



# The Assessment of Physical and Microbial Properties of Traditional Fruit Leathers in Tehran

Payam Safaei<sup>1,\*</sup>, Zahra Sadeghi<sup>2</sup> and Gholamreza Jahed Khaniki<sup>2</sup>

<sup>1</sup>Student's Scientific Research Center, Tehran University of Medical Sciences, Tehran, Iran

<sup>2</sup>Department of Environmental Health, Faculty of Health, Tehran University of Medical Sciences, Tehran, Iran

\*Corresponding author: Department of Environmental Health, Faculty of Health, Tehran University of Medical Sciences, Tehran, Iran. Tel: +98-9187563104, Email: p.safaei2013@gmail.com

Received 2018 October 27; Revised 2018 December 02; Accepted 2018 December 15.

## Abstract

**Background:** Fruit leathers are nutritional products that are made by dehydrating a thin layer of fruit puree or juice under specific conditions, to obtain a chewy snack. Usually, the sun drying system is used to make traditional leathers, consequently takes a relatively long time and infections may occur during this period.

**Objectives:** The aim of this work was to assess the chemical and microbial quality of traditional fruit leathers.

**Methods:** In this study, 30 traditional sample leathers were obtained from the local market of Tehran. The pH, moisture, and microbiological load were measured in calculated samples. The pH was determined using a pH meter and moisture content values of all fruit leather samples were determined using the AOAC official method. For the assessment of aerobic mesophilic bacteria, yeast, and mold, dilutions were prepared and inoculated, followed by incubation for two and seven days at 37°C and 25°C, respectively. Also, this research used Green Bile Broth Brilliant Culture medium and most probable number (MPN) method for *E. coli* detection.

**Results:** The measured pH of the samples was in the range of 2.3 to 3.6 and 56% of the samples had a higher moisture content than the standard. Measuring the microbial load of the samples showed that they were over-contaminated in 16% of cases. Also, *E. coli* was identified in four samples.

**Conclusions:** This study showed that the high microbial load of traditional leathers, due to unhealthy production, is predictable. Thus, the correction of the traditional method for this product is recommended.

**Keywords:** Fruit Leather, Dehydration, Physical Properties, Microbial Properties

## 1. Background

Fruit leathers, also known as fruit bars are dehydrated, concentrated fruit-based products, made by certain kinds of fruits and classified as snack products (1, 2). Due to their attractive appearance and easy storage, fruit leathers are an effective way to increase fruit solids consumption, mainly for children and young people; therefore, producing fruit leather from fresh fruits is a useful way for the preservation of fruits. There are many types of fruits that are used in leather production, such as apple, blueberry, apricot, plum, jackfruit, banana, lemon, and orange (3). These products contain considerable quantities of dietary fibers, carbohydrates, minerals, vitamins, and antioxidants with less than 100 kcal per serving (4, 5). Fruit leathers are manufactured by drying a very thin layer of fruit puree or a mixture of fruit juice concentrated to a leather-like sheet with or without additives. However, in the leather production process, humidity is removed from the wet puree or fruit juice by direct sunlight or industrial dryers (2, 6). A good fruit leather contains low moisture (10% to 20%), intermediate water activity (less than 0.7), and chewy texture, and can be consumed directly (7).

To make traditional leathers, sunlight is more commonly used for drying. Generally, incorrect drying methods cause damage to the quality of the last product, which makes it unsuitable for consumption (8). Furthermore, dehydration of the leather is usually carried out below 80°C, in order to keep a better final quality (9), which leads to stability of microbial activity during and after the preparation (10). Quality changes, such as a microbial growth that occurs during storage, can be avoided by cold storage and proper packaging (11). On the other hand, many diseases are caused by consumption of contaminated food. Microbial foodborne disease cases continue to take a considerable public health toll, primarily in developing countries. According to recent World Health Organization (WHO) reports, at least 600 million cases of foodborne illness and 420000 associated deaths occur each year, and monitoring of food products can prevent them from occurring (12). In the recent years, the popularity of leathers has increased. They are becoming an industrial and economic product that should receive more attention for their quality. Packaging products for fruit leather are needed to extend shelf-life, and is related to the stability of water activity, microbiological stability, sensory properties, and physicochemical

attributes (13).

## 2. Objectives

The objectives of this study were to evaluate the chemical and microbial properties of fruit leathers in order to determine their quality and therefore to prevent foodborne diseases. The result is expected to provide useful information about this product.

## 3. Methods

### 3.1. Materials

Thirty traditional samples including pomegranate, apricot, plum, apple, kiwi, and cherry fruit leathers were obtained randomly from different local markets of Tehran. Leathers were sorted at room temperature until further analysis. The samples obtained under the optimum conditions were submitted to physicochemical and microbial analyses.

### 3.2. Moisture Content and pH

The pH was determined using a pH meter (S20 SevenEasy, Mettler Toledo, USA) that was previously calibrated with pH 4 and pH 7 tampon solutions, according to a standard (AOAC 981.12, 1998) (14). Overall, 10 g of leather sample was homogenized in water and after preparing the 10% solution, pH was measured.

Moisture content values of all fruit leather samples were determined using the AOAC official method (15). According to this method, the moisture content of the samples was determined by drying in an oven at 104°C for four hours. First, 10 g of the sample was taken and left in an oven at 104°C, until constant weight was reached (16). The sample weight was measured before and after placement in the oven, and thus the moisture content of the sample was calculated according to the following formula.

It should be mentioned that the standard levels of moisture should be lower than 15% and this standard for pH is 2.5 to 4.5.

$$M = \frac{w_1 - w_2}{w_s} \times 100 \quad (1)$$

M = amount of moisture

w<sub>1</sub> = the weight of sample before placement in the oven

w<sub>2</sub> = the weight of sample after placement in the oven

w<sub>s</sub> = sample weight

### 3.3. Microbiology Tests

Aerobic mesophilic bacteria, *Escherichia coli*, mold, and yeasts counts, was determined according to the guidelines of the American Public Health Association (17). First, a range of sample dilutions were prepared. To prepare a 1/10 dilution series, the researchers mixed 10 g of each sample in a dilution bottle containing 90 mL of normal saline. In this way, a 1/10 uniform dilution of the samples was obtained.

### 3.4. Aerobic Mesophilic Bacteria Count

Sample suspension (1 mL) from fruit leathers with 1/10 dilution was pipetted onto the surface of a sterile plate (two plates for each dilution) in the vicinity of the flame. In the next step, 15 mL of agar solutions at a temperature of less than 45°C was poured on sterile petri-dishes, and then the plates were gently swirled until the sample was mixed with the culture medium and dried. Plates were placed in an incubator at 37°C for 48 ± 3 hours. After the incubation, the number of colonies per plate was counted for 24 and 48 hours.

### 3.5. *Escherichia coli* Detection

For *E. coli* analysis, the most probable number (MPN) method was used. The MPN conditions were as follows: Preparation of the three dilutions of tubes containing BGLB broth culture (10, 1, and 1/10). Examination of the tubes was done to make sure that the inner vial was full of liquid with no air bubbles. Plates were shaken gently to mix the sample with the medium and incubated for 48 hours at 37°C. After 24 hours, the tubes were examined for gas production or color change. If no tubes appeared positive, re-incubation up to 48 hours was done. Using a sterile pipet, the inocula were transferred from positive tubes to the confirmation medium (the tubes containing peptone water) and incubated for 48 hours at 44°C. After incubation, 0.5 mL of indole reagent was added to tubes and mixed well. After one-minute, color change to red confirmed results and was regarded as positive.

### 3.6. Total Yeast and Mold Count

Total yeast and mold count were determined using the Sabouraud Dextrose Agar (SDA) culture medium. One milliliter of prepared food dilution was pipetted onto the center of the Petri dish. Then, 15 mL of liquid medium (45°C) was added to plates and mixed slowly. The inoculated plate was incubated at 25°C for five to seven days and the number of colonies was counted. The results of Green-smith showed a value of 1000 CFU/gr (yeast and moulds) as the maximum acceptable limit for dehydrated fruits (18).

### 3.7. Statistical Analysis

Statistical analyses were carried out using SPSS software 16.00 (SPSS Inc., Chicago, IL, USA). The results were compared with one-sample *t* test analysis.

## 4. Results

### 4.1. Moisture Content and pH

Mean ± standard deviation of pH and moisture content of samples are shown in Table 1. According to the results, apricot and the mixture of plum and apple leathers had the highest and lowest moisture among each of formulations, respectively. Also, maximum pH was related to

**Table 1.** Mean  $\pm$  Standard Deviation of pH and Moisture Content of Samples

Fruit Leathers	Moisture (%)	Rang (Min - Max)	pH	Rang (Min - Max)
Plum fruit leathers	17.27 $\pm$ 3.63	12.5 - 21.2	3.25 $\pm$ 0.29	2.7 - 3.6
Kiwi fruit leathers	13.83 $\pm$ 4.14	9.1 - 16.8	2.46 $\pm$ 0.05	2.4 - 2.5
Cherry fruit leathers	14.75 $\pm$ 2.36	11.1 - 18.5	2.50 $\pm$ 0.06	2.4 - 2.6
Pomegranate fruit leathers	15.96 $\pm$ 3.61	13.14 - 9	2.32 $\pm$ 0.03	2.3 - 2.4
Apple fruit leather	14.66 $\pm$ 1.52	13 - 14.2	2.53 $\pm$ 0.11	2.4
Apricot fruit leathers	20.76 $\pm$ 1.36	19.3 - 20	2.80 $\pm$ 0.20	2.6 - 2.7
Plum and apple leathers	13.05 $\pm$ 1.34	12.1 - 12.8	2.66 $\pm$ 0.11	2.6

**Table 2.** Microbiological Feature of Samples

Fruit Leather	Total Yeast and Mold Count	Aerobic Mesophilic Bacteria Count
Plum fruit leather	10 - 2.9 $\times 10^2$	10 - 3 $\times 10^2$
Kiwi fruit leathers	3 $\times 10$ - 3.1 $\times 10^2$	10 - 3 $\times 10^2$
Cherry fruit leathers	< 10 - 4 $\times 10^2$	10 - 3 $\times 10^2$
Pomegranate fruit leathers	10 - 2 $\times 10^1$	< 10 - 4 $\times 10^1$
Apple fruit leather	1 $\times 10^2$ - 1.3 $\times 10^2$	10 - 1.2 $\times 10^1$
Apricot fruit leathers	< 10	1.4 $\times 10$ - 4 $\times 10^1$
Plum and apple leathers	10 - 1.4 $\times 10^1$	< 10 - 10

plum fruit leathers with a mean of 3.25, and pomegranate fruit leathers with an average value of 2.32 have a minimum pH.

#### 4.2. Microbiology Tests

The microbiological feature of samples, including total yeast and mold, and aerobic mesophilic bacteria, are presented in Table 2. The results showed that total yeast and mold in samples were in the range of  $\leq 10$  to  $4 \times 10^2$ . Cherry and apricot fruit leathers had maximum and minimum range of total yeast and mold, respectively. In addition, the range of aerobic mesophilic bacteria count was  $\leq 10$  to  $3 \times 10^2$ . The results indicated that a mixture of plum and apple leathers had a minimum value amongst samples.

### 5. Discussion

#### 5.1. Moisture Content and pH

According to the standards, the moisture content of leather must be lower than 15%. Suna et al. showed that a moisture content of leather below 15% prevents microbial growth (19). Due to the fact that in the traditional process of leather (sun drying) it is not possible to control the drying conditions, and the completion time of the process is determined by observing the tissue and not the laboratory test, high humidity is probable in some samples. The results of moisture content of fruit leathers were similar to the results of Torres et al. who found that the moisture

content of their fruit leathers was 18 kg water/100 kg of last products (5).

According to the standard, the pH of the leathers must be between 2.5 and 4.5. In this study, the mean pH was 2.75 (Table 1) and was below the lower limit for bacterial growth (4.0), yet could allow the growth of some fungi and yeasts (20). This may be related to the use of edible acids during the production process. Thus, these leathers are expected to have a stable shelf-life for several months without the need for chemical preservatives. In another study, the mean pH for apples was 3.6 and 3.3 in quince fruit leathers. However, these amounts are lower than the bacterial growth limits (5).

Low moisture content and pH can inhibit microbial growth and prolong shelf-life, which subsequently affected the consumer's health.

#### 5.2. Microbiology Tests

Moisture and low pH of the leathers cause unfavorable conditions for the growth of common microorganisms, yet some of them, such as aerobic bacterial spores, lactobacillus, yeast, and molds are more resistant than other microorganisms to grow and survive in these conditions. Similarly, the results of Radmard Ghadiri and Kalbasi Ashtari showed that microbial flora of apple leather mainly included aerobic bacterial spores, lactobacillus, yeast, and molds (21). According to the standard, the number of aerobic bacteria, as well as total mold and yeast should be less than 200 items per gram. The results proved that 16% of samples have higher microbial load than the standard. The results were also consistent with the results of a previous study, performed with durian leather samples. In that study, the storage stability of durian leather at room temperature was tested. Microbial analyses showed that Total Mesophilic Bacteria (TMB) and Total Molds and Yeast (TMY) counts were low, where after a 12-week storage, TMB and TMY were less than 60 and 140 cfu g<sup>-1</sup>, respectively; this could be due to the secondary contamination (13). In another study, apples and quince leathers were obtained from local producers in Chile. The microbiological test showed that aerobic mesophilic bacteria, yeast and mold, *Escherichia coli*, and *Staphylococcus aureus* counts were less than 10 CFU/g in all fruit leathers after accelerated storage at 30°C (5).

These results were expected since leathers had low pH and intermediate moisture. The *E. coli* detection test showed that the bacteria was present in samples 2, 3, 21, and 25. These bacteria are fecal coliform and can result in inappropriate sanitary conditions during the preparation as well as unsuitable packaging during product transportation, which leads to food-borne disease. Common symptoms of foodborne diseases are diarrhea, vomiting, and nausea. Such diseases can also have neurological, immunological, even cancer, and other symptoms (22).

### 5.3. Conclusions

Fruit leathers are a healthy alternative to junk foods, particularly for children, due to their texture and nutritive value. Considering the low pH of fruits, aerobic bacterial spores, lactobacillus, yeast, and molds are considered as corruption factors. Cross-contamination can increase the corruption and microbial load. The high microbial load of 16% of the samples in this test compared with the standard can be due to one of these issues. Also, the presence of *E. coli* as a fecal contamination indicator is a critical risk. Therefore, due to the lack of a test for confirmation of the traditional leathers, according to the standard and difference in their quality, the necessity of using a health product that is manufactured in accordance with the standard is needed. Compared to traditional methods, a new drying technique recommended for high-quality leather production. The data from this paper will be useful in the food industry and for consumers, who are health-conscious.

### Acknowledgments

The authors would like to thank the Department of Environmental Health and Tehran University of Medical Sciences for their support in carrying out the project.

### Footnotes

**Authors' Contribution:** Payam Safaei: Analysis and interpretation of data, writing the manuscript. Zahra Sadeghi: Contributed to the development of the protocol and statistical analysis. Gholamreza Jahed Khaniki: Study concept and design.

**Conflict of Interests:** The authors of this article declare that they had no conflict of interest.

**Ethical Considerations:** The ethical approval code was 9411439004.

**Funding/Support:** This research was supported by the Tehran University of Medical Sciences.

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