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Effects of Enrichment with Globe Artichoke Roots and Jerusalem Artichoke Tubers on Nutritional and Functional Properties of Whey Beverages

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Abstract: Whey is a major by-product obtained from cheese manufacture often disposed as waste. Whey has the capability to work as an antioxidant, antitumor, hypolipidemic, antihypertensive, chelating agent and antiviral. Traditionally Globe artichoke roots (*Cynara scolymus* L.) and Jerusalem artichoke tubers (*Helianthus tuberosus* L.), have long been used as food and medicine. Nowadays, they are considered as functional foods, owing to their nutritional properties and chemical composition and are rich source of bioactive phenolic compounds and also inulin, fibre, minerals and Vitamins. In the present study, whey beverages, fortified with Globe artichoke roots and Jerusalem artichoke tubers in order to combine the high nutritive value. Changes were observed in phenolic and flavonoid compounds, vitamin B complex and C by HPLC, minerals and counts of total bacterial, *L. delbrueckii* subsp. *bulgaricus* and *S. salivarius* subsp. *thermophilus* of different produced functional whey beverages during storage of the beverages at 4°C. Sensory properties were also evaluated. It is interesting to mention that whey based functional beverages with 5% Jerusalem artichoke tubers (W_1) had better flavour and overall acceptability. The shelf-life of the beverages with Globe artichoke roots; Jerusalem artichoke tubers and each of them together as functional beverages was estimated to be 28 days under refrigerated storage (at 4°C).

Key words: Globe Artichoke Roots · Jerusalem artichoke Tubers · Functional Foods · Functional Whey Beverage

INTRODUCTION

Production of whey based beverages started in 1970's and until today a wide range of different whey beverages has been developed. Depending on the type of casein coagulation, whey can be sweet or acid. Composition and properties of whey mainly depend on the technology of cheese manufacture and on the quality of milk used cheese production [1]. Liquid whey consists approximately 93% of water and contains almost 50% of total solids present in the milk of which lactose is the main constituent [2]. Lactose is the main constituent of whey while proteins represent less than 1% of total solids. In fewer amount also minerals and vitamins are present.

Whey beverages include wide range of products obtained by mixing native sweet, diluted or acid whey with different additives like tropical fruits (But also other fruits like apples, pears, strawberries or cranberries), some scientists have applied the addition of other flavouring agents like chocolate, coca, vanilla, vegetables, cereals (Mostly rice, oat and barley), honey, etc. Special attention is being paid to development of whey beverages production by whey fermentation with probiotic bacteria where the most important step is the choice of suitable culture of bacteria in order to produce functional beverage with high nutrient value and acceptable sensory characteristics. There are even some indications that fermentation of whey using yoghurt

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culture (*Lactobacillus bulgaricus* and *Streptococcus thermophilus*) produces a more intense yoghurt flavour compared to the one obtained when skim milk is fermented. This suggests the possibility of producing beverages from whey with similar sensory profiles to those of fermented milk drinks or with some flavour attributes of drinking yoghurt, following manufacturing procedures conventionally used for milk [3].

Globe artichoke roots (*Cynara scolymus* L.) and Jerusalem artichoke tubers (*Helianthus tuberosus* L.) are an ancient herbaceous perennial plant, originating from the Mediterranean area basin including Egypt, which today are widely cultivated all over the world. Globe artichoke roots and Jerusalem artichoke tubers are considered a healthy food due to its nutritional and chemical composition [4]. Globe artichoke roots and Jerusalem artichoke tubers contain proteins, minerals, low amount of fat and calories, so it is an excellent choice for traditional weight loss diets, dietary fibers and high proportion of phenolic [5].

Additions of extraction of Globe artichoke roots and Jerusalem artichoke tubers, which are known as a good source of iron and antioxidants, have proved to be very useful. That is especially important in production of whey beverages with improved nutritional value. They have proved that long-term consumption of this drink had an impact on reduction in the prevalence of anemia in children and adolescents. Also extraction of Globe artichoke roots and Jerusalem artichoke tubers is a rich source of bioactive phenolic compounds that exhibit lots of pharmacological activities. Different studies about artichoke have demonstrated their health-protective potential, especially their hepatoprotective anticarcinogenic and hypocholesterolemic activities [6]. In terms of natural polyphenolics from dietary plants, there is increasing evidence that a diet with an increased intake of these compounds is associated with reduced risk of cardiovascular diseases and certain types of cancer [7].

The aim of this research was to improve the production of functional whey-based beverages with highly nutritional and functional properties and investigate the effect of enrichment with Globe artichoke roots and Jerusalem artichoke tubers extracts.

MATERIALS AND METHODS

Materials: The roots of artichoke (*Cynara scolymus* L.,) were obtained from Kafer El Dawar district and tubers of Jerusalem artichoke (*Helianthus tuberosus* L.) were obtained from Sabahia Horticultural Research Station, Agriculture Research Center, Alexandria, Egypt. Whey was obtained from dairy farm in Sadat City, Egypt. Freeze dried DVS-ABY-1Nu-TRISH yoghurt cultures containing *S. salivarius* subsp. *thermophilus* and *L. delbrueckii* subsp *bulgaricus* strains were obtained from Chr. Hansen Inc. Laboratories, Denmark, by Misr Food Additives (MIFAD), Egypt.

Methods: Globe artichoke roots and Jerusalem artichoke tubers were washed with tap water and any deteriorated parts were removed, than sliced in dividedly to the reasonable thickness by conventional food slicing machine. The slices were immersed immediately in boiling water for 5 min. followed by immediate dipping in cold citric acid solution (1%) to inhibit polyphenol oxidase activity. After that slices were dried in electronic air oven at 55-65°C until samples reached constant weight. The recovered powder was kept in tight polyethylene bags and stored under freezing until use.

Preparation of Functional Whey Beverage Containing Jerusalem Artichoke Tubers and Globe Artichoke Roots: A volume of 2000 ml of whey with pH 6.2 was poured into 4sterile glass bottles (C, W_1, W_2 and W_3) in each 500 mL. Whey was pasteurized at 85°C for 15 min. After that, bottles cooled at fermentation temperature 42°C and immediately inoculated by adding 2% (v/v) of yoghurt cultures (*S. thermophilus* and *L. bulgaricus*), in each glass bottles. Jerusalem artichoke tubers and Globe artichoke roots were added in three glass bottles as follows: 5% Jerusalem artichoke tubers (W_1); 5% Globe artichoke roots (W_2) and 2.5% Jerusalem artichoke tubers + 2.5% Globe artichoke roots (W_3), respectively (Preliminary experiment was done to select the best concentration from Jerusalem artichoke tubers and Globe artichoke roots) and the fourth bottle without any additives (C). Fermentation was carried out until pH 4.6 was attained. During the incubation time samples were withdrawn every 1h for determination of pH value. When pH was reached, fermentations were stopped by quick cooling. The resulting beverages were distributed in sterile plastic bottles in triplicates and stored at 4°C for 28 days. Viable cell count (CFU/mL) was determined every 7 days of the storage.

Chemical Analysis: Moisture, total protein content, fat, crude fiber and ash contents were determined according to AOAC [8]. Carbohydrate content was determined by differences of total contents (Moisture, protein, fat, crude fiber and ash) from 100. Total, reducing and non-reducing sugars were determined by the Lane-Eynon method AOAC [8].

Determination of Inulin: Jerusalem artichoke tubers and Globe artichoke roots were homogenized with water (1:2 w/v) and heated at 120°C for 20 min (1 atm.) in a vertical retort Luferco; the treated sample materials were then filtered. Sample analysis was performed using a water's high performance liquid chromatography (HPLC) under the following conditions: column- (Aminex HPX-87C); detector- refractive index detector, Waters model 2414; eluent: water; flow rate: 0.3 ml/min; injected volume: 20 µl: column temperature: 80°C; detector temperature: 40°C. For the inulin quantitation, a commercial standard (Fluka-BioChemika 57614) was used. The sample was homogenized with water (1:2 w/v) and heated at 120°C for 20 min (1 atm.) in a vertical retort Luferco; the treated sample materials were then filtered and subjected to chromatography.

Minerals Content: The minerals (K, Ca, Mg, Na, Fe and Zn) were determined according to the AOAC [8] in the bio-products and plus Mn in the raw materials (Globe artichoke roots and Jerusalem artichoke tubers). Sodium and Potassium were determined by coring flame photometer (Model 410, Japan), while calcium, magnesium, iron and zinc were determined using a Perkin Elmer Atomic Absorption Spectrophotometer (Model 2:6650, Shimazy) Japan.

Determination of Antioxidant Activity: The antioxidant activity of the sample was evaluated by using 2, 2-diphenyl-1-picryl-hydrazyl (DPPH) assay [9]. One hundred microliters of the ethanolic extract were added to 5 ml of 0.004 % (w/v) of DPPH in methanol. The mixture was vortexed for 15 s and then left to stand at room temperature for 30 min. Absorbance was measured at 517 nm against ethanol. The inhibition of DPPH free radicals (1%) was calculated according to the following formula: % inhibition = $\{(1-A_1)/A_0) \times 100\}$; where A_0 is the absorbance of the control and A_1 is the absorbance of the DPPH-sample.

For the functional whey beverage samples, extraction of antioxidant activity was carried out according to Li *et al*. [10] with some modifications as follows: addition of 20 ml of solvent (Each 100 ml contains 15 ml 1N HCL and 85 ml ethanol 95%) to 10g of functional whey beverage in 50 ml brown bottles and shaking for 90 min at 30 °C in a rotary shaker set at 200 rpm were done. Then, the mixture was centrifuged at 2500 g for 45 min at 5 °C. The supernatant fluids were analyzed for antioxidant activity as described for Globe artichoke roots and Jerusalem artichoke tubers.

Identification of Phenolic and Flavonoid Compounds by HPLC: The HPLC system was used for analysis of the methanolic extract of samples to identify the phenolic and flavonoid compounds according to the methods of Mattila *et al*. [11]. Ultraviolet (UV) detector at wavelength 280 nm and quaternary HP pump (Series 1100) was used for phenolic compounds, while Ultraviolet (UV) detector at wavelength 330 nm and quarter HP pump (Series 1050) was used for flavonoid compounds. The column was (Agilent Zerbax ODS C18 column 150×4.5 mm) and temperature was maintained at 35°C. Gradient separation was carried out with methanol and acetonitrile as mobile phase at flow rate of 1 ml/min. Phenolic acids and flavonoid standards from Sigma Co. were dissolved in a mobile phase and injected into HPLC. Retention time and peak area were used to calculate the concentration of the previous compounds by the data analysis of Hewllet Packard software.

Identification of Vitamin C by HPLC: Vitamin C content was analyzed using HPLC described by Romeu-Nadal *et al*. [12]. The samples were protected from light by wrapping tubes and flasks with aluminum foil and preparing the samples in a darkened room. 300µl of sample mixed with 300 µl of 0.56% (w/v) meta-phosphoric acid solution were added to the same centrifuge and filtration tube, which was shaken for 30 seconds and centrifuged at 10°C (10 minutes, 3000Xg). Ascorbic acid was identified by comparing the retention time of the sample peak with that of the ascorbic standard at 254 nm. Quantification was carried out using external standardization.

Identification of Vitamin B Complex by HPLC: Vitamin B complex of samples was determined by reversed-phase high-performance liquid chromatography (RP-HPLC) according to the method of Papadoyannis *et al*. [13] with some modifications. The ground sample (0.5 g) was macerated in a glass blender containing 5 mL 30% metaphosphate. The macerate was diluted to 25 ml in a graduated flask with distilled water. The diluted solution was then filtered through a 0.45 µm filter. The filtrate was then injected directly into the HPLC with the injection volume 10 µl, column (Hypersil™ODS C18 µm, 100 x 4.6 mm), temperature 30°C, flow rate (0.8 ml/min), detector (Ultraviolet detector, wavelength was set at 280 nm) and a mobile phase of phosphoric acid and methanol at different time intervals 0, 20 and 25 min in a ratio of 90:10, 30:70 and 30:70, respectively.

Estimation of Shelf-Life: The self-life of the functional whey beverages was defined as the period of refrigerated storage (4°C) during which pH remained above 4.0 and the number of viable cell counts above 10^6 cfu/ml. Storage period was carried out for 28 days with periodical observation of pH and viability of starter culture.

Determination of pH: The pH was determined using glass electrode pH meter (Persic model pH 900, Switzerland).

Microbiology Analysis: Serial decimal dilutions in sterile peptone water (0.1%) were prepared from every beverages kind (1 ml sample). Then 1 cm^3 of aliquots was plated over selected culture media as follows: The total bacterial count was determined; Lactobacilli count was estimated on the selective medium for lactobacilli (MRS) and streptococci count on M17 agar medium respectively. Coliform bacteria and Moulds & Yeasts were enumerated according to IDF [14].

Sensory Properties: The sensory properties of fresh and stored functional whey beverages from different treatments were assessed by a taste panel of 10 persons of staff members of the Dairy Department who assessed the produced four beverage samples. The four beverage samples were scored for colour, taste, texture, flavour and overall acceptability. Scores were based on a hedonic scale of 1 to 9 where: 1 = dislike very much (Very bad) and 9 = like very much (Excellent).

Statistical Analysis: Results from the facial hedonic scale record sheets were collated and input into a SPSS version 15 database, mean, standard deviations and p-values were calculated for each sample. P-values less than 0.05were considered statistically significant.

RESULTS AND DISCUSSION

Gross Chemical Composition of Whey: Whey is a fairly dilute product with total solids of 6.5±0.55%. As mentioned before the solids are basically consisted by lactose, proteins, fat and ash. According to its average composition whey was 93.5±0.68% Moisture and contains above 50% of total solids present in the milk of which lactose was the main constituent 5.00±0.18; proteins constituted 0.80±0.06% of dry matter. Ash and milk fat were also present but in less amounts (0.5±0.08 and0.05±0.04), respectively. These results are in agreement with the values reported by Beucler *et al*. [2].

Table 1: Gross chemical composition of Jerusalem artichoke tubers and Globe artichoke roots

Components (%)	Jerusalem artichoke tuber	Globe artichoke roots
Moisture content	6.80±0.84	3.80±0.59
Total solids	93.20±0.89	96.20±0.70
Crude protein	2.6±0.11	2.1±09
crude fat	0.8±0.07	1.3±0.07
Ash	5.2±0.05	5.3±0.04
Curd fiber	4.4±0.09	0.7±0.08
Total carbohydrate	80.2±0.08	86.8±0.06
Total sugar	45, 46±0.79	38.2±0.70
Reducing sugar	4.41±0.69	3.9±0.71
Non reducing sugar	41.05±0.29	34.3±0.31
Inulin	21.46	13.24
Antioxidant activity (%)	67.22	59.57

Table 2: Minerals (mg/100g); Phenolic compounds (g/100g); Flavonoids compounds (g/100g) and Vitamins composition (mg/100g) dry weight of Jerusalem artichoke tubers and Globe artichoke roots

Minerals (mg/100g)	Jerusalem artichoke tuber	Globe artichoke roots
K	208, 39	147.86
Ca	43.38	31.54
Mg	13.49	11.22
Na	41.38	32.73
Fe	7.12	5.33
Zn	4.76	2.37
Mn	0.39	0.36
Phenolic compounds (mg/100g)		
Pyrogallol	379.77	407.12
Chlorogenic	40, 688	98.955
Caffeic	34.854	37.810
p-OH-benzoic	9.651	41.950
3-Hydroxy-tyroso	17.105	19.830
Catechol	9.266	5.188
Catechein	7.280	1.739
Flavonoid compounds (mg/100g)		
Luteolin 6-arbinose 8-glucose	69.78	76.63
Luteolin 6- glucose 8-arbinose	7.13	7.81
Luteolin 7- glucose	2.04	2.65
Luteolin	1.04	1.61
Apigenin 6-arbinose 8-glucose	25.39	28.60
Apigenin 6- glucose 8-rhamnose	19.74	11.20
Apigenin 6- rhamnose 8- glucose	1.19	1.10
Apigenin 7-O-neohespiroside	0.39	0.93
Apigenin 7- glucose	0.34	0.52
Apigenin	0.06	0.09
Vitamins composition (mg/100g)		
Vitamin B 6 (Pyridoxine)	267.404	274.086
Vitamin B1 (Thiamine)	195.721	200.612
Vitamin B3 (Niacin)	157.553	29.897
Vitamin B9 (Folic acid)	18.255	13.078
Vitamin B2 (Riboflavin)	6.644	3.962
Vitamin B 12(Cobalamine)	N.D	N.D
Vitamin C (Ascorbic Acid)	17.077	4.802

N.D: Not Defined

Gross Chemical Composition of Jerusalem Artichoke Tubers and Globe Artichoke Roots: Gross chemical composition of Jerusalem artichoke tubers and Globe artichoke roots investigated in the present study was given in Table (1). It was obvious that Jerusalem artichoke tubers and Globe artichoke roots are characterized by their high content of carbohydrates (80.2 and 86.8 mg/100g), respectively. The principal storage carbohydrate of Jerusalem artichoke tubers and Globe artichoke roots is inulin, the amounts of inulin in Jerusalem artichoke tubers and Globe artichoke roots were (21.46 and 13.24 mg/100g), respectively. The data showed that Jerusalem artichoke tubers and Globe artichoke roots were rich source of sugar containing high concentrations of total sugars (45.46% and 38.2), reducing sugars (4.41% and 3.9) and non-reducing sugars (41.05 % and 34.3), respectively. Along with moderate content of crude protein (2.6 and 2.1 g/100g), crude fat (0.8 and 1.3 g/100g) and Curd fiber (4.4 and 0.7 g/100g), respectively [5]. Globe artichoke roots and Jerusalem artichoke tubers contain proteins, minerals, low amount of lipids and dietary fibers. Ash was (5.2 and 5.3 g/100g) on dry weight basis in Jerusalem artichoke tubers and Globe artichoke roots, respectively. Data presented are in a good agreement with the reports given ash content as high as 4.7% [15].

Globe artichoke roots and Jerusalem artichoke tubers contain proteins, minerals, low amount of lipids, dietary fibers and high proportion of phenolic [5]. Globe artichoke roots and Jerusalem artichoke tubers extracts were low in fat and calories, so it was an excellent choice for traditional weight loss diets.

Mineral Content of Jerusalem Artichoke Tubers and Globe Artichoke Roots: Jerusalem artichoke tubers and Globe artichoke roots have a high mineral content. The data given in Table (2) showed that K (208. 39 mg/100g), Ca (43.38 mg/100g) and Na (41.38 mg/100g) were the most abundant mineral elements of Jerusalem artichoke tubers. This was also true for the Globe artichoke roots, since it contained 147.86, 31.54 and 32.73mg/100g of K, Ca and Na, respectively. It was obvious that Mg contents were (13.49 and 11.22 mg/100g) for Jerusalem artichoke tubers and Globe artichoke roots, respectively. They especially were rich in Fe content, it was 7.12 mg/100g and 5.33 mg/100g Iron concentrations, for instance, are around three times higher than in potatoes [16], While relatively little Mn contents were (0.39 and 0.36 mg/100g) and Zn (4.76 and 2.37 mg/100g) for Jerusalem artichoke tubers and Globe artichoke roots, respectively. The data presented in Table (2) are in a good agreement with the data reported by Van Loo *et al*. [17].

Antioxidant Activity of Jerusalem Artichoke Tubers and Globe Artichoke Roots: The data presented in Table (2) indicated that Jerusalem artichoke tubers exhibited antioxidant activity (67.22%) higher than of Globe artichoke roots, (59.57%). Globe artichoke roots and Jerusalem artichoke tubers were healthy addition to any eating plan due to containing antioxidants and phytonutrients which have been recognized as important compounds that reduce the risk of several chronic diseases, which are thought to protect cells against the attack by free radicals [4].

Identification of Phenolic and Flavonoid Compounds of Jerusalem Artichoke Tubers and Globe Artichoke Roots by HPLC: Phenolic and flavonoid compounds of methanolic extracts of Jerusalem artichoke tubers and Globe artichoke roots were shown in Table (2). Moreover, Jerusalem artichoke tubers and Globe artichoke roots had interesting nutritional characteristics related to its high content of phenolic and flavonoids compounds as the major chemical components. Seven phenolic compounds and ten flavonoids compounds were identified in Jerusalem artichoke tubers and Globe artichoke roots.

Table (2) showed the phenolic compounds in Jerusalem artichoke tubers and Globe artichoke roots. Three major compounds in Jerusalem artichoke tubers and Globe artichoke roots were the salts of Pyrogallol, chlorogenic acid (5-O-caffeoylquinic acid) and Caffeic. Pyrogallol was found to account (379.77 and 407.12 mg/100g), followed by Chlorogenic acid was found to account (40.688 and 98.955 mg/100g), followed by Caffeic (34.854 and 37.810 mg/100g), p-OH-benzoic (9.651 and 41.950 mg/100g), 3-Hydroxy-tyroso (17.105 and 19.830 mg/100g), Catechol (9.266 and 5.188 mg/100g) and Catechein (7.280 and 1.739 mg/100g), respectively. Lattanzio *et al.* [18] reported that the main phenolic compounds were the caffeic acid derivatives which include the caffeoylquinic acid derivatives. Using the International Union of Pure and Applied Chemistry (IUPAC) nomenclature, 5-O-caffeoylquinic acid (Chlorogenic acid) was the most abundant single substance (39%), followed by 1, 5-O-dicaffeoylquinic acid (21%) and 3, 4-O-dicaffeoylquinic acid (11%), based on total caffeoylquinic acid content. Artichoke contains

chlorogenicacid (5-O-caffeoyl-quinic acid) which has cholagogic properties and induced sweetness. Furthermore, the Ferulic content in methanolic extracts of artichoke was very low (0.702 and 0.807 mg/100g). The above phenolic substances have an important scavenging activity against reactive oxygen species (ROS) and free radicals and perform as a protective shield against oxidative damage to biological molecules, such as proteins, lipids and DNA [19].

Flavonoids are large group of compounds occurring ubiquitously in food plants. They occur as glycosides and contain several phenolic hydroxyl groups on their ring structures. The most important property of almost every group of flavonoids is their capacity to act as antioxidants [20]. Table (2) showed the flavonoid compounds in Jerusalem artichoke tubers and Globe artichoke roots. Luteolin 6-arbinose 8-glucose was the most abundant flavonoid in Jerusalem artichoke tubers and Globe artichoke roots (69.78 and 76.63 mg/100g), respectively followed by Apigenin 6-arbinose 8-glucose (25.39 and 28.60 mg/100g), Apigenin 6- glucose 8-rhamnose (19.74 and 11.20 mg/100g) and Luteolin 6-glucose 8-arbinose (7.13 and 7.81 mg/100g). More recently, three flavanone glycosides (Apigenin, Apigenin 7- glucose and Apigenin 7-O-neohespiroside) have been identified as minor phenolic compounds in Jerusalem artichoke tubers and Globe artichoke roots [21]. However, these observations require verification. Brown and Rice-Evans [22] found that luteolin is a strong antioxidant that protects low density lipoproteins from oxidation, plays an important role in the appearance of food plants and therefore in food acceptance by consumers.

Vitamins of Jerusalem Artichoke Tubers and Globe Artichoke Roots: Table 2 showed the values of water soluble vitamin B complex (B1, B2, B3, B6, B9 and B12) and Vitamin C content by HPLC of Jerusalem artichoke tubers and Globe artichoke roots. The results in Table (2) showed that Jerusalem artichoke tuber and Globe artichoke roots had relatively high levels of Pyridoxine (Vitamin B6), Thiamin (Vitamin B1) and Niacin (Vitamin B3) [(267.404 and 274.086 mg/100g), (195.721 and 200.612 mg/100g) and (157.553 and 29.897 mg/100g)], respectively while other vitamins in the B complex were presented {Folic (Vitamin B_9) and Riboflavin (Vitamin B2)} for Jerusalem artichoke tuber and Globe artichoke roots [(18.255 and 13.078 mg/100g) and (6.644 and 3.962 mg/100g)], respectively. Ascorbic acid (Vitamin C) concentrations (17.077 and 4.802 mg/100g) were recorded for Jerusalem artichoke tubers and Globe artichoke roots, respectively. All varieties of artichoke contain the same vitamins and minerals such as: vitamins C, B1, B2, B3 and Inulin which helps those suffering from diabetes, liver problems, high blood pressure and other bowel disorders [23]. Jerusalem artichoke tubers and Globeartichoke roots are a good source of vitamins B complex and vitamin C [17]. In the present study Vitamin B12 was not detected for Jerusalem artichoke tubers and Globe artichoke roots.

Minerals Content in Different Produced Functional Whey Beverage Samples: Table 3 showed minerals i.e. potassium (K), calcium (Ca), sodium (Na), magnesium (Mg), iron (Fe) and zinc (Zn) content of different beverages. Results indicated that minerals content increased as a result of Jerusalem artichoke tuber and Globe artichoke roots added compared to control. Samples W_1 and W_3 contained the highest value of K (260.6 mg/100g, 232.38 mg/100g), respectively followed by sample (W_2) 202.28 mg/100g while the lowest value was 102.80 mg/100g for sample (C). The values (Table 3) recorded for sodium 73.70, 71.74, 70.69 and 63.76 mg/100g for different beverage samples (W1, W3, W2 and C), respectively. The composition of different beverages expounded low sodium to potassium ratio that is essential for maintenance of blood pressure. The instant results for minerals contents in different beverages based functional drink are in conformity with the previous findings of Jindal *et al*. [24]. Different beverages are also an excellent source of bioavailable calcium that improve bone health. Moreover, calcium from the different beverages were readily absorbed in the intestine, facilitated by the presence of lactose. Calcium content ranged 86.18 to 130.12 mg/100g. The highest value was recorded in sample (W_1). The increment in Fe content of produced beverages as a result of Jerusalem artichoke tuber and Globe artichoke roots that added, this means Jerusalem artichoke tuber and Globe artichoke roots were rich source of Fe. Regarding to iron content data showed that iron content of sample (W_2) and (W_3) were 13.69 and 11.11 mg/100g, followed by (W_1). The addition of Jerusalem artichoke tuber and Globe artichoke roots which were known as a good source of iron and antioxidants had proved to be very useful. That was especially important in production of whey beverages with improved nutritional value. Best example for supporting this work was Brazilian group

Table 3: Phenolic compounds; Flavonoid compounds content; Vitamins and Minerals composition of different produced functional whey beverage samples (g/100g) dry weight

Phenolic compounds (g/100g)	Storage (days)	Control	5 % Jerusalem artichoke tuber (W_1)	5% Globe artichoke roots (W_2)	2.5 % Jerusalem artichoke tuber + 2.5 % Globe artichoke roots (W_3)
Pyrogallol	Fresh	0.89	4.71	4.82	4.61
	14	0.52	4.29	4.73	2.37
	28	0.43	1.17	3.7	1.01
Chlorogenic	Fresh	0.58	3.38	3.6	82
	14	0.38	2.97	3.2	0.67
	28	0.72	2.92	0.73	0.6
Caffeic	Fresh	0.02	0.85	0.93	0.71
	14	0.02	0.59	0.7	0.52
	28	0.02	0.57	0.61	0.49
3-Hydroxy-tyroso	Fresh	0.01	0.42	0.87	0.36
	14	0.01	0.41	0.81	0.38
	28	0.01	0.37	0.76	0.23
p-OH-benzoic	Fresh	0.01	0.27	0.31	0.21
	14	0.01	0.24	0.3	0.22
	28	0.01	0.2	0.27	0.18
Catechin	Fresh	0.04	0.53	0.45	0.34
	14	0.04	0.45	0.38	0.29
	28	0.01	0.37	0.32	0.22
Catechol	Fresh	0.04	0.4	0.38	0.34
	14	0.04	0.37	0.35	0.3
	28	0.04	0.35	0.32	0.29
Flavonoid compounds (g/100g)					
Luteolin 6-arbinose 8-glucose	Fresh	0.33	15.96	17.47	10.57
	14	0.31	15.71	16.82	7.62
	28	0.23	14.47	15.73	3.43
Luteolin 6- glucose 8-arbinose	Fresh	0.29	1.93	2.19	0.81
	14	0,26	1.78	1.92	0.27
	28	0.22	1.71	1.84	0.23
Luteolin 7- glucose	Fresh	0.02	0.16	0,08	0.14
	14	0.01	0.06	0.05	0.02
	28	0.01	0.05	0.04	0.01
Luteolin	Fresh	0.02	0.13	0.04	0.12
	14	0.02	0.09	0.13	0.02
	28	0.01	0.07	0.11	0.01
Apigenin 6- glucose 8-rhamnose	Fresh	0.17	0.93	0.46	0.82
	14	0.09	0.77	0.46	0.46
	28	0.07	0.44	0.42	0.41
Apigenin 6-arbinose 8-glucose	Fresh	0.01	0.40	0.42	0.33
	14	0.01	0.31	0.31	0.19
	28	0.01	0.30	0.09	0.03
Apigenin 6- rhamnose 8- glucose	Fresh	0.01	0.34	0.23	0.06
	14	0.01	27.0	17	0.05
	28	0.01	0.11	0.04	0.03
Apigenin 7- glucose	Fresh	0.03	0.15	0,55	0.52
	14	0.01	0.13	0.51	0.44
	28	0.01	0.12	0.18	0.16
Apigenin	Fresh	0.12	0.11	0.03	0.05
	14	0.11	0.11	0.01	0.01
	28	0.06	0.06	0.01	0.01
Apigenin 7-O-neohespiroside	Fresh	0,01	0,07	0,08	0.04
	14	0,01	0.04	0.05	0.02
	28	0.01	0.03	0.03	0,01

Table 3: Continued

Vitamins composition (g/100g)					
Vitamin B1 (Thiamine)	Fresh	0.23	2.99	0.59	0.3
	14	0.22	2.99	0.58	0.3
	28	0.18	2.80	0.45	0.28
Vitamin B3 (Niacin)	Fresh	0.48	0.77	0.72	0.41
	14	0.48	0.76	0.7	0.41
	28	0.27	0.59	0.49	0.05
Vitamin B2 (Riboflavin)	Fresh	0.79	2.85	2.49	0.67
	14	0.78	2.84	2.49	0.66
	28	0.77	2.78	2.26	0.48
Vitamin B6 (Pyridoxine	Fresh	0.19	2.95	1.82	0.32
	14	0.19	2.93	1.79	0.32
	28	0.18	2.83	1.25	0.3
Vitamin B_9 (Folic)	Fresh	0.15	1.23	1.21	0.54
	14	0.15	1.22	1.19	0.54
	28	0.08	0.58	0.26	0.05
Vitamin C (Ascorbic Acid)	Fresh	N.D	9.48	6.49	3.68
	14	N.D	9.40	6.46	3.65
	28	N.D	4.51	4.43	2.87
Minerals (mg/100)					
K		102.8	260.6	202.28	232.38
Ca		86.18	130.12	109.03	122
Na		63.76	73.7	70.69	71.74
Mg		19.74	35.36	34.24	33.49
Fe		7.01	10.42	13.69	11.11
Zn		2.87	4.51	4.07	3.81

C: control, W1: 5% Jerusalem artichoke tuber W2: 5% Globe artichoke roots, W3: 2.5% Jerusalem artichoke tuber + 2.5% Globe artichoke roots. N.D: Not Defined

of scientists who had developed a whey drink flavored by addition of strawberry concentrate and fortified with ferrous bisglycinate. They had proved that long-term consumption of this drink had an impact on reduction in the prevalence of anemia in children and adolescents [25]. Zinc contents in samples (W1) and (W2) increased with Jerusalem artichoke tuber and Globe artichoke roots added amounted in 4.51 and 4.07 mg/100g. This means Jerusalem artichoke tuber and Globe artichoke roots were good source of zinc. These results are in a good agreement with the data reported by Van Loo *et al*. [17].

Identification of Phenolic and Flavonoid Compounds of Different Produced Functional Whey Beverage Samples by HPLC: Phenolic and flavonoid compounds of methanolic extracts of different produced functional whey beverage samples as determined by HPLC analysis are shown in Table (3). Results indicated that Phenolic and flavonoid compounds increased as a result of addition of Jerusalem artichoke tuber and Globe artichoke roots which contained high proportion of phenolic and flavonoid compounds [5]. Seven phenolic compounds and ten flavonoids compounds were identified in water extracts in whey beverage.

Table (3) showed the Phenolic compounds of different produced functional whey beverage samples. Samples W_1 and W_2 contained the highest value of Pyrogallol (4.71 and 4.82 mg/100g) followed by Chlorogenic (3.38 and 3.60 mg/100g) followed by Caffeic (0.85 and 0.93 mg/100g), p-Hyd-benzoic (0.42 and 0.87 mg/100g), 3-Hyd-tyraso (0.27 and 0, 31 mg/100g), Catechol (0.53 and 0.45 mg/100g) and Catechin (4.40 and 0.38 mg/100g), respectively. Artichoke by-products were also a potential good source of antioxidant activity because it contains large amounts of polyphenols that possess high antioxidant activity. Also Wang *et al*. [26] mentioned that the chlorogenic acid (5-O-caffeoyl-quinic acid), an ester of caffeic acid with quinic acid, have received considerable attentions for their wide distribution and potential biological effects.

Table (3) showed the most flavonoid compounds identified in different produced functional whey beverage samples. Luteolin 6-arbinose 8-glucose, Luteolin 6- glucose 8-arbinose, Apigenin 6- glucose 8-rhamnose, Apigenin 6-arbinose 8-glucose and Apigenin 6- rhamnose 8- glucose. Luteolin 6-arbinose 8-glucose was the most abundant flavonoid. Samples W_1 and W_2 contained the highest value of (15.96 and 17.47 mg/100g), respectively

followed by Luteolin 6- glucose 8-arbinose (1.93 and 2.19 mg/100g), Apigenin 6- glucose 8-rhamnose (0.93 and 0.46 mg/100g), Apigenin 6-arbinose 8-glucose (0.40 and 0.42 mg/100g) and Apigenin 6- rhamnose 8- glucose (0.34 and 0.23 mg/100g). More recently, two flavanone (Apigenin and Apigenin 7-O-neohespiroside) have been identified as minor flavonoid compounds in different produced functional whey beverage samples However, these observations require verification. Sa´nchez-Rabaneda *et al*. [21]. However, these observations require verification. Brown & Rice-Evans [22] found that luteolin is a strong antioxidant that protects low density lipoproteins from oxidation, plays an important role in the appearance of food plants and therefore in food acceptance by consumers.

Vitamins Composition of Different Produced Functional Whey Beverage: Table 3 showed the values of water soluble vitamin B complex (B1, B2, B3, B6, B9 and B12) and Vitamin C content by HPLC of functional whey beverage samples. Results indicated that vitamin B complex and vitamin C contents increased in the functional whey beverage as a result of Jerusalem artichoke tuber and Globe artichoke roots added compared to control. Van Loo *et al*. [17] stated that Jerusalem artichoke tuber and Globe artichoke roots are a good source of vitamins B complex and vitamin C, an important antioxidant.

The obtained values of the vitamin B complex content in W_1, W_2 functional whey beverage samples were higher than W_3 and control. W_1 samples for Thiamin, Niacin, Riboflavin, Pyridoxine, Folic and Ascorbic acid, were found to be (2.99, 0.76, 2.84, 2.93, 1.22 and 9.40 mg/100g), respectively. While W_2 samples for Thiamin, Niacin, Riboflavin, Pyridoxine, Folic and Ascorbic acid, were found to be (0.58, 0.70, 2.49, 1.79, 1.19 and 6.46 mg/100g), respectively throughout 14 days of storage period and then decreased throughout 28 days storage period. In the present study Vitamin B12 (Was not defined for all functional whey beverage samples during storage period and ascorbic acid was not detected for all control samples during storage period.

Changes in pH of Different Produced Functional Whey Beverages During the Storage Period: The effects of Jerusalem artichoke tuber and Globe artichoke roots addition on pH values of functional whey beverage samples during refrigerated storage for up to 28 days are shown Fig. 1. The pH level above 4 is generally required for a functional whey beverage samples throughout storage [27]. Results in Fig. 1 showed that the addition of Jerusalem artichoke tuber and Globe artichoke roots to the whey beverages affected the initial pH value from (4.62 to 4.82) and the pH values for all whey beverage samples were ranged from 4.46 to 4.34 at the end of storage. Average pH value of functional whey beverage samples obtained with the Jerusalem artichoke tuber and Globe artichoke roots addition were found to be higher than the control samples. For all samples analyzed, the pH values decreased throughout the storage period. This is can be explained by further metabolic activity of starter culture during refrigerated storage. These results are in line with those reported by Kim and Liu (2002) [28]. Akalin *et al*. [29] found that probiotic microorganisms reduced pH values of different types of yoghurts from 4.51 to 4.40, after 28 days of refrigerated storage at 4°C.

Microbiology Analysis of Different Produced Functional Whey Beverages During the Storage Period: Fermented lactic beverage can be defined as a type of fermented milk, resultant from the mixture of milk and cheese whey containing lactic culture and other dairy products [5]. Microbiological analysis data presented in Figure 2 (a; b and c) included total bacterial, *L. bulgaricus* and *S. thermophilus* counts (CFU/g) of beverage samples during 28 days of storage period. The mean values obtained with respect to total plate count (TPC) in freshly prepared beverages C; W_1; W_2 and W_3 were (3.43±0.10; 3.45±0.01; 3.15±0.01 and 3.43±0.10 $x10^6$ cfu/mL), respectively. The total plate count was increased during the first two weeks of storage for C; W_1; W_2 and W_3 to (4.90±0.10; 5.55±0.01; 5.75±0.01 and 5.93±0.10$x10^6$cfu/mL), respectively. These increased was accepted because whey was pasteurized before beverage production. Earlier, similar trend for total plate count in whey based fruit beverage was also recorded by Patel *et al*. [30]. They observed a rise in total viable count from 2.6 to 2.76$x10^4$ cfu/mL from initiation to termination of 30 days trial. The current findings of Sakhale *et al*. [31] are analogous to the instant results as they expounded significant increase 1.93 to 2.73$x10^4$ cfu/mL in total plate count of whey based mango beverage after one month storage.

The mean values obtained with respect to *L. delbrueckii* subsp. *bulgaricus* and *S. thermophills* counts in freshly prepared beverages C; W_1; W_2 and W_3 were {(3.20 ± 0.10 and 3.23 ± 0.10); (3.50 ± 0.11 and 3.11 ± 0.10); (3.32 ± 0.01 and 3.12 ± 0.11) and (3.40 ± 0.11 and 3.30 ± 0.01) x 10^4 cfu/mL}, respectively. The *L. delbrueckii* subsp. *bulgaricus* and *S. thermophilus* count was increased during the first two weeks of storage for C; W_1;

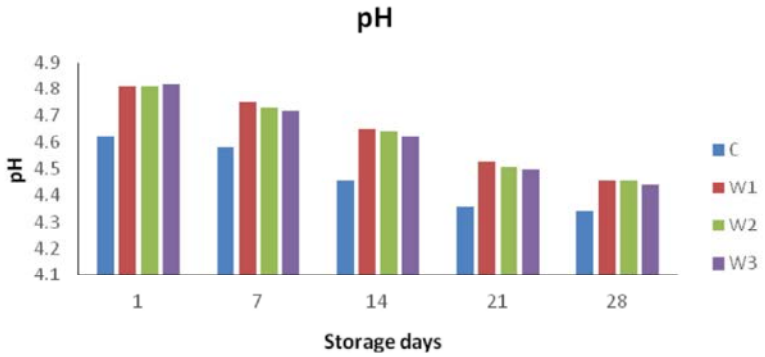

Fig. 1: The changes of pH of different produced functional whey beverages during the storage period. C: control, W_1: 5% Jerusalem artichoke tuber W_2: 5% Globe artichoke roots, W_3: 2.5% Jerusalem artichoke tuber + 2.5% Globe artichoke roots

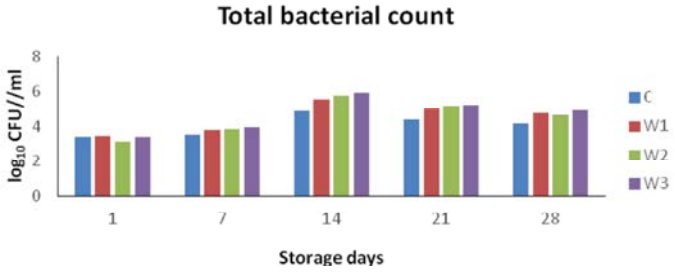

Fig. 2a: Growth of Total bacterial count in different produced functional whey beverage samples during the storage period. C: control, W_1: 5% Jerusalem artichoke tuber W_2: 5% Globe artichoke roots, W_3: 2.5% Jerusalem artichoke tuber + 2.5% Globe artichoke roots

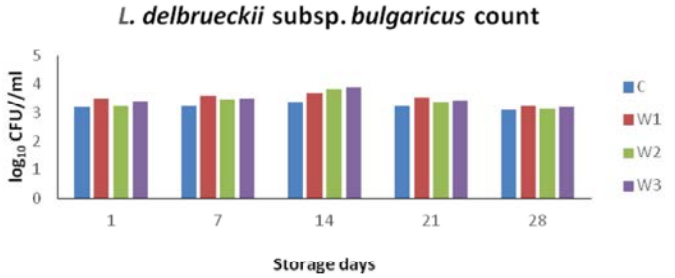

Fig. 2b: Growth of *L. delbrueckii* subsp. *bulgaricus* count in of different produced functional whey beverage samples during the storage period. C: control, W_1: 5% Jerusalem artichoke tuber W_2: 5% Globe artichoke roots, W_3: 2.5% Jerusalem artichoke tuber + 2.5% Globe artichoke roots

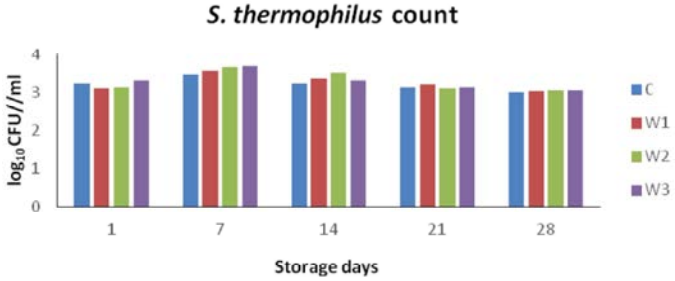

Fig. 2c: Growth of *S. thermophilus* count in of different produced functional whey beverage samples during the storage period. C: control, W_1: 5% Jerusalem artichoke tuber W_2: 5% Globe artichoke roots, W_3: 2.5% Jerusalem artichoke tuber + 2.5% Globe artichoke roots

Table 4: Sensory properties of different produced functional whey beverage samples:

Sample	Colour Fresh 28 days	Taste Fresh 28 days	Flavour Fresh 28 days	Texture Fresh 28 days	Overall acceptability Fresh 28 days
C	7.8± 0.2×10^{8a} 7.90.1×10^{8d}	8.2±0.01×10^{8c}8.0±0.2×10^{8d}	8.0±0.1×10^{8b}7.9±0.2×10^{8a}	8.2±0.1×10^{8a}8.1± 0.3×10^{8c}	8.4 ± 0.2×10^{8b}8.1 0.3×10^{8a}
W_1	7.9±0.2×10^{8b} 7.9 0.1×10^{8a}	8.5±0.2×10^{8c} 8.1± 0.1×10^{8b}	8.2±0.2×10^{8c}7.9±0.1×10^{8d}	8.4±0.2×10^{8b}8.3±0.1×10^{8d}	8.5±0.2×10^{8a}8.2 ± 0.1×10^{8c}
W_2	7.7±0.2×10^{8b} 7.9 ±0.1×10^{8c}	8.1±0.2×10^{8b} 7.9± 0.1×10^{8d}	8.1±0.2×10^{8a}7.9± 0.1×10^{8c}	8.3±0.2×10^{8c}8.0±0.1×10^{8b}	8.2±0.2 ×10^{9d}8.1 ± 0.1×10^{8b}
W_3	7.6± 0.2×10^{8d}7.9 ±0.1×10^{8a}	7.9± 0.2×10^{8a}7.8± 0.1×10^{8a}	8.1±0.2×10^{8a}7.9±0.1×10^{8b}	8.2±0.2×10^{8c}8.1±0.1×10^{8b}	8.3±0.2×10^{8c} 8.0 ± 0.1×10^{8d}

Means that bearing different superscripts are significantly at (P<0.05)

C: control, W1: 5% Jerusalem artichoke tuber W2: 5% Globe artichoke roots, W3: 2.5% Jerusalem artichoke tuber + 2.5% Globe artichoke roots

W_2 and W_3 to {(3.11 ± 0.01 and 3.00 ± 0.11); (3.24 ± 0.11 and 3.02 ± 0.10); (3.15 ± 0.10 and 3.05 ± 0.11) and (3.20 ± 0.11 and 3.06 ±0.10) x10^{4}cfu/mL}, respectively. Vinderola *et al.* [32] reported that counts of probiotic strains decreased during storage that is convergent with observations in this work. Dave and Shah [33] researched the viability of bacteria from commercial starter cultures during yoghurt manufacture and storage. Moreover, several studies have shown that yoghurt bacteria survive well in yoghurt throughout the shelf life [34].

Mould and yeasts were not detected in any beverage samples during the storage period. Also the coliform bacteria were not detected in both fresh and till the end of the storage period in all treatments. This might be due to the severity of heat treatments of whey and the role of lactic acid bacteria in the preservation of the products.

Sensory Properties of Different Produced Functional Whey Beverage Samples: Data presented in Table 4 illustrated the sensory properties of different produced functional whey beverage samples during storage period. The data revealed that, the sensory properties such as colour, taste, flavour, texture and overall acceptability of whey beverage were affected by addition of Jerusalem artichoke tuber and Globe artichoke roots and during storage periods.

Addition of 5% Jerusalem artichoke tuber (W_1) had more acceptable than that of control beverage, while the sensory properties of beverages when addition of 2.5% Jerusalem artichoke tuber + 2.5% Globe artichoke roots (W_3) fortification decreased. Fortification with (W_1) had more acceptable than (W_2).

In general, the presence of Jerusalem artichoke tuber and Globe artichoke roots increased the nutritional quality of beverages. Inclusion of prebiotics in food is a natural way to provide healthy ingredients to the consumers. Most of the prebiotics were easily consumable and give desired functionality to the food items. Considering the results of sensory response, it is deduced that overall acceptability of the functional drinks was maintained best up to 28 days of storage. The results of present study are also in line with the work of Yadav *et al*. [35], they elucidated that appearance, taste, colour, flavour and overall acceptability of whey based banana herbal beverage decreased significantly during 20 days storage at refrigeration temperature. Additionally, Sakhale *et al*. [31] also revealed a significant declining trend in the sensory attributes of whey based mango beverage during 30 days of storage.

CONCLUSIONS

Jerusalem artichoke tubers and Globe artichoke roots have important nutritional values due to its particularly high content of bioactive phenolic and flavonoids compounds, but also due to substantial amounts of Vitamins, inulin, fibres and minerals. From these results, it could be concluded that the use of Jerusalem artichoke tubers and Globe artichoke roots at 5% by-functional whey beverages improved phenolic and flavonoid compounds, vitamin B complex and C, minerals and counts of total bacterial, *L. bulgaricus* and *S. thermophilus*. However, despite of all difficulties, fresh whey processing has proved to be the most economical technological solution. Shelf-life study revealed that during 28 days storage at 4°C, pH of functional whey beverage samples remained above 4, while numbers of *S. thermophilus* and *L. delbrueckii* subsp. *bulgaricus* count remained above 8 log cfu/ml This study showed a new possibility to make an acceptable fermented product based mainly on Jerusalem artichoke tuber or Globe artichoke roots which are suitable substrates that's can support high cell viability during cold storage for 28 days for different probiotic strains. Therefore many efforts have been made in development of beverages with addition of Jerusalem artichoke tuber and Globe artichoke roots in order to produce a drink with acceptable sensory properties especially regarding flavour.

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