Food ScienTech Journal Vol. 6 (1) 2024 ISSN : 2685-4279 DOI:10.33512/fsj.v6i1.23489

Potato Peels Waste Extract as Natural Antioxidant and Antimicrobial in Lemon Carbonated Soft Drink

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Submitted: 18.12.2024; Revised: 15.05.2024; Accepted: 09.06.2024

ABSTRACT

Most soft drinks are generally considered ready media for spoilage microbial growth. Deterioration of these products by undesirable microorganisms can alter the sensory quality of the product which also poses a major public health risk. The soft drink category with lemon made up 85% of all bacterial contamination and was considered microbiologically unsafe due to a lack of preservatives. Over the last decade the world has been generating a high quantity of potato peel waste. These peels have several economic benefits but there is mismanagement or inappropriate valorization that could present risks to environment and public health. Potato peels waste contain several bioactive components. These components are known to provide human health benefits including antioxidant and antimicrobial properties. The use of potato peels as natural antimicrobial compounds are believed to play important role in reduce or inhibit microbial growth in food stuff. Results confirmed that the antimicrobial action of potato peel methanol extract (PPME) at 200 ppm concentration, was higher and achievable than using weak-acid preservatives (citric acid), where total bacterial count, aciduric bacteria, yeast and mold viable counts of standard lemon product (without PPME) showed tolerance to low pH and was associate to spoilage the final product even using citric acid. Otherwise, both standard and PPME lemon soft drink formulations were free of pathogenic bacteria (Coliform & *E. coli*) after production and during 6 months storage period. All quality sensory attributes gradually decreased ($P \leq 0.05$) up to the sixth month of storage. After production, taste, appearance and overall acceptability of PPME- lemon soft drink formulation had lower ($P \leq 0.05$) values than standard lemon soft drink formulation (control). In contrast, PPME-lemon soft drink formulation had higher ($P \leq 0.05$) quality sensory values than standard lemon soft drink formulation (control) as affected by PPME addition.

Keywords: Antimicrobial, antioxidant, soft drinks, natural preservatives, potato peel waste

INTRODUCTION

The manufacturing of carbonated soft drinks is a very large industry. Mexico was the country with the highest carbonated soft drink consumption, namely over 630-ounces servings per capita per year. The United

States stood in second place, with almost the same quantity, while Brazil, which ranked third, consumed less than half the soft drinks Mexicans drank that year. Also, approximately 5.3 billion liters of carbonates were consumed in the UK. Total volume of carbonated soft drinks consumed in European Union per capita was 243.6 liter (Helal, 2020). Soft drinks are susceptible to microbial spoilage because their high carbon-to-nitrogen (C/N) ratio and low pH (3.5) allows the growth of acetic and lactic acid bacteria, molds, and yeasts (Garavaglia et al., 2019). Non-alcoholic beverages are highly prone to microbial contamination. While high-level microbial contamination can cause economic loss through product spoilage and consumer rejection, lower and usually inconspicuous levels may, if uncontrolled, pose grave human health problems (Oranusi, 1994). Antioxidant supplemented carbonated beverages faced a lot of challenges, increase the need for additional antioxidants in the diet or require antioxidant supplements is not clear (Urso and Clarkson, 2003). A diversity of factors, such as methodological differences, the antioxidant tested or the nutritional status of the subjects participating in the test (Levine et al. 1990, Sacheck and Blumberg 2001, Morillas-Ruiz et al. 2005). Further studies are necessary to confirm the real beneficial effects of antioxidant supplementation. Under different defections, researchers create recipes of soft drinks and beverages containing different antioxidants. Such as, antioxidants blended- beverages (Awe et al. 2013). Potato peel waste containing phenols and flavonoids give it advantages in high antioxidant and antimicrobial activities. Therefore, this study was designed to enhance a high category of soft drink most expose to microbial contamination with antioxidants and antimicrobial components extracted from potato peels waste, and then evaluate their microbiological and sensory properties comparing with lemon soft drink standard formulation.

MATERIALS AND METHODS

Plant Material

Potato peels processing (20 Kg) waste (Solanum tuberosum L.) was collected from peeling carborundum machine unit, chips production factory (6th of October city, Giza Governorate, Egypt) during the winter season of 2017. The collected peels were hand-sorted to remove foreign particles, filtered to remove accumulated water generated from collection zone, then stored in polyethylene bags in the freezer (Ideal, Delta Home Appliance, Egypt) at -18°C ± 1 until used.

Collection of lemon soft drink standard formulation

Lemon soft drink standard formulation packaged in 1.0-liter polyethylene terephthalate (PET) bottle were collected after production during winter season (2016), from 6th of October city, Giza Governorate, Egypt, then stored at laboratory temperature (22°C±2).

Preparation of plant material

Frozen potato peels processing waste was dried in a ventilated oven (memmert, GmbH+CoKG, UF 55, Germany), at 40°C±1 until dryness. The sample was ground by using a laboratory disc mill (Perten, Model LM 3100, Sweden), sieved in 25 mm sieve and kept in a tightly closed PET bottle at -18°C ± 1 in freezer for further analysis.

Extraction of antioxidant and antimicrobials:

Dried ground potato peels were extracted under optimum extraction conditions, with 80% methanol for overnight (16 hr) at room temperature (22ºC ± 2). The samples were sonicated for 20 min in ultra-sonication device (Elmasonic 15 Hans Schmidbauer GmbH), and then filtrated through Whatman No.1 filter paper. The precipitates were re-extracted under the same conditions twice

and the filtrates were combined. Evaporation of methanol performed via a rotary evaporator (Buchi 011, Buchi, Switzerland) below (40ºC). The residues were stored in dark tight glass at -18°C±1 until further use.

Evaluation of potato peel methanol extract antioxidant activity

Potato peel extracts were investigated for the scavenging effect on 1, 1-diphenyl-2-picrylhydrazyl (DPPH) radical according to the method of Brand-Williams et al. (1995).

Preparation of lemon soft drink mixed with potato peels methanol extract:

Table 1 lists the specifications of lemon soft drink standard formulation used in the study. The dosage of potato peel methanol extract (200 ppm) in the lemon soft drink formulation were defined in early published paper with indicator microorganisms (Helal et al., 2020). The amount of 200 ppm of potato peels methanol extract (PPME) was mixed under aseptic conditions with 1000 ml of lemon soft drink standard formulation in PET filling unit and then closed immediately, then stored at laboratory temperature (22°C±2). All microbiological and sensory properties were analysed after production immediately, during 6 months storage period, compared with the control standard formulation of lemon soft drink under controlled temperature conditions.

Physicochemical Methods

The degassing of lemon soft drinks (Standard formulation & PPME-formulation) were accomplished according to the method described by Victoria Bedenko (2010) using Commercial Somex Degassing Unit Somex Soft drink Degasser (Bally vourney CO. Cork, Ireland). The pH was measured using a pH meter (Jenway 3510 pH Meter, England) as described by Rangana (1977), according to manufacture manual, the Anton Paar Carbo Qc (DMA 48 / DMA 58, Austira) measuring system for monitoring and measuring CO2 & O2 was used, (Anton Paar Manual, 2010). Density was measured as described by (Steinbach et al., 2014). The titratable acidity, citric acid (ppm), sugars (ºBrix, refractometer, ATAGO Model 5000 DCX, Research Analytical, Japan) measurements were performed in triplicate following AOAC official methods.

Microbiological Methods

A membrane filter procedure for enumerating total bacterial, yeast, molds and aciduric bacteria, total coliform and Escherichia coli counts was developed and evaluated with some modifications as follows:

Appropriate volumes (100 ml) of standard and PPME lemon soft drinks formulations samples were passed through 0.45µm gridded membrane filters (MCE), by using vacuum funnel assembly. Then, samples were allowed to drawn completely via vacuum pump through the filter and the filters then were placed on the selected medium, incubated at the proper temperature and for the appropriate time period, then counted to confirm the colonies.

Total bacterial count

Total bacterial viable count was carried out according to Anon (2002). Standard and PPME lemon soft drinks formulations samples (100 ml) were transferred by vacuum to each three separate Petri dishes contained plate count broth (total count and plate count broths and agar). The plates were then incubated in an incubator at 37°C for 48 hr. A colony counter was used to count the viable bacteria.

Yeast, mold and aciduric bacteria counts

Yeast, mold and aciduric bacteria viable count was carried out according to Anon (2002). Standard and PPME lemon soft drinks formulations (100 ml) were transferred by vacuum to each three separate Petri dishes contained M-Green Yeast and

Mold Broth. The plates were then incubated in an incubator at 25°C for 120 hr. A colony counter was used to count.

Total coliform bacteria detection

A total coliform bacterium was detected according to AOAC (2005). (COLIFORM METHOD: PRESENCE/ ABSENCE). Standard and PPME lemon soft drinks formulations (100 ml) were transferred by vacuum to each three separate Petri dishes, Colilert reagent was added. The plates were then incubated in an incubator at 35°C for 24 and 48 hr. A change in color form the initial colorless to yellow indicates the presence of total coliform bacteria.

***Escherichia coli* detection**

Escherichia coli (E. coli) detection was carried out according to Downes and Ito (2001). MI Broth and Agar was selected as medium for general use with membrane filtration technique to determine *E. coli* count. Standard and PPME lemon soft drinks formulations samples (100 ml) were transferred by vacuum to each three separate petri dishes, Colilert reagent was added. The plates were then incubated in an incubator at 35°C for 24 and 48 hr. Fluorescence of a bright light blue color when exposed to UV light indicates the presence of *E. coli.*

Sensory method

Standard and PPME lemon soft drinks formulations were subjected to sensory evaluation directly after production and every month during six months storage periods for color, appearance, taste and overall acceptability by trained panel consisted of ten members (average age mid-30s) selected from laboratory staff and team of sensory test, using Hedonic scale rating 1-9 points (1 = dislike very much, 9=like very much) to assess the differences (Abeker, 2009). Panelists evaluated of soft drink samples which were offered at the same time in specific area of sensory test in the soft drink samples plant quality assurance laboratory without special lighting. Water was provided for rinsing purposes.

Statistical analysis

Statistical analysis was conducted according to Snedecor and Cochran (1994). Sensory properties of standard and PPME lemon soft drinks formulations were determined as the mean of ten replicates, while physico-chemical properties of standard and PPME lemon soft drinks formulations were determined as the mean of three replications. Two-way (Factorial Design) analysis of variance was used for the physico-chemical and sensory properties of lemon soft drinks formulation. Least significant difference (LSD) was used for comparison among means, considering significance at 0.05% level, using Costat version 6.311 (Copyright 1998-2005, CoHort software).

RESULTS AND DISCUSSION

Properties of potato peel methanol extract

Based on our previous published data (Helal et al., 2020), potato peel antioxidants and antimicrobials extraction were optimized by using 80 % methanol, yield (10.42±0.00), total phenols (3.78±0.00 mg GAE g-1 DW), total flavonoids (0.13±0.00 mg Rutine g-1 DW) and antioxidants activity (80.86±1.21 %). Potato peel methanol extract at 400 ppm was more effective in inhibiting gram-positive and gram-negative bacteria and *C. albicans* as indicator microorganisms than ampicillin.

Physicochemical changes after production and during 6 months storage period

The CO_2, pH, Density, O_2, TSS, titratable acidity, and citric acid were evaluated as these parameters are essential for controlling the lemon soft drinks formulations (control and PPE) quality Table (1). All parameters were initially within acceptable quality limits

for tested lemon carbonated soft drinks formulations (control and PPE-lemon carbonated soft drinks formulations). After production, the standards lemon carbonated soft drink and PPE lemon carbonated soft drink formulation had carbonation volume 4.46±0.03 (v/v) and 4.32±0.03 (v/v), respectively. After 6 months of storage (Table 2), carbonation volume decreased gradually for both formulations 1.62±0.02 (v/v) and 2.15±0.08 (v/v). This may be due to the permeation mechanism through bottles walls (Ondulati, 2014). During the storage at room temperature, the pH of the tested standard and PPE lemon carbonated soft drinks formulations changed ($P \leq 0.05$) values recorded 3.25±0.05 and 3.20±0.06, respectively at the beginning of production. During the storage period (6 months) pH values were significantly ($P \leq 0.05$) decreased. Contrarily, titratable acidity increased significantly in both formulations, indicating that the addition of PPE (400 ppm) does not affect pH and titratable acidity of lemon carbonated soft drink formulation comparing with control formulation. This is may be due to chemical equilibrium modifications inside the PET bottles (Nyman et al., 2010). During the storage, precipitation and dissolving of salts (Dias et al., 2011) and even the incorporation of oxygen during bottling and storage (Arisseto et al., 2013) are typical. The increment of O2 content was practically observed during the whole storage period, due to the permeability of soft drinks packages resin. Following this finding (Profaizer and Ordulanti, 2007) found that O2 content raised from 0.3 to 6.5 ppm after 6 months of storage period. No literature was found regarding density values among storage period

Regarding citric acid levels (%) of standard and PPE- LCSD were 0.17±0.03% and 0.16 ±0.03% respectively (after production). Non-significant changes were detected among 6 months storage period. The results were in accordance with (Gravaglia, 2019, Brima and Abbas, 2014) citric acids concentrations in CSD were ranged between 0.17% and 0.18 %. There is no recommended levels of citric acids in soft drinks however, USA soft drinks contain varying quantities of 0.131- 0.350%, it i varying quantities of 0.131- 0.350%, it is generally considered safe by Food and Drug Administration (Terry-McElrath et al., 2014). Hence, the results of citric acid concentrations follow USA soft drinks standards that can be used for formulating health policy.

Sugar hydrolysis was significantly observed in standard LCSD. After production, TSS gave 11.25±0.03% and gradually decreased to 10.85 % by 6th month of the storage period. This is may be due to the transformation under the action of microbial spoilage and/ or chemical reactions. The decline of TSS values of tested soft drinks for 10 months storage was detected due to hydrolysis of sugars caused by the acidic pH of these drinks upon storage (Idris et al., 2016). Significant differences were not observed for sugar values when comparing the beginning and the ending of shelf life for PPE-LCSD formulation, no sugar hydrolysis or transformation under the action of microorganisms and/ or chemical reactions were detected, this may be due to the enhancement of PPE as antimicrobial constituents. Storage time exhibited a significant ($P \leq 0.05$) effect on the physical properties of the PPE-lemon carbonated soft drink formulation. However, the addition of 400 ppm of PPE to lemon soft drink formulation didn't influence physicochemical attributes.

Microbiological changes after production and during 6 months storage period

The total bacterial count, aciduric bacteria, yeast and mold, coliform, and E.coli counts were determined after production and during 6 months storage period of both standard and

PPE lemon carbonated soft drinks are presented in Table (3). Mainly after production, both standard and PPE lemon soft drinks formulations were considered unstable to microbial contamination of total count, molds and yeasts. However, pathogen's (Coliform & E.coli) detected zero counts. Also, after production, aciduric bacteria was zero in PPE-lemon soft drink formulation. (Oransui et al., 1994, Hoffman, 1997) found that bacterial counts of lemon soft drinks were 75 and 100 CFU/100 ml, respectively.

For the shelf life, the optimum dose of natural preservatives (400 ppm PPE). Table (4) shows the effect of PPE against bacteria, mold, and yeast during 120 days of storage compared with standard formulation (applied citric acid as weak-acid preservatives).

The total count of bacteria population in standard lemon soft drinks during shelf life was increased after production up to 3rd month of storage (160, 203, 256, 318 CFU/100ml, respectively). After four months of storage till the end of the shelf life, the active bacteria count declined in order 297, 254 and 209 CFU/100 ml, respectively. These results are higher than those reported by (Gravaglia, 2019). Who reported that the total bacterial count of lemon soft drink formulation after 4th months of storage was 5 CFU/100 ml. Saint Lucia Bureau of Standards (2004) specified that the total bacterial count should be less than 50 CFU/100 ml in carbonated soft drinks. The aciduric bacteria count of standard lemon soft drink were increased after one month of storage period up to the 3rd month of the storage period, still active by the end of 6 months (50 CFU/100 ml). The standard lemon soft drink formulation during the storage period at room temperature for 6 months were utterly free of coliform bacteria and E. coli. Using citric acid as week-acid preservatives, at 6 months shelf life, 98 & 4 log CFU/100 mL of yeast and mold were detected after 1st of storage, increased gradually till 2nd month of storage by 75 & 10 2 log CFU/100 ml. Our results are conformed with (Saint Lucia Bureau of Standards 2004). They revealed that coliform and E.coli counts must be less than 1.0 CFU/100 ml and 0.0 CFU/100 ml, respectively. The yeast and mold growth were preserved at 10 & 1 log CFU/100 mL, respectively, by the end of the storage period for all conditions tested (Table 3 and Fig 2). However, results indicated that preservatives in standard LCSD could not inhibit microbial spoilage to the 0.0 limits. demonstrated that yeast and mold counts were 60 and 34 CFU/100 ml of tested carbonated soft drinks (Oransui et al., 2014, Hiko and Muktar, 2020).

The microbial population in PPE-lemon soft drink during shelf life was reduced efficiently compared with standard lemon soft drink formulation showing that treatment with PPE had an antimicrobial effect. , The total bacterial count, was shown using citric acid (0.17 %) and PPE (400 ppm) with 5.60 CFU/100mL, followed by 3.30, 2.0, 1.60, 0.60, and 0.0 CFU/100mL, respectively by the end of storage. Aciduric bacteria, coliform and E. coil were absent during 120 days of PPE- lemon soft drink formulation storage. Using 400 ppm of PPE-lemon soft drink formulation, at 6 months shelf life, gradually reduced the development of yeast and mold growth by 3.30 & 3.60 log CFU/100 mL after 1st month of storage. The yeast and mold growth were preserved at 0.0 log CFU/100 mL after 6th & 4th months of storage, respectively. Present data are confirmed that the antimicrobial action of PPE (400 ppm) was higher and achievable than using weak-acid preservatives (citric acid) where yeast and mold viable counts of standard- LCSD show tolerance to low pH, and was associated to spoilage the final product even using citric acid. The results are

similar to the previous findings (Saint Lucia Bureau 2004), yeast and mold viable counts of carbonated soft drinks during 6 months should be less than 50 CFU/100 ml

Deterioration factor (DF) of standard and PPE-lemon carbonated soft drinks formulations:

Deterioration factor (DF) data of PPE-lemon carbonated soft drink and standard lemon drink (control) formulations are shown in Table (5). The results show that DF of lemon carbonated soft drink (control) was higher than PPE-lemon carbonated soft drink in all microbial growth parameters (total bacterial count & aciduric bacteria and yeast & mold). Pathogenic bacteria and coliform were absent in both formulations. However, when using PPE, the DF was lower than the formulation with weak-acid preservatives (citric acid), indicating that its use may decrease DF to minimal limits (0.0%), as aciduric bacteria growth compared with the control formulation (225.72 %).

When creating a new product, DF assessment is essential for anticipating the performance of the product. The PPE-LCSD gave the lowest values of DF compared with standard-LCSD. Except, DF of pathogenic bacteria was negative in both tested formulations, showing that the PPE-LCSD suppressed standard- LCSD.

4. Sensory changes after production and during 6 months storage period

Results of sensory evaluation of standard and PPE-lemon carbonated soft drinks formulations are shown in Table (6) After production, taste, appearance and overall acceptability of PPE-lemon soft drink formulation had lower ($P \leq 0.05$) values than standard lemon soft drink (control) as affected by PPE addition. This is may be due to astringency related to PPE phenolic content. Otherwise, the odor didn't show ($P \geq 0.05$) difference. After production, the addition of PPE seems to have no effect for deteriorating taste, odor, appearance, and overall acceptability.

Periodical analysis for sensory attributes manifested a mild decline ($P \leq 0.05$) during the 6-months storage period in standard and PPE-lemon soft drinks formulations. However, PPE-lemon soft drink formulation had higher ($P \leq 0.05$) than standard lemon soft drink formulation. Also, the PPE-lemon carbonated soft drink formulation was still liked by judges for taste and overall acceptability up to 3rd month of storage period. Odor and appearance were still liked up to the 4th and 5th months of storage, respectively. A uniform pattern of decline in these sensory attributes of the PPE-lemon carbonated soft drink formulation is evident concerning storage time albeit the samples were not rejected for taste and overall acceptability up to 3rd months of storage. Deterioration of lemon soft drink brands' sensory characteristics could be explained by the formation of undesired chemical and microbial compounds resulted from the high microbial load of this category hence, microbial spoilage inhibition in the mentioned beverages-category via addition of bioactive (antimicrobial) compounds could be necessary to ensure shelf-stable product, maintain sensory and quality attributes during storage period. In this content, lemon carbonated soft drinks sensory characteristics start to decline by increasing storage period (El-Faki, and Eisa, 2010). Additionally, lemon soft drink quality shelf life reduced by high microbial load even after 120 days of storage and using natural preservatives could enhance microbial stability of lemon soft drink against microbial growth (Gravaglia et al. 2019).

No studies have confirmed the effect of adding PPE as bioactive-antimicrobial constituents and subsequent storage at ambient temperature of the lemon soft drink samples on the sensory

characteristics. However, no data are reported in the literature, in particular PPE, which can be assumed to confer to the considered drink (lemon soft drink) to investigate sensory attributes after production and /or during storage time.

CONCLUSION

Potato peel extract (PPE) contains antimicrobial constituents and can be used to reduce and/or inhibit microbial spoilage of sensitive lemon soft drinks formulations and enhanced Physicochemical attributes. For example, at dose of 400 ppm of potato peel extract, it was possible to inhibit the growth aciduric bacteria, yeast and mold cultures, reducing total bacteria counts to safe limits even after 120 days, using the same dose, lemon soft drinks sugar levels remained stable. Besides the PPE-lemon carbonated soft drink produced (combined with citric acid) offered good quality with a composition that adhere legal and technological specifications. Therefore, it's recommended to extend studies on the application of potato peels as natural antioxidant and antimicrobial source in microbial contamination-prone carbonated soft drinks.

ACKNOWLEDGMENT

The author thanks Pepsico Egypt for supporting this study.

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Table 1. Physcio-chemical properties of standard and PPME-lemon soft drinks formulations used in the tests.

Physico-chemical characteristics	Standard formulation	PPME-formulation
CO_2 (V/V)	4.46^{a}±0.02	4.32^{b}±0.04
pH	3.23^{a}±0.04	3.20^{a}±0.05
Density (gm/cm^3)	1.0534^{a}±0.0003	1.0463^{a}±0.0002
O_2 (ppm)	1.66^{b}±0.05	3.00^{a}±0.05
TSS (°Brix) (%)	11.25^{a}±0.05	11.09^{b}±0.04
Titratable acidity % (TA %)	24.70^{a}±0.05	24.15^{b}±0.03
Citric acid (%)	0.17^{a}±0.01	0.16^{a}±0.02

PPME (ppm)	-	200
PPME Antioxidants Activity (%)	-	80.68±1.21

Means with different letter in the same row are significantly different ($p \leq 0.05$)
PPME: Potato peel methanol extract.

Table 2. Physicochemical evaluation from formulations of lemon carbonated soft drinks after production using standard formulation (citric acid) and formulation containing PPE. Values expressed are means of triplicate measurements

Physicochemical characteristics	Standard formulation	PPE-formulation
CO_2 (V/V)	4.46^{a}±0.02	4.32^{b}±0.04
pH	3.23^{a}±0.04	3.20^{a}±0.05
Density (gm/cm^3)	1.0534^{a}±0.0003	1.0463^{a}±0.0002
O_2 (ppm)	1.66^{b}±0.05	3.00^{a}±0.05
TSS (°Brix) (%)	11.25^{a}±0.05	11.09^{b}±0.04
Titratable acidity % (TA %)	24.70^{a}±0.05	24.15^{b}±0.03
Citric acid (%)	0.17^{a}±0.01	0.16^{a}±0.02
PPE (ppm)	-	200
PPE Antioxidants Activity (%)	-	80.68±1.21

Means with different letter in the same row are significantly different ($p \leq 0.05$)

Table 3. physicochemical evaluation from formulations of lemon soft drinks after production and during 120 days using standard formulation (citric acid) and formulation containing PPE. Values expressed are means of triplicate measurements

Physicochemical characteristics	Lemon soft drink formulation	Storage period						
		0	1	2	3	4	5	6
CO_2 (V/V)	Standard	$4.46^{a} \pm 0.03$	$3.76^{b} \pm 0.05$	$3.42^{c} \pm 0.02$	$3.17^{d} \pm 0.01$	$2.20^{e} \pm 0.02$	$1.95^{f} \pm 0.01$	$1.62^{g} \pm 0.02$
	PPME	$4.32^{a} \pm 0.03$	$3.36^{b} \pm 0.02$	$3.07^{c} \pm 0.05$	$2.76^{d} \pm 0.05$	$2.53^{e} \pm 0.01$	$2.29^{f} \pm 0.03$	$2.15^{g} \pm 0.08$
pH	Standard	$3.23^{a} \pm 0.05$	$3.12^{b} \pm 0.05$	$3.01^{c} \pm 0.02$	$2.90^{d} \pm 0.05$	$2.85^{de} \pm 0.01$	$2.75^{e} \pm 0.05$	$2.46^{f} \pm 0.01$
	PPME	$3.20^{a} \pm 0.06$	$3.00^{b} \pm 0.05$	$2.94^{c} \pm 0.01$	$2.89^{d} \pm 0.02$	$2.63^{e} \pm 0.02$	$2.44^{f} \pm 0.02$	$2.21^{g} \pm 0.05$
Density (gm/cm^3)	Standard	$1.0534^{a} \pm 0.0002$	$1.0420^{b} \pm 0.0005$	$1.0426^{b} \pm 0.0004$	$1.0430^{bc} \pm 0.0018$	$1.0425^{bc} \pm 0.0004$	$1.0432^{c} \pm 0.0002$	$1.0443^{d} \pm 0.0002$
	PPME	$1.0463^{a} \pm 0.001$	$1.0457^{b} \pm 0.0008$	$1.0442^{c} \pm 0.001$	$1.0426^{d} \pm 0.0005$	$1.0423^{d} \pm 0.0004$	$1.0421^{de} \pm 0.0005$	$1.0417^{e} \pm 0.0007$
O_2 (ppm)	Standard	$1.66^{g} \pm 0.00$	$2.00^{f} \pm 0.0$	$7.00^{e} \pm 0.0$	$10.00^{d} \pm 0.0$	$13.00^{c} \pm 0.0$	$16.60^{b} \pm 1.15$	$24.00^{a} \pm 4.0$
	PPME	$3.00^{g} \pm 0.00$	$9.00^{f} \pm 0.0$	$11.00^{e} \pm 0.0$	$14.00^{d} \pm 0.0$	$20.33^{c} \pm 0.9$	$27.66^{b} \pm 1.12$	$31.95^{a} \pm 0.8$
TSS (°Brix) (%)	Standard	$11.25^{a} \pm 0.03$	$11.19^{b} \pm 0.01$	$11.14^{c} \pm 0.01$	$11.09^{d} \pm 0.05$	$11.04^{e} \pm 0.05$	$10.93^{f} \pm 0.05$	$10.85^{g} \pm 0.05$
	PPME	$11.09^{a} \pm 0.07$	$11.08^{a} \pm 0.03$	$11.06^{a} \pm 0.02$	$11.05^{a} \pm 0.03$	$11.05^{a} \pm 0.03$	$11.02^{ab} \pm 0.08$	$11.02^{ab} \pm 0.01$
Titratable acidity % (TA %)	Standard	$24.70^{a} \pm 0.03$	$24.17^{b} \pm 0.01$	$24.26^{c} \pm 0.03$	$24.84^{d} \pm 0.04$	$25.04^{e} \pm 0.03$	$25.35^{f} \pm 0.02$	$25.64^{g} \pm 0.05$
	PPME	$24.15^{g} \pm 0.05$	$24.27^{f} \pm 0.12$	$24.69^{e} \pm 0.18$	$24.80^{d} \pm 0.03$	$24.98^{c} \pm 0.06$	$25.10^{b} \pm 0.03$	$25.34^{a} \pm 0.01$
Citric acid (%)	Standard	$0.17^{a} \pm 0.03$	$0.17^{a} \pm 0.01$	$0.17^{a} \pm 0.0$	$0.17^{a} \pm 0.02$	$0.17^{a} \pm 0.02$	$0.17^{a} \pm 0.01$	$0.16^{a} \pm 0.05$
	PPME	$0.16^{a} \pm 0.03$	$0.16^{a} \pm 0.02$	$0.16^{a} \pm 0.03$	$0.15^{a} \pm 0.03$	$0.15^{a} \pm 0.03$	$0.15^{a} \pm 0.03$	$0.15^{a} \pm 0.03$

Means with different letter in the same row are significantly different ($p \leq 0.05$)

Table 4. Growth of microorganisms (CFU/100ml) from formulations of lemon soft drinks after 120 days using standard formulation (citric acid) and formulation containing PPE. Values expressed are means of triplicate measurements

Microorganism	Lemon soft drink formulation	Storage period (months)						
		0	1	2	3	4	5	6
Yeast	Standard	50	98	75	67	50	25	10
	PPME	2.50	3.30	7.30	3.50	0.60	0.60	0.0
Mold	Standard	2.0	4.0	10	9.0	5.0	1.0	1.0
	PPME	1.0	3.60	1.50	0.50	0.0	0.0	0.0

Table 5. Growth of microorganisms (CFU/100ml) from formulations of lemon soft drinks after 120 days using standard formulation (citric acid) and formulation containing PPE. Values expressed are means of triplicate measurements

Microorganism	Lemon soft drink formulation	Storage period (months)						
		0	1	2	3	4	5	6
Bacteria count	Standard	160	203	256	318	297	254	209
	PPME	2.3	5.60	3.30	2.00	1.60	0.60	0.0
Aciduric bacteria	Standard	22	45	64	104	87	80	50
	PPME	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Coliform	Standard	0.0	0.0					
	PPME	0.0	0.0					
E. *coli*	Standard	0.0	0.0					
	PPME	0.0	0.0					

Table 6. Deterioration factor from formulations of lemon carbonated soft drinks using standard formulation (citric acid) and formulation containing PPE. Values expressed are means of triplicate measurements

Microbial analysis	Standard formulation	PPE-formulation
Total count	60.06 %	8.69 %
Aciduric bacteria	225.72 %	0.0 %
Yeast	8.2 %	2.0 %
Mold	60.0 %	7.0 %
E. coli & coliform	0.0 %	0.0 %

* Deterioration factor = Control - Average of 6-months/ Control × 100
PPE: Potato peels extract

Table 7. Sensory evaluation from formulations of lemon soft drinks after production and during 120 days using standard formulation (citric acid) and formulation containing PPE. Values expressed are means of triplicate measurements

Sensory characteristics	Lemon soft drink formulation	Storage period						
		0	1	2	3	4	5	6
Taste	Standard	$8.15^{a} \pm 0.51$	$7.00^{b} \pm 0.00$	$6.1^{c} \pm 0.51$	$4.70^{d} \pm 0.64$	$4.70^{d} \pm 0.73$	$4.70^{d} \pm 2.04$	$2.50^{e} \pm 1.18$
	PPME	$7.40^{a} \pm 1.19$	$7.40^{a} \pm 2.01$	$6.10^{b} \pm 0.20$	$6.00^{b} \pm 1.55$	$5.70^{b} \pm 1.17$	$4.00^{c} \pm 0.64$	$3.00^{c} \pm 0.81$
Odor	Standard	$8.09^{a} \pm 0.31$	$7.5^{b} \pm 1.17$	$5.50^{c} \pm 1.64$	$4.60^{cd} \pm 0.67$	$4.15^{d} \pm 0.56$	$4.00^{d} \pm 1.80$	$2.50^{e} \pm 1.54$
	PPME	$8.00^{a} \pm 1.02$	$8.00^{a} \pm 1.28$	$6.80^{b} \pm 0.78$	$5.60^{c} \pm 1.15$	$5.00^{c} \pm 0.70$	$5.00^{c} \pm 1.80$	$4.00^{d} \pm 1.03$
Appearance	Standard	$8.54^{a} \pm 0.47$	$7.90^{ab} \pm 0.71$	$7.70^{b} \pm 0.54$	$7.70^{b} \pm 0.98$	$7.70^{b} \pm 1.72$	$7.20^{bc} \pm 1.90$	$6.60^{c} \pm 0.73$
	PPME	$8.10^{b} \pm 1.11$	$8.10^{a} \pm 0.40$	$7.80^{a} \pm 1.00$	$7.20^{ab} \pm 1.15$	$6.70^{b} \pm 0.68$	$5.30^{c} \pm 2.89$	$5.10^{c} \pm 2.70$
Overall acceptability	Standard	$8.26^{a} \pm 2.18$	$7.46^{b} \pm 1.18$	$6.33^{c} \pm 2.82$	$5.93^{cd} \pm 2.03$	$5.51^{d} \pm 0.87$	$5.31^{d} \pm 1.96$	$3.53^{e} \pm 1.27$
	PPME	$7.83^{b} \pm 1.48$	$7.83^{a} \pm 0.33$	$6.90^{b} \pm 0.97$	$6.26^{bc} \pm 1.24$	$5.83^{c} \pm 0.97$	$4.76^{d} \pm 1.42$	$4.03^{d} \pm 1.02$

Means with different letter in the same row are significantly different ($p \leq 0.05$)