mean \pm S.D (standard deviation). Statistical analysis was assisted by using student t-tests. p<0.05(*), p<0.001(***) were considered statistically significant.

3. Result and discussion

Tangail is famous for producing Giant Q and Honey Queen variants of pineapple, and wholesalers from across the country are flocked there to supply the sweet and sour juicy pineapples in the markets. The majority of the farmers in the study area cultivate Giant Kew, Honey Queen, Ghurashal, Singapuri variants of pineapple, and other different crops. Thus fruit variants, planting area, cost of input, working experience significantly affect pineapple production. Most of the farmers are illiterate and use agrochemicals to a great extent. Insufficient knowledge of farming, lack of training, and scarcity of high production with profit are the main reasons for using agrochemicals, especially growth hormones and artificial ripening agents [27]. In addition, farmers use chemicals to quicken the ripening time, enhance color and size, and reduce production loss. Fourteen different types of chemicals have been used during pineapple cultivation, from planting to storage, to prolong the shelf-life in the storage [28]. Different growth hormones and artificial ripening agents are usually used to grow more giant pineapples, resulting in losing the original taste. For example, Seprfix, Gibberellic acid, and Ethiopian are used as growth hormones, whereas calcium carbide and ethylene are

used as artificial ripen agents [29]. Ethephon (2-chloroethyl phosphonic acid) was the primary ripening agent in the study area.

Farmers follow the same procedure in the study area for commercial pineapple cultivation. They use compost, fertilizer, calcium carbonate to prepare their land. Besides compost, they use inorganic fertilizers such as Urea, Potassium phosphate, Zinc, and Triple superphosphate (T.S.P.). Growth hormones (e.g., 4.5% w/w α-naphthyl acetic acid) are commonly used after 15 to 30 days of plantation for the rapid growth of the plants, while used in the fruit after 21-22 days of its appearance. Moreover, insecticides are also used in the granular form at the earlier stage of the plantation. Pineapples in the Tangail district drop in the original flavor and taste because of the unrestricted use of growth hormones and chemicals. After harvesting, harmful chemicals such as calcium carbide, formaldehyde, ethephon, and superfix are being used to protect microbially and reduce production loss [30]. In this study, farmers were asked for a face-to-face interview while they were in their leisure period. They were conducted with a structured questionnaire and information related to this study, summarized in **Table 1**.

Table 1: Information about pineapple varieties, growth hormones, ripening agents, and land preparation was collected on a face-to-face interview with the farmers in the study area.

To expose the internal changes and the differences between the naturally ripened and artificially ripened pineapples, we evaluated the physicochemical analysis in the samples from the Giant Kew species pineapple. The moisture content was slightly lower

in the artificially ripened pineapples (81.20 ± 1.96%) compared to the naturally ripened ones (82.60 ± 2.2%), although statistically not significant. Likewise, the ash content was lower in artificially ripened pineapples (0.22 ± 0.06% Vs. 0.26 ± 0.05%) but not statistically significant. Moreover, the protein content was lower in artificially ripened pineapples (1.51 \pm 0.1%) than the naturally ripened ones (1.66 \pm 0.1%) but not statistically significant. Unlike moisture, ash, and protein, the fat content was higher in the artificially ripened pineapples (0.24 ± 0.02% Vs. 0.15 ± 0.03%), but statistically nonsignificant. In addition, the crude fiber amount was also higher in the artificially ripened pineapples (12.43 ± 0.28% Vs. 8.31 ± 0.7%), but statistically nonsignificant. Fasting blood sugar levels is an indicator of diabetes [31]. Moreover, chemically treated fruit contains a high amount of total sugar due to organic acids and starch hydrolysis to soluble sugars [32]. Thus it is essential to unmask the sugar content between naturally ripened and artificially ripened pineapples. Interestingly, there is no significant difference in reducing sugar (4.5 \pm 0.2% Vs. 4.6 \pm 0.1%) and total sugar (9.17 \pm 0.2% Vs. 9.41 \pm 0.3%) contents between the naturally ripened and artificially ripened pineapples. Thus we may conclude that the artificially ripened pineapples may not aggravate sugar levels compared to the naturally ripened ones. Interestingly, titratable acidity (a critical indicator of acid's influence on flavor) in naturally ripened pineapples (1.61 ± 0.2%) was higher than the artificially ripened ones (1.14 ± 0.2%), suggesting that precaution is decisive to chronic kidney disease patients in case of excessive pineapple consumption [33]. No arsenic was found in both samples. Importantly, vitamin C content was significantly (p<0.01) higher in naturally ripened pineapples (19.83 ± 1.1 mg/g) compared to the artificially ripened ones (14.56 ± 1.2 mg/g) (Figure 1), suggesting that naturally ripened pineapple is the better antioxidant reservoir, and chemical treatment may lead to the loss of vitamin C in artificially ripened pineapples. Importantly, vitamin C can repair DNA damage [34]. Fresh pineapples without chemical treatment contain bromelain that has anti-inflammatory actions, may reduce swelling in inflammatory conditions such as arthritis, acute sinusitis, sore throat, and can help recover from injuries and surgery [35].

Figure 1: Physicochemical analysis between naturally ripened and artificially ripened pineapples. Results were expressed as mean \pm (S.D.). Statistical analysis was executed using the t-test (*p<0.05).

Organoleptic tests depend on the smell, sweetness, appearance, color, flavor, sourness, and taste of the fruits [26]. The regular form of the organoleptic hedonic scale [36] was applied in this study. The comprehensive band score of both types of pineapples was common in essence, but their unique properties were found slightly different by the trial panelists. From the sensory test, we found that there were no significant differences in appearance $(6.79 \pm 0.65 \text{ Vs. } 6.53 \pm 0.86)$, and color $(6.05 \pm 1.26 \text{ Vs. } 6.57 \pm 0.86)$ between the naturally and artificially ripened pineapples, indicating that recognition of artificially ripened pineapples is not an easy task in an open eye. On the other hand, some studies reported that artificial ripening agents produce a more acceptable color than naturally ripened ones [37, 38]. Nonetheless, sweetness, flavor intensity, and volatile aroma positively impact the sensory acceptability of pineapples. Here, we found a significant difference in flavor $(7.1 \pm 0.99 \text{ Vs. } 6.05 \pm 1.26)$ between the naturally ripened and artificially ripened pineapples. Interestingly, naturally ripened pineapples were sweeter than artificially ripened pineapples $(6.37 \pm 0.92 \text{ Vs. } 5.21 \text{ Vs.$

0.81). Surprisingly, the sourness was also higher in naturally ripened pineapples than in artificially ripened ones (6.21 \pm 0.96 Vs. 4.73 \pm 0.74). The same scenario was also found in the case of overall acceptance (6.68 \pm 0.78 Vs. 5.37 \pm 0.69) between the naturally ripened and artificially ripened pineapples (**Figure 2**).

Figure 2: Different organoleptic quality parameters (Sensory Evaluation) of naturally ripened and artificially ripened pineapples. Data were expressed as Mean ± S.D. Statistical analysis was performed using the t-test (***p<0.001).

4. Conclusion

Our results delineate that artificial ripening agents and growth hormones are frequently used in pineapple cultivation, creating health concerns. Pineapples treated with different chemicals help to reduce ripening time and produce a more attractive color, but it is responsible for decreasing nutritional values. Moreover, our study points out that farmers are using chemical agents that are harmful to human health and prone to act as carcinogens [39, 40]. Vitamin C in the artificially ripened pineapples was significantly lower than in the naturally ripened pineapples. Some of the farmers are cultivated pure natural pineapple for their consumption. In addition, our study points out that there is no tolerance limit for chemicals in fruits in the study area. This study recommends that scientific communities find safer ways for ripening that minimize the loss, and consumers should be very selective in purchasing seasonal fruits. We also recommend complimentary transportation and proper cold storage facilities, mainly for seasonal fruits. Finally, compile involvement of all sectors people may help to reduce the problem of chemical use in fruit ripening.

Author contribution

R.B.R. ran the experiments, analyzed the data, and wrote the manuscript. R.T. contributed to the sample collection and experiments. M.A.Z. supervised the project. S.K. prepared the figures and table, rewrote, and edited the manuscript.

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Conflict of interest

The authors declare no conflict of interest.

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Table 1

A. Cultivation variants

Pineapple variants	Crops cultivated with pineapple
✓ Giant kew	√ Zinger
✓ Honey-queen	√ Banana
✓ Giant kew	✓ Edible root
✓ Ghurashal	
✓ Singapuri	

C. Land preparation

✓ Ingredients →	✓ Calcium Carbonate, Fertilizer, Compost
✓ Soil types \rightarrow	√ Bele duash
✓ Fertilizers →	✓ TSP, Urea, Potash, Zinc
✓ Flowering time→	✓ 18 months
✓ Maturity time →	✓ 3 months

B. Ripening agents

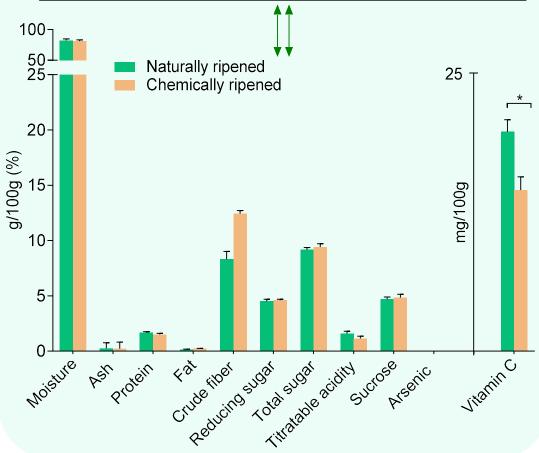
Hormones	Chemical agents	
	✓ Calcium carbide	
✓ 4.5% w/w α- napthyl acetic acid	✓ Ethephon	
	✓ Ethylene	
⊕ [Exposure duration] ⊕		
√ 15-30 days from flowering	✓ 20-30 days from flowering	
[Advantages]		

[Advantages]

- ✓ Increase size ✓ Enhance color
- ✓ Quick ripen √ Reduce transportation loss

Figure 1

Parameters	Naturally ripened (g/100g; n=3)	Chemically ripened (g/100g; n=3)
Moisture	82.60 ± 2.2	81.20 ± 1.96
Ash	0.26 ± 0.05	0.22 ± 0.06
Protein	1.66 ± 0.1	1.51 ± 0.1
Fat	0.15 ± 0.03	0.24 ± 0.02
Crude fiber	8.31 ± 0.7	12.43 ± 0.28
Reducing sugar	4.5 ± 0.2	4.6 ± 0.1
Total sugar	9.17 ± 0.2	9.41 ± 0.3
Titratable acidity	1.61 ± 0.2	1.14 ± 0.2
Sucrose	4.69 ± 0.3	4.81 ± 0.3
Arsenic	0.00 ± 0	0.00 ± 0
Vitamin C (mg/100g)	19.83 ± 1.1	14.56 ± 1.2*





Parameters	Natural ripened (Score; n=10)	Chemical ripened (Score; n=10)
Appearance	6.79 ± 0.65	6.53 ± 0.86
Color	6.05 ± 1.26	6.57 ± 0.86
Flavor	7.1 ± 0.99	6.05 ± 1.26***
Sweetness	6.37 ± 0.92	5.21 ± 0.81***
Sourness	6.21 ± 0.96	4.73 ± 0.74***
Overall acceptance	6.68 ± 0.78	5.37 ± 0.69***

