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Impact of appertization on the physicochemical, phytochemical properties, nutritional and microbiological qualities of a lemon-enriched tomato juice

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Abstract

Aim: This study attempts to offer a viable and sustainable solution related to the tomato value chain, which plays an active role in human diets but deteriorates very fast due to its short shelf life.

Methods: Fresh lemons (*Citrus limon*) and tomatoes (*Solanum lycopersicum* L.) were purchased from the local market. Previously, varying percentages of lemon juice (0%, 1%, 3%, 5%, and 10%) were added to clear jars containing peeled and pasted tomatoes, which were then sterilized. The physicochemical, antioxidant, nutritional, and microbiological characteristics of appertized tomato samples were assessed through the use of standardized techniques.

Results: The addition of lemon juice significantly ($P < 0.05$) reduced the total phenolic content of appertized tomatoes, while increasing the titratable acidity ($P < 0.05$) and decreasing the hydrogen potential (pH) content ($P < 0.05$). However, the addition of 10 g of lemon juice recorded the high flavonoid content (0.01 mg CE/g) and carotenoid content (16.52 mg/100 g) of samples. In terms of nutritional value, adding lemon juice to appertized tomatoes considerably reduced ($P < 0.05$) their protein content while increasing their carbohydrate content. Regarding the mineral composition, the addition of lemon juice considerably ($P < 0.05$) raised the amounts of calcium (Ca), phosphorus (P), and magnesium (Mg) in the appertized tomato samples. The results of this investigation fall within the ranges of the daily allowances that are advised. Pathogens including *Salmonella*, *Clostridium*, and *Escherichia coli* are inhibited, and yeasts and molds are destroyed, ensuring the product's microbiological quality [476.57 to 0 colony-forming unit (CFU)].

Conclusions: Lemon juice helps to preserve consumer health and improve the preservation of appertized tomatoes.

Keywords

Appertized tomatoes, lemon juice, antioxidant properties, pathogens inhibition, nutritional quality

Introduction

The tomato (*Solanum lycopersicum*) is a plant species belonging to the family of Solanaceae, widely cultivated for its fruit, which makes it one of the most consumed vegetables in the world. It is highly appreciated for its high content of minerals, vitamins, proteins, essential amino acids (leucine, threonine, valine, histidine, lysine, arginine), monounsaturated fatty acids (linoleic and linolenic acids), carotenoids (lycopene and β-carotenoids) and phytosterols (β-sitosterol, campesterol and stigmasterol) [1], as an important source of income for farmers [2]. Under rainfed conditions, its cultivation is limited during the dry months of the year and abundant during the wet months. However, being a very perishable crop, losses are usually high after harvest. This results in both food waste for consumers and financial losses for producers, estimated to be about one-third of the production of some tropical countries like Cameroon [3]. Hence, some countries resort to importing this vegetable as canned food to ensure their food security. These canned foods are often packaged with chemical additives that often result in some dysfunctions (allergies, digestive disorders, neurological disorders) and damage to some physiological organs in some consumers [4].

In resource-poor communities, local producers and small and medium-sized enterprises (SMEs) resort to the use of basic techniques (drying, salting, sugaring, dehydration, etc.) to preserve the vegetables while richer operators use more sophisticated cold storage methods, which, although more efficient, are costly and do not always fully guarantee the long-term preservation of their quality [5]. To address this limitation, combinations of other methods have been used, like some quantities of common salt and lemon juice to preserve the color and the tomato's bioactive compounds [6]. Some processors use more efficient hot preservation canning techniques that seem to preserve the food for slight periods, of up to six months [7]. Indeed, despite some fluctuations observed in the food's composition, canning allows for the preservation of the physico-chemical and nutritional properties of tomato paste [8]. With the increasing demand worldwide for organic foods whose cultivation is more environment-friendly, synthetic preservatives such as ascorbic acid (E300) and citric acid (E330) could conveniently be substituted with canned tomatoes with other natural products like lemon juice [9].

The lemon [*Citrus limon* (L.) Burm f.] is considered the third most important citrus species in the world, only behind orange and tangerine, and is socially known for its valuable nutritional, medical, pharmacological, industrial, and cosmetic properties and uses [10]. In Cameroon, in the locality of Bokito, the Centre region, lemons are produced abundantly and consistently, unlike other citrus fruits where two peaks of production are observed per year (June–July and October–November). These lemons are used in the composition of several cooking recipes or traditional pharmacopoeia, and they provide income to producers, especially during the production period from October to November, coinciding with a very high demand for lemons in the Yaoundé market [11]. In addition, Djeuga Youga et al. [12] revealed that lemon juice is used to treat stomach ache in the Western highlands of Cameroon. The lemon (*Citrus limon*) has a high concentration of citric and other organic acids [13] that could effectively control the proliferation of some micro-organisms and even inactivate *Escherichia coli* (*E. coli*) bacteria [14], and inhibit *Staphylococcus aureus* and *Pseudomonas aeruginosa* growth [15]. Thus, limiting the losses within this highly prized food product via viable processing or preservation techniques becomes important, and this would undoubtedly also contribute to the achievement of the objectives of Sustainable Development Goals, particularly the fight against hunger and poverty [8]. This study was therefore undertaken as an alternative to synthetic

preservatives used in the canning of tomato paste while improving their nutritional and microbiological qualities.

Materials and methods

Materials and sample preparation

The plant materials used in this study were fruits of the *Rio Grande* tomato variety, which are bright red in color, mature, firm, round, and medium-sized with no visible signs of infestation. This variety is the most commercialized in the market and the most popular with consumers. The lemon (*Citrus limon*) used was light green, mature, and also without any visible signs of pest attacks. All chemicals and reagents used were of analytical grade. After collection at Mfoundi market of Yaoundé, Centre region of Cameroon, the tomato and lemon samples were transported in food bags to the laboratory, sorted the crushed soft ones from those of good firmness, and then graded according to sizes to retain only those of the same size and shape for the preparation of appertized tomatoes. As shown in Figure 1, which summarizes the steps used for sample preparation, tomatoes and lemon were cleaned with potable water, soaked separately in potable water for 1 h, thoroughly washed with distilled water, and then placed in separate water baths (Memmert WNB 29, Germany) at 100°C for 30 s. Thereafter, they were retrieved and soaked in cold water to facilitate the peeling off of their outer shells, and the peeled tomatoes were then cut into thin strips 3 mm thick, stripped of their seeds, and crushed using a domestic blender (Moulinex®, France). The pastes obtained were placed in previously cleaned and sterilized transparent glass jars. Lemon juices that had been prepared in parallel were introduced in increasing proportions: 0% of lemon juice (AT0), 1% of lemon juice (AT1), 3% of lemon juice (AT3), 5% of lemon juice (AT5), and 10% of lemon juice (AT10). These correspond to 0 g, 1 g, 3 g, 5 g, and 10 g, respectively, of lemon juice in each jar to have a final mass of 100 g (w/w). Each jar was then tightly sealed, shaken vigorously, and sterilized in an autoclave (SANOclav™, Germany) for 45 min at 120°C (with F0 value of 35 min). After sterilization, each jar was retrieved from the autoclave, allowed to cool in cold water, and then stored away in the dark.

Determination of physicochemical properties

The hydrogen potential (pH) which provides information on the total acidity of a sample, was determined according to Boumendjel et al. [16] using a calibrated pH meter (Hanna edge, USA) operating at 25°C.

The titratable acidity of appertized tomato samples, which gives the total natural organic acids content, was carried out by titrating with a strong base (0.1 mol/L NaOH) per change in a colored phenolphthalein indicator. The concentration of organic acids was determined by titrating a test portion with NaOH to a pH of 8.1. Citric acid monohydrate which is the predominant acid in tomatoes, was used in the expression of results according to the standard NF V05-501 method [17]. The titratable acidity is expressed in mmoles of hydrogen ion (H^+) per liter of product.

Proximate composition analysis

The AOAC standard analytical methods were used for the determination of the moisture, ash, protein, and fat contents on wet-based tomato samples [18]. The moisture content was determined by drying the samples in an oven at 105° ± 2°C to constant weight, which was achieved according to the AOAC procedures 925.40. The ash content was evaluated by incineration of the samples for 2 h at 550°C according to the AOAC procedures 942.05. The nitrogen (N) content was analyzed using the micro-Kjeldahl method according to AOAC procedures 984.13, and the crude protein content was calculated as N × 6.25. The lipid content was determined using the Soxhlet method, following the AOAC 963.15 methodology. The fiber content was assessed following the AOAC method [19]. The carbohydrate content was obtained via subtraction [18]. In this context, the crude protein, lipid, moisture, ash, and fiber contents were deducted from 100 to obtain the carbohydrate content. The values of proximate composition analysis were expressed in percentage (%).

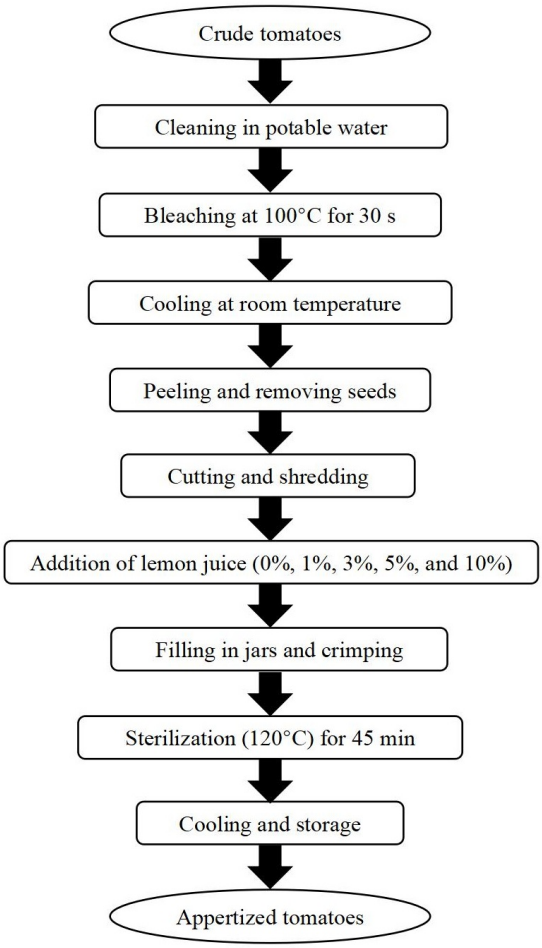

Figure 1. Diagram of the production of canned tomatoes

Mineral content analysis

To determine the mineral content [calcium (Ca), sodium (Na), potassium (K), phosphorus (P), magnesium (Mg), and iron (Fe)] of appertized tomato pastes, accurately weighed test portions were first ashed at 550°C, and the ash obtained was then boiled with 10 mL of 20% HCl in a beaker and later filtered into a 100 mL standard flask. The P content was determined using a PerkinElmer UV-Visible Spectrophotometer (Norwalk CT, USA) [19], while the mineral content of the digested samples was determined by flame atomic absorption spectrophotometry using a Varian 220FS Spectra AA apparatus (Varian, USA) for the rest of the elements (Ca, Mg, Na, K, and Fe). The mineral content of each sample was determined from calibration curves of standard minerals.

Evaluation of phytochemical contents

Before the determination of the phytochemical content, 10 mL of appertized tomato from each sample was filtered using Whatman No. 1 paper, and the filtrate was oven-dried (BMT Medical Technology, Czech Republic) at 40°C to constant mass to obtain extract.

The total phenolic content (TPC) of the canned tomato samples was determined via the Folin-Ciocalteu colorimetric method, as described by Gao et al. [20]. In a 5 mL test tube, 20 µL of a 2 mg/mL aqueous extract solution was added, followed by 0.2 mL Folin-Ciocalteu reagent and 2 mL distilled water. The whole was incubated for 3 min at room temperature. Thereafter, 1 mL of 20% Na_2CO_3 solution was added, and the mixture was further incubated for 20 min under the same conditions. The absorbance of the resulting blue-colored solution was measured at 765 nm via a spectrophotometer (BioMate, Germany). The TPC of the extract was calculated from the gallic acid standard curve and expressed as milligrams of gallic acid equivalent (GAE) per gram of extract.

The flavonoid content was determined according to the method described by Marinova et al. [21]. Here, approximately 0.1 mL of extract was mixed with 1.4 mL of distilled water and 0.03 mL of 5% $NaNO_2$ solution, and the whole was stored at room temperature for 5 min. Thereafter, 0.2 mL of 10% $AlCl_3$ solution was added and the whole rested at room temperature for a further 5 min. Then, 0.2 mL of 10% NaOH solution and 0.24 mL of distilled water were added, and the absorbance was measured at 510 nm via a spectrophotometer (BioMate, Germany). The flavonoid content was determined via a standard curve obtained with catechin and the contents were expressed as mg catechin equivalent (CE) per gram of extract.

The total carotenoid content was evaluated using a modified spectrophotometric method as described by De Leenheer et al. [22]. To extract the carotenoids, a 1 g portion of appertized paste was poured in 10 mL of a hexane-acetone mixture (30/70; v/v), and the whole was stirred for 1–2 h and then filtered using a Whatman No. 1 paper. The filtrates were then transferred to tanks and read on a spectrophotometer (BioMate, Germany) at 450 nm against vitamin A used as the blank solution. Calibration ranges formed by pure vitamin A were used and the results are expressed as mg/100 g of wet matter.

Microbial analysis

For microbial enumerations, 25 g of each appertized tomato sample was ground under aseptic conditions with 225 mL of peptone saline using a Stomacher® 400 circulator (United Kingdom) homogenizer and rested at room temperature for 30 min. From this preparation, serial dilutions were made to determine the presence of yeasts and molds, *E. coli* bacteria, *Salmonella* spp., and Clostridia.

Yeasts and molds were quantified at 25°C in potato dextrose agar (PDA) over 72 h. The detection of *E. coli* in appertized tomato samples was performed using a liquid selective enrichment medium (selective lauryl sulfate broth) with a determined amount of the initial suspension of the test sample at 37°C for 48 h. A subculture was made in a tube containing free peptone water and the tube was examined for indole production resulting from the degradation of tryptophan into peptone constituents [23].

The presence of *Salmonella* spp. in the samples was determined via the method proposed by ISO 6579-1 [24]. *Salmonella* spp. were isolated by inoculating 100 µL of the samples on *Salmonella-Shigella* agar (SSA) and allowing them to incubate for 18–24 h at 37°C. Uncolored colonies with black centers were considered *Salmonella* spp. Microscopic (gram staining) and biochemical tests (catalase, sugar fermentation, methyl red, indole, and Voges-Proskauer) were performed on presumptive colonies for confirmation.

Clostridia are sulfite-reducing spore-forming bacteria that ferment lactose for gas production. Their culture is based on growth on a selective medium equipped with Na, Fe, and Al, which is carried out on Mannitol Salt Agar (MSA, LiofilChem, Italy). Clostridia reduce Na_2SO_3 to sulfides under anaerobic conditions to produce energy. Colonies that are positive for sulfite reduction are more correctly referred to as colony-forming units (CFUs) of "sulfide-reducing bacteria". This test is used as an indicator of fecal or soil contamination in dairy plants given that species from the genus *Clostridium* are isolated both from the soil and the feces of warm-blooded mammals [25].

After the different incubation periods, well-individualized colonies on Petri dishes were counted via a colony counter, and the average results from triplicate measurements were calculated and expressed as the logarithm of CFUs per gram of sample (log CFU/g).

Statistical analysis

The results obtained were subjected to analysis of variance (ANOVA) with the Newman–Keuls multiple comparison test to evaluate the statistical significance of the data expressed as the means ± standard deviations. A probability value at the 95% confidence limit ($P < 0.05$) was considered statistically significant via GraphPad Prism 5 software.

Results

Physicochemical properties

The values of the titratable acidity of the appertized tomato samples are presented in Figure 2. In general, the addition of lemon juice increased the titratable acidity of the appertized tomato, where the sample supplemented with 10 g of lemon juice (AT10) presented a high value (0.156) compared with the other samples ($P < 0.05$).

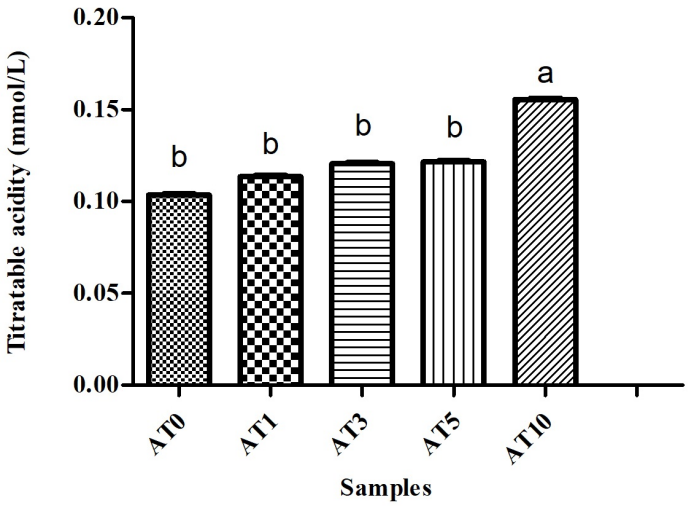

Figure 2. Changes in the titratable acidity of appertized tomato samples. The results are presented as the means ± standard errors ($n = 3$), and columns with different letters differ significantly ($P < 0.05$). AT0: appertized tomato without lemon juice; AT1: appertized tomato + 1 g of lemon juice; AT3: appertized tomato + 3 g of lemon juice; AT5: appertized tomato + 5 g of lemon juice; and AT10: appertized tomato + 10 g of lemon juice

For the change in pH, there was a significant difference ($P < 0.05$) for all samples, where the values varied from 3.52 to 4.29 for AT10 and AT0, respectively (Figure 3). Each concentration of lemon juice increases the acidity of the medium, resulting in a decrease in pH.

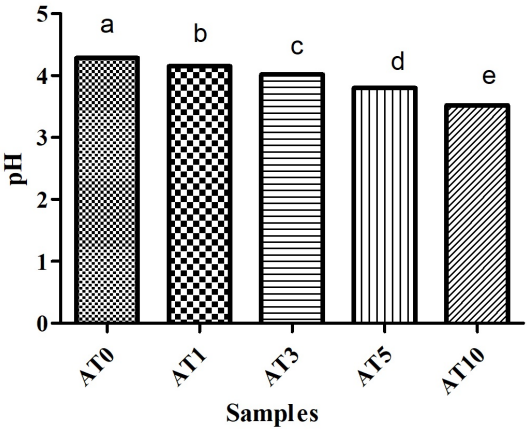

Figure 3. Changes in the pH values of appertized tomato samples. The results are presented as the means ± standard errors ($n = 3$), and columns with different letters differ significantly ($P < 0.05$). AT0: appertized tomato without lemon juice; AT1: appertized tomato + 1 g of lemon juice; AT3: appertized tomato + 3 g of lemon juice; AT5: appertized tomato + 5 g of lemon juice; and AT10: appertized tomato + 10 g of lemon juice. pH: hydrogen potential

Phytochemical contents

The TPC of the different samples is shown in Figure 4. There was a significant decrease ($P < 0.05$) in this parameter in the appertized tomato samples supplemented with lemon juice compared with the raw sample (AT0), which presented the highest value (0.10 mg GAE/g) compared to other samples where no significant difference ($P > 0.05$) was recorded between them.

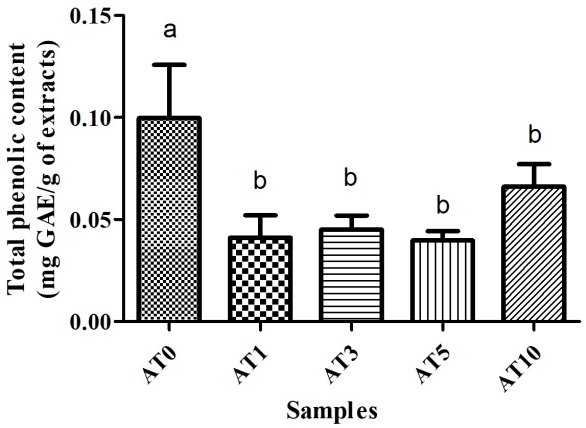

Figure 4. Changes in the total phenolic content of appertized tomato samples. The results are presented as the means ± standard errors (n = 3), and columns with different letters differ significantly (P < 0.05). AT0: appertized tomato without lemon juice; AT1: appertized tomato + 1 g of lemon juice; AT3: appertized tomato + 3 g of lemon juice; AT5: appertized tomato + 5 g of lemon juice; and AT10: appertized tomato + 10 g of lemon juice. GAE: gallic acid equivalent

The change in the total flavonoid content of the appertized tomato samples is presented in Figure 5. The highest values were obtained with the appertized tomato sample supplemented with 10 g of lemon juice (0.01 mg CE/g) and the appertized tomato sample supplemented with 3 g of lemon juice (0.009 mg CE/g). The values recorded for these samples were significantly different (P < 0.05) from those of the other samples, where the samples supplemented with 1 g of lemon juice presented the lowest total flavonoid content (0.001 mg CE/g).

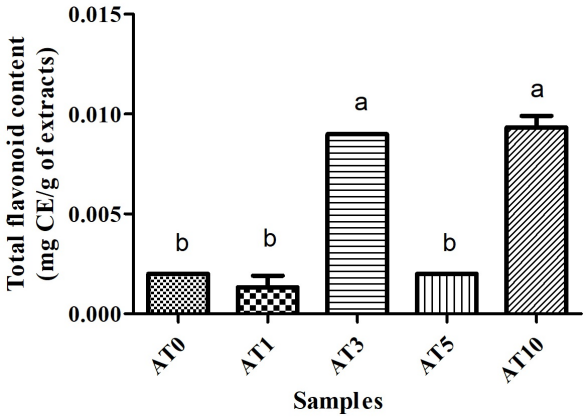

Figure 5. Changes in the total flavonoid content of appertized tomato samples. The results are presented as the means ± standard errors (n = 3), and columns with different letters differ significantly (P < 0.05). AT0: appertized tomato without lemon juice; AT1: appertized tomato + 1 g of lemon juice; AT3: appertized tomato + 3 g of lemon juice; AT5: appertized tomato + 5 g of lemon juice; and AT10: appertized tomato + 10 g of lemon juice. CE: catechin equivalent

The total carotenoid content significantly differed (P < 0.05) between the different appertized tomato samples (Figure 6). The highest value was recorded for AT10 (16.52 mg/100 g), followed by AT1 (16.14 mg/100 g), and sample AT3 presented the lowest value (15.22 mg/100 g).

Proximate composition

The changes in the proximate compositions of different appertized tomato samples supplemented with or without lemon juice are presented in Table 1. There was no significant difference (P > 0.05) in moisture content among all the samples, with values varying from 58.49 to 59.35% for AT5 and AT10, respectively. In terms of protein content, supplementation with lemon juice significantly decreased (P < 0.05) this parameter, where the AT0 sample presented the highest value (15.79%), whereas the lowest values were recorded for the AT3 (8.26%) and AT10 (8.56%) samples. The determination of lipid content revealed that supplementation with lemon juice significantly increased (P < 0.05) the number of lipids, with values varying from 1.2 to 2.07% for AT0 and AT5, respectively. However, the results also revealed that a very

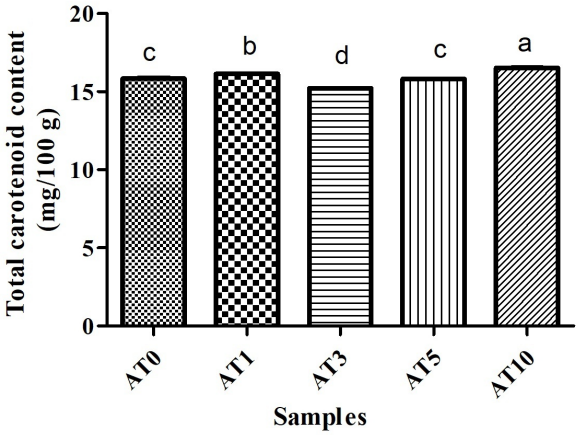

Figure 6. Changes in the total carotenoid content of appertized tomato samples. The results are presented as the means ± standard errors (n = 3), and columns with different letters differ significantly (P < 0.05). AT0: appertized tomato without lemon juice; AT1: appertized tomato + 1 g of lemon juice; AT3: appertized tomato + 3 g of lemon juice; AT5: appertized tomato + 5 g of lemon juice; and AT10: appertized tomato + 10 g of lemon juice

high amount of lemon juice significantly decreased (P < 0.05) this parameter compared with that of the other samples, with a lipid content of 0.60% obtained with AT10. In terms of ash content, there was no significant difference (P > 0.05) between samples AT0 and AT5, whose ash content was 7.5%, which was significantly greater (P < 0.05) than the values obtained for the other samples, which varied from 6 to 6.5% for AT1, AT3, and AT10, respectively. The amount of dietary fiber varied from 6.41 to 9.75% for samples AT0 and AT5, respectively. However, there was no significant difference (P > 0.05) in this parameter between samples AT0 and AT10 (6.41% and 6.74%), indicating that supplementation with high amounts of lemon juice tends to decrease (P < 0.05) the level of dietary fiber in appertized tomato samples. Concerning the carbohydrate content, except for samples AT1 and AT3 (15.31% and 16.06%), which did not significantly differ (P > 0.05), there was a significant difference (P < 0.05) between the other samples, with the highest value recorded with AT10 (18.25%) and the lowest value obtained with AT5 (9.85%).

Table 1. Changes in the proximate compositions of appertized tomato samples

Parameters	Moisture content (%)	Protein content (%)	Lipid content (%)	Ash content (%)	Fiber content (%)	Carbohydrate content (%)
AT0	58.88 ± 0.29^{a}	15.79 ± 0.04^{a}	1.2 ± 0.01^{c}	7.5 ± 0.70^{a}	6.41 ± 0.04^{c}	10.21 ± 1.13^{c}
AT1	59.05 ± 0.30^{a}	10.75 ± 0.03^{c}	1.2 ± 0.01^{c}	6.00 ± 0.00^{b}	7.68 ± 0.03^{b}	15.31 ± 0.84^{b}
AT3	58.99 ± 0.12^{a}	8.26 ± 0.01^{d}	1.61 ± 0.01^{b}	6.5 ± 0.00^{b}	8.57 ± 0.04^{a}	16.06 ± 1.25^{b}
AT5	58.49 ± 0.32^{a}	12.35 ± 0.01^{b}	2.07 ± 0.00^{a}	7.5 ± 0.70^{a}	9.75 ± 0.04^{a}	9.85 ± 1.55^{d}
AT10	59.35 ± 0.28^{a}	8.56 ± 0.02^{d}	0.60 ± 0.01^{d}	6.5 ± 0.00^{b}	6.74 ± 0.03^{c}	18.25 ± 1.65^{a}

The results are presented as the means ± standard errors (n = 3), and the values in the same column with different letters differ significantly (P < 0.05). AT0: appertized tomato without lemon juice; AT1: appertized tomato + 1 g of lemon juice; AT3: appertized tomato + 3 g of lemon juice; AT5: appertized tomato + 5 g of lemon juice; and AT10: appertized tomato + 10 g of lemon juice

Mineral composition

The changes in the mineral contents of the samples are presented in Table 2. For the Fe content, there was a significant difference (P < 0.05) between samples, where values ranged from 4.8 to 31.09 mg/100 g for AT3 and AT5, respectively. The supplementation of lemon juice significantly increased (P < 0.05) the Ca content of appertized tomato samples; the lowest value was observed with AT0 (112.25 mg/100 g), whereas the highest value was recorded with AT1 (704.5 mg/100 g). The same trend was observed for the P content (P < 0.05), where the values ranged from 151.17 to 389.37 mg/100 g for AT0 and AT5, respectively. In addition, lemon juice tends to significantly increase (P < 0.05) the Mg content of supplemented samples; here, the lowest value was recorded with AT0 (19.49 mg/100 g), and the highest value was recorded with AT3 (233.33 mg/100 g). The K content significantly differed (P < 0.05) among the samples, with values ranging from 2,189.36 to 4,304.34 mg/100 g for AT5 and AT1, respectively. However, the values recorded

for AT0 (2,345.41 mg/100 g) and AT10 (2,425.43 mg/100 g) were not significantly different ($P > 0.05$) from those recorded for AT5. The addition of a high amount of lemon juice significantly decreased ($P < 0.05$) the Na content of the appertized tomato, and the AT10 sample presented the lowest value (217.78 mg/100 g). However, there was no significant difference ($P > 0.05$) among the other samples, with values varying from 242.42 to 295.5 mg/100 g for AT0 and AT1, respectively.

Table 2. Changes in the mineral content of the appertized tomato samples

Parameters	Fe (mg/100 g)	Ca (mg/100 g)	P (mg/100 g)	Mg (mg/100 g)	K (mg/100 g)	Na (mg/100 g)	Ca/P	Na/K
AT0	6.17 ± 0.00^{c}	112.25 ± 0.35^{d}	151.17 ± 0.56^{e}	19.49 ± 0.14^{d}	2,345.41 ± 0.70^{c}	242.42 ± 0.70^{a}	0.74	0.10
AT1	5.42 ± 0.00^{d}	704.5 ± 0.75^{a}	309.19 ± 0.70^{b}	165.29 ± 0.72^{b}	4,304.34 ± 0.15^{a}	295.5 ± 0.75^{a}	2.28	0.07
AT3	4.8 ± 0.10^{e}	512.5 ± 0.70^{b}	289.75 ± 0.77^{c}	233.33 ± 0.70^{a}	3,205.28 ± 0.70^{b}	268.33 ± 0.70^{a}	1.77	0.08
AT5	31.09 ± 0.50^{a}	288.5 ± 0.76^{c}	389.37 ± 0.70^{a}	106.97 ± 0.71^{c}	2,189.36 ± 0.71^{c}	268.33 ± 0.80^{a}	0.74	0.12
AT10	14.44 ± 0.70^{b}	560.5 ± 0.70^{b}	192.57 ± 0.78^{d}	155.57 ± 0.70^{b}	2,425.43 ± 0.70^{c}	217.78 ± 0.70^{b}	2.91	0.09

The results are presented as the means ± standard errors ($n = 3$), and the values in the same column with different letters differ significantly ($P < 0.05$). AT0: appertized tomato without lemon juice; AT1: appertized tomato + 1 g of lemon juice; AT3: appertized tomato + 3 g of lemon juice; AT5: appertized tomato + 5 g of lemon juice; and AT10: appertized tomato + 10 g of lemon juice

Microbial analysis

The evaluation of the microbiological quality of the canned tomato samples is shown in Table 3. For the enumeration of yeasts and molds, the addition of lemon juice led to the inhibition of these pathogens in the different samples. Indeed, the number of yeasts and molds decreased significantly with increasing lemon concentration until zero values (not determined) were reached from the addition of 3% lemon juice (AT3). Compared with the sample not supplemented with lemon (AT0), which presented a value of 476.57 log CFU/g, the presence of yeasts and molds (43.19 log CFU/g) was recorded for AT1. Concerning the detection of *E. coli*, *Salmonella* spp, and *Clostridium botulinum*, no signs of infestation of appertized tomato samples by these pathogens were recorded during this study.

Table 3. Microbial analysis of the appertized tomato samples

Samples	Yeast and molds (log CFU/g)	*E. coli*	*Salmonella* spp.	*Clostridium botulinum*
AT0*	476.57 ± 2.89	nd	nd	nd
AT1*	43.19 ± 2.90	nd	nd	nd
AT3*	nd	nd	nd	nd
AT5*	nd	nd	nd	nd
AT10*	nd	nd	nd	nd

*: Results were expressed in triplicate; nd: not determined. AT0: appertized tomato without lemon juice; AT1: appertized tomato + 1 g of lemon juice; AT3: appertized tomato + 3 g of lemon juice; AT5: appertized tomato + 5 g of lemon juice; and AT10: appertized tomato + 10 g of lemon juice. CFU: colony-forming unit; *E. coli*: *Escherichia coli*

Discussion

In terms of physicochemical properties, the addition of lemon juice significantly increased the titratable acidity and reduced the pH of the appertized tomato samples. These changes could be due to the large amount of organic acid present in the lemon juice. According to Canene-Adams et al. [26], folic acid, malic acid, citric acid, and bioflavonoids (including lycopene, α-, and β-carotene) are the most common compounds found in tomatoes (*Lycopersicon esculentum*, Mill.). In addition, in canning, the addition of lemon juice significantly alters the pH of food (usually below 4.6), impacting flavor, texture, color, and preservation in the cooking of canned fruits and vegetables. Lemon juice can kickstart fermentation by creating an acidic environment that acts as a hurdle to the growth of spoilage and pathogenic microbes, to the detriment of beneficial bacteria [27]. Additionally, Novelina et al. [28] revealed that the increase in total acid content is affected by the addition of lemon juice in the preparation of tomato jelly drinks. According to

these authors, the main organic acids contained in fruit juice are citric acid and malic acid. The element that causes a sour taste is the H^+, which can be derived from an organic acid molecule that is found mainly in the lemon, and an ionized molecule releases its proton H^+. A greater presence of H^+ results in a more acidic solution and a lower pH. These results are following those of the Codex Stan [29], which revealed that the pH of appertized tomatoes should be less than 4.5. According to Pratt et al. [30], the results obtained for the titratable acidity and pH could preserve the stability of the sample and thus make it unattractive against certain microorganisms, such as *E. coli* and *Clostridium botulinum*.

Phenolics are the major antioxidant compounds in plant extracts and might contribute 60% to 70% of the antioxidant activity of extracts [31]. However, the TPC was significantly lower ($P < 0.05$) in the appertized tomato samples supplemented with lemon juice than in the raw samples. The decrease in TPC observed during this study may be due to the breakdown of the cellular structure that caused their lysis with heating time and processing steps of different tomato products. Berinyuy et al. [32] reported a decrease in TPC after the boiling of tomato samples at 98°C from the 20th to the 30th minute, and they attributed these changes to the decomposition of antioxidants, which have limited thermal stability under certain conditions. In addition, Ca has been shown to have positive effects on the accumulation of carotenoids, vitamin C, and phenolic acids in fruits and vegetables [33]. Based on the results of the proximate composition recorded in this study, the supplemented samples presented higher Ca contents than the raw samples. Moreover, the values obtained with TPC by our experiments also presented higher values for fresh tomatoes than those reported in the literature, ranging from 0.6 to 0.9 mg GAE/g fresh matter [34] and 0.14 mg GAE/g fresh matter [35].

Numerous edible fruits and vegetables contain flavonoids, one of the most common classes of secondary plant metabolites [36]. In addition, owing to their ability to lower blood cholesterol and prevent obesity, diabetes, cardiovascular disease, and some types of cancer, lemon fruits are high in flavonoids and are an essential component of a balanced diet [37]. The highest flavonoid content was obtained with the hydrated tomato sample supplemented with 10 g of lemon juice. The flavonoid content was determined based on the addition of lemon juice to the appertized tomato samples. The beneficial effects of dietary citrus fruits can be attributed not only to vitamin C, minerals, dietary fiber, organic acids, and carotenoids but also to the antioxidant activity of their flavonoids, and this property is linked particularly to the chemical structures of these flavonoids [38]. According to Garg et al. [39], three structural groups are important for evaluating the antioxidant capacity of flavonoids, namely, the ortho-dihydroxy (catechol) structure in the B-ring; the 2,3-double bond in conjugation with a 4-oxo functional group; and the presence of both 3-(*a*)- and 5-(*b*)-hydroxyl groups. Indeed, vitamin C and flavonoids are, in part, synergistic for the biological activities of citrus fruits. However, the flavonoid contents recorded in this study are lower than those reported by Riahi and Hdider [40] for Tunisian tomato samples, which range from 0.109 to 0.113 mg CE/g (fresh matter). Moreover, Boumendjel et al. [8] reported an increase in the flavonoid content of tomato slices subjected to microwave heating for 30 s (2.57 mg CE/g fresh matter) and 300 s (10.68 mg CE/g fresh matter) compared with that of the control group (2.33 mg CE/g fresh matter), and they concluded that heat treatment helped release these antioxidant components from the cell matrix of fruits.

Tomato (*Solanum lycopersicum* L.) is consumed fresh and processed for its nutritional and bioactive antioxidants, such as carotenoids [41]. According to Phung et al. [42], lycopene is the predominant carotenoid in tomatoes, and it has the highest antioxidant activity and singlet oxygen-quenching ability of all dietary carotenoids. As observed previously for the flavonoid content, the highest value for the carotenoid content was also obtained with the appertized tomato sample supplemented with 10 g of lemon juice, whereas the lowest value was obtained with the sample supplemented with 3 g of lemon juice. In this study, the carotenoid content recorded was dependent on the concentration of lemon juice added to the hydrated tomato samples. In fact, lemon is a rich source of carotenoids, and these constituents protect against photooxidative damage. However, the concentration of carotenoids is strongly dependent on the citrus variety and growing conditions [43]. According to Melendez-Martínez et al. [44], lemon contains reasonable quantities of carotenoids for daily nutrition. In addition, it is well known that processing

tomatoes (via heat and mechanical treatment) releases lycopene from the food matrix and results in better bioavailability of lycopene [45]. Authors revealed that microwave treatment for 5 min released the maximum phenolic and carotenoid concentrations in tomato slices [8]. Jacob et al. [46] reported that heat treatment resulted in a concentration of lycopene of 9.89 mg/100 g fresh matter and 9.83 mg/100 g fresh matter in tomato samples paste at 110°C for 15 min (RT1) and at 110°C for 30 min (RT2) compared with raw tomato samples (1.98 mg lycopene/100 g). The same observations were recorded, with the total amount of β-carotene varying from 0.25 mg/100 g fresh matter for the raw sample to 0.70 mg/100 g fresh matter and 0.91 mg/100 g fresh matter for RT1 and RT2, respectively [46]. However, the number of carotenoids obtained in this study was greater than that reported by these authors, which could be explained by the number of carotenoids present in lemon juice added to the appertized tomato samples.

The quality and nutritional value of fresh products such as tomatoes are affected by postharvest handling and storage conditions. Therefore, good and protective storage methods are needed to increase the shelf-life and physical quality of these products [47]. Concerning the proximate composition of the tomato samples, supplementation with lemon juice significantly reduced ($P < 0.05$) the protein content compared with that of the unsupplemented samples. According to Klimek-Szczykutowicz et al. [27], the acidic nature of lemon juice can denature proteins and affect enzyme activity, leading to changes in food structure and function. The protein contents obtained in this study are greater than those reported by Abdullahi et al. [48] for commercialized canned tomatoes from the Tarauni market in Kano State, Nigeria, where values ranged from 4.16–4.83%. However, these values are lower than those reported for dried tomato waste (17.62%) by Nour et al. [49]. The lipid content of the tomato samples found in this study varied from 0.6 to 2.07%. These values are lower than those reported by González et al. [50] and Elbadrawy and Sello [51] for dried-peeled tomatoes, who obtained crude fat contents of 0.6 and 4.04%, respectively. However, these values are significantly greater than those obtained by Abdullahi et al. [48] for commercialized canned tomatoes in Kano State, Nigeria, where values ranged from 0.14 to 0.28%. The lowest value of lipid content recorded with AT10 could be explained by the way that high acid content can decrease the polyunsaturated fatty acid content of tomatoes, as demonstrated by Wickramanayake et al. [52]. The ash content found in our samples (6% to 7.5%) is greater than that reported in previous studies. Nour et al. [49] reported an ash content of 4.21% in dried tomato waste (skins and seeds). The highest ash content in tomato may be a result of its ability to absorb minerals from the soil [51]. The values of fiber content recorded in this study (6.41–9.75%) are higher than those obtained by Abdullahi et al. [48] for commercialized canned tomato samples, with values varying from 4.97–6.16%. According to [49], a high fiber content is metabolized slowly for energy contribution and digested slowly, which makes it a great supplement for processed fiber-rich foods. Carbohydrates are essential nutrients in the body, as they constitute the major energy source. The carbohydrate contents recorded in this study (9.85–18.25%) are similar to those reported by Abdullahi et al. [48] for commercialized canned tomato samples (13.7–15.18%). According to Agbemafe et al. [53], the differences in the nutrient contents of tomato samples can be attributed to various influences, environmental conditions, and other agronomical practices during production. Based on sensory analysis, a study revealed that natural juice formulated with 5% lemon juice was the most organoleptically accepted. On the other hand, a formulated natural drink supplemented with 10% of lemon juice + sugar was shown to be the most nutritious, and recorded the least overall acceptability amongst the five formulations [54].

In terms of mineral composition, among the microelements, the high concentration of lemon juice increased the Fe content of the canned tomato samples. According to González-Molina et al. [43], Fe is an essential part of hemoglobin, and lemon contains trace elements of Fe (0.6 mg/100 g). The range of obtained values for Fe in the canned tomato samples ranged from 4.8 to 31.09 mg/100 g, which is higher than those recorded with commercialized canned tomato samples (1.08 to 3.44 mg/100 g) [48]. Among the macroelements, supplementation with lemon juice significantly increased the Ca, P, and Mg contents of appertized tomato samples compared with those of unsupplemented samples. This increase could be due to the amount of these minerals present in the added lemon juice. In contrast, only a high concentration of

lemon juice (AT10) reduced the Na content of appertized tomato samples. According to [43], K is the main mineral present in lemon, although other minerals, such as Ca, Mg, and P, are also present at minor levels, whereas Na, Fe, and Zn are present at trace levels. K is an essential mineral for human health since it is essential for maintaining the water-acid balance and participates in the transmission of nerve impulses to muscle [55]. Ca is the main constituent of bones and teeth and has key metabolic functions; Mg is essential in muscle contraction; P is a component of DNA materials and is involved in energy distribution; and Na is necessary for maintaining the balance of the physical fluid system, which is also required for nerve and muscle function [40]. These results are in agreement with those reported by Zharkova et al. [56], who reported that some varieties of tomatoes grown in the village of Khokhol of the Russian Federation have high mineral saturation and are rich in macro- and micronutrients (K, Ca, Mg, P, Mn, and Fe). The values recorded in this study are higher than those obtained by Abdullahi et al. [48] for Na (12.7 to 16.3 mg/100 g), Mg (6.6 to 13.2 mg/100 g), K (7.18 to 8.90 mg/100 g) and Fe (1.08 to 2.7 mg/100 g). For supplemented samples, the Ca, K contents obtained in this study ranged (200–1,300 mg/100 g for Ca, and 300–4,700 mg/100 g for K) of the FAO/WHO standard for covering the mineral requirements of weaning-age children [57]. In addition, for better Ca absorption, the Ca/P ratio must be between 0.5 and 1 [57]. Similarly, according to Perez and Chang [58], the ratio of Na/K in food should be less than or equal to 1, and it is a good indicator for the prevention of cardiovascular diseases.

The assessment of the microbiological quality of the canned tomato samples revealed that the number of yeasts and molds decreased significantly with increasing lemon juice concentration. Several components of lemon are responsible for different targets in infectious entities, and lemon essential oils can be used as antimicrobial and antifungal agents [59]. In addition, the antimicrobial effect of essential oils may be considered with care, although essential oils may constitute a good alternative to combat the increasing resistance of pathogens [60]. According to Klimek-Szczykutowicz et al. [27], most microorganisms that cause food spoilage and foodborne illnesses thrive in a neutral to slightly acidic environment (pH 4.6 to 7.0); thus, by lowering the pH of canned foods to 4.6 or lower, the growth of these harmful microorganisms is inhibited, significantly reducing the risk of spoilage and foodborne diseases.

Tomato sample treated with 5% of lemon juice extract was more effective in preserving chemical parameters of the appertized tomato samples. Meanwhile, supplemented tomato samples with 3%, 5% and 10% of lemon juice are recommended for preserving tomatoes against higher microbial loads and ensuring the health of consumers.

Conclusions

The supplementation of lemon juice influenced the physicochemical properties of the appertized tomato samples by increasing their titratable acidity and reducing their pH content. Phytochemical analysis revealed that the addition of lemon juice increased the flavonoid and carotenoid contents while reducing the TPC of the appertized tomato. Additionally, supplementation influences nutritional value by increasing the level of essential minerals (Ca, P, and Mg) in appertized tomato samples. Lemon juice inhibits the growth of yeasts and molds and can be useful for increasing the shelf-life of appertized tomato samples. So, lemon juice can be used as a natural conservator to preserve the quality of appertized tomatoes.

Abbreviations

CE: catechin equivalent

CFUs: colony-forming units

E. coli: *Escherichia coli*

GAE: gallic acid equivalent

H^+: hydrogen ion

pH: hydrogen potential

TPC: total phenolic content

Declarations

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Author contributions

SCHN: Conceptualization, Methodology, Formal analysis, Writing—original draft. FB: Investigation, Methodology, Formal analysis. BZZ: Methodology, Formal analysis, Writing—original draft. EEE: Supervision, Formal analysis, Writing—original draft. RT: Methodology, Writing—original draft. HMW: Supervision, Validation, Writing—review & editing. All authors read and approved the submitted version.

Conflicts of interest

The authors declare that there are no conflicts of interest.

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Consent to participate

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