Journal of Dairy Research

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Quality and antioxidant potential of goat's milk paneer prepared from different citrus juices and its whey

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Research Article

Cite this article: Qureshi TM, Saeed MM, Nadeem M, Muhammad G, Murtaza MA and Ibrahim SA (2024). Quality and antioxidant potential of goat's milk paneer prepared from different citrus juices and its whey. *Journal of Dairy Research* **91**, 99–107. https://doi.org/10.1017/S0022029924000190

Received: 26 July 2023
Revised: 28 January 2024
Accepted: 6 February 2024
First published online: 16 April 2024

Keywords:
Antioxidant potential; citrus fruit juices; paneer; physico-chemical characteristics; sensorial quality

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Abstract

The experiments reported in this research paper aimed to evaluate the physico-chemical and sensory characteristics, microbial quality and antioxidant potential of goat's milk paneer during storage (0–12 d, 4 ± 1°C). The juices from five different citrus fruits were used as coagulant (treatments) to make goat's milk paneer. The pH of all paneer samples decreased during storage whereas titratable acidity increased. Ash (%) fat (%) and protein (%) of paneer increased slightly during storage, whereas sensory perception decreased. The juices from all the citrus fruit varieties showed high contents of total phenolics and total flavonoids which ultimately influenced ferric reducing antioxidant power, total antioxidant capacity and radical scavenging activities. As the contents of different juices were also retained in the paneer matrix, so paneer coagulated with citrus juices also showed encouraging results in terms of total phenolic and flavonoid contents, ferric reducing antioxidant power and radical scavenging activities. Amongst all the paneers, the most promising was that coagulated by kinnow juice. In addition, the whey obtained from paneer coagulated by citrus juices also showed appreciable quantities of total phenolic and flavonoid contents, thereby beneficially influencing ferric reducing antioxidant power andradical scavenging activities. It is concluded that citrus juices improve the sensorial quality and antioxidant potential of goat's milk paneer and its whey.

Goat's milk has different composition compared to the more commonly consumed cow's milk. It has potentially better digestibility, higher buffering capacity and lower contents of αs_1-casein (which may be responsible for causing allergic reactions) than cow's milk (Biadala and Konieczny, 2018). Goat's milk is also reported to contain more free amino acids and have higher antioxidant potential than cow's milk (Biadala and Konieczny, 2018). Simos *et al.* (2011) also investigated the antioxidant activity of goat, donkey and cow's milk and concluded that goat's milk provided the best antioxidant capacity. Moreover, its casein precipitates more readily into more fine particles as compared to cow's milk (Ulusoy, 2015). Many potential health benefits, like for instance anti-diabetic, antihypertensive, antioxidant, anti-microbial and anti-inflammatory activities, have been attributed to goat's milk (Nayik *et al.*, 2021).

Goat's milk can be converted into many products, such as cheese, yogurt, milk powder, ice-cream, butter and fermented milk products (Ribeiro and Ribeiro, 2010; Nayik *et al.*, 2021) and could potentially have use in the production of infant formula (Prosser, 2020; Vázquez-Garcíaet *al.* (2021) analyzed antioxidant peptides in Mexican goat cheese fractions (acid-soluble and non-protein nitrogen) during ripening. They suggested that the ripened Mexican goat cheese showed DPPH radical scavenging activity owing to the presence of peptides which were either present originally in the milk or released by the actions of starter culture during ripening. Another study also reported that goat cheese had high antioxidant activities due to peptides released by the action of rennet during cheese manufacturing (Hernández-Galán *et al.*, 2017). Further, López-Villafaña *et al.* (2023) also observed appreciable DPPH radical scavenging activity (%) of Mexican Panela goat's milk cheese.

Citrus fruit crops are being cultivated across the globe due to the economic and nutritional values of their fruits (Addi *et al.*, 2022).The juices from different citrus fruits were observed to contain antioxidants and other functional compounds. For example, lemon (*Citrus limon*) juice has been characterized for its composition by Alfadul and Hassan (2016) who found appreciable quantities of vitamin C (23.9 mg/100 g juice) and minerals (Ca, Mg, P, K, Na). Its juice was reported to contain tannins, flavonoids, terpenoids and phenols (Rauf *et al.*, 2014). Being a rich source of antioxidants, lemon juice has been found to have protective

and antioxidant features concerning improved hematological and biochemical parameters of oxidatively stressed mice (Ali *et al.*, 2020). Grapefruit (*Citrus* × *paradisi*) juice was found to contain appreciable quantities of flavanones such as naringin and narirutin (Sicari *et al.*, 2018). Sweet lime (*Citrus limetta*) is commonly known as 'Mosambi' in the culture of the Indian subcontinent region. This fruit has been reported to have multiple traditional uses such as in the treatment of scurvy, constipation, indigestion, ulcers, diabetes and urinary disorder (Khan *et al.*, 2016). The mandarin variety 'Feutrell's Early' (*Citrus reticulata* Blanco) is a delicious citrus fruit in terms of its sweet taste and has been reported to contain appreciable quantities of vitamin c, DPPH radical scavenging activities, total phenolics, flavonoids, flavonols and carotenoids (Nawaz *et al.*, 2020; Manzoor *et al.*, 2023). Kinnow (*Citrus reticulate* L.) is a cross between the King Orange (*Citrus nobilis*) and Willow Leaf Orange (*Citrus deliciosa*) which was developed by Dr H.B. Frost in California in 1935 (Sharma *et al.*, 2016). It was brought to the Indian sub-continent and is now grown in Pakistan, India and Bangladesh. The juice of kinnow is rich in vitamin B, vitamin C, β-carotene, minerals, and other bioactive compounds (Mahawar *et al.*, 2020). All the above mentioned citrus fruit varieties are grown in Pakistan, especially in Punjab and are available in abundant quantities during winter season.

Among the dairy products, paneer is considered a primitive cheese variety which is produced through heat and acid coagulation of milk (Qureshi *et al.*, 2019). People in Pakistan do not have a tradition to consume paneer either alone or with bread. In our previous study, traditional spices were incorporated into buffalo milk paneer matrix so as to increase its consumption (Qureshi *et al.*, 2023). Concerning goat's milk, some people avoid it due to its distinct flavor owing to the presence of more free amino acids and higher amounts of short-chain fatty acids (Ulusoy, 2015). In this context, it would be of interest to be able to convert goat's milk into value added product(s) so as to boost its consumption. Synthetic citric acid is usually used for the coagulation of milk in order to produce paneer at sweet shops or bakeries. Citrus fruit juices contain abundant quantities of natural ascorbic acid which may also act as a coagulant in the preparation of paneer. Moreover, citrus fruit juices are potentially health promoting due to the presence of abundant quantities of bioactive compounds. Therefore, in order to avoid deleterious effects of synthetic acids (for coagulation) as well as to enhance the nutrition and taste of goat's milk paneer, it would be advantageous to coagulate goat's milk with natural citrus fruit juices. To our knowledge, no study has been previously published regarding the quality and antioxidant potential of goat's milk paneer coagulated with juices from citrus varieties. Thus, the objective of the present study was to explore the physico-chemical characteristics, microbiological quality, sensorial acceptability and antioxidant potential of goat's milk paneer prepared through coagulation by the juices from different citrus varieties.

Materials and methods

Procurement of raw materials for development of paneer

Goat's milk used for paneer manufacturing was collected from a goat farm in a village of Bahawalpur city near Cholistan University of Veterinary and Animal Sciences, Bahawalpur. It was filtered using muslin cloth before being standardized: fresh goat's milk was separated to obtain skim milk and cream, which were then used to standardize the milk to 2.5% fat (7.5% solids-not-fat) before paneer manufacture. Different citrus fruits were purchased from the local fruit market.

Chemicals and apparatus

DPPH (2, 2-diphenyl-1-picrylhydrazyl), ascorbic acid, quercetin, Folin-Ciocalteu reagent and various other chemicals/standards were purchased from Merck (KgaA, Darmstadt, Germany). All other chemicals were also of analytical grade. All the apparatus and glassware was properly cleaned and manufacturing was done under hygienic condition in the laboratory. For the packaging of paneer, plastic sheet and aluminum foil were also bought from the local market.

Treatment plan and manufacturing

The citrus fruit juices and milk were used for the manufacturing of paneer. The six different paneers were made with citric acid (control), lemon juice, grapefruit juice, musambi juice, mandarin juice (Feutrell's Early) and kinnow juice. The pH and total soluble solids (°Brix) of the juices were determined previously, and in our preliminary trials the quantities of the different citrus juices required for coagulation were noted. These formed the basis of the paneer preparation, as detailed in online Supplementary Table S1. The pH of the whole paneer mixture was adjusted to 5.70–5.80 because proper coagulation occurred at that pH, and at that point addition of coagulants was stopped so that the coagulum attained should not be hard.

The standardized milk was transferred to a stainless steel container and heated for 5 min at 85°C, then cooled to 75°C with continual stirring before coagulation with different coagulants. The whey was then drained out by pouring it into the muslin fabric, and the resulting curd was placed in a hoop lined with the muslin cloth. Then, the paneer of each treatment was separately pressed for 30 min, yielding a small block of paneer. The paneer block was cut into five equal pieces. For each treatment, the same method was implemented. The cut paneer pieces were then carefully wrapped, first in a plastic sheet to prevent moisture loss and contamination, and then in aluminum foil. The wrapped pieces were stored at refrigeration temperature (4°C) for 3, 6, 9 or 12 d. One piece from each treatment was immediately frozen as fresh (day 0).

Physico-chemical analysis

Physico-chemical analysis of paneer was carried out during storage period. Gerber method was used to determine the fat in paneer. Moisture contents of paneer were determined according to IDF standard 4/ISO 5534 (IDF, 2004) through oven drying. After calibrating with buffers of pH 4.0 and pH 7.0, the pH of paneer was measured using a pH meter. With a few drops of water, the electrode was placed in grated paneer (20 g) (Ardö and Polychroniadou, 1999).Total nitrogen was calculated using the IDF standard 20B (IDF, 1993). Protein contents (%) were determined by multiplying TN (%) by 6.38. The titratable acidity was measured according to the method described in AOAC (2016).

Microbiological analysis

Paneer was microbiologically analyzed for total plate counts (TPC, log CFU/g of paneer), yeasts and molds (YMC, log CFU/

https://doi.org/10.1017/S0022029924000190 Published online by Cambridge University Press

g of paneer) during storage period as described by Broadbent *et al.* (2013).

Sensory evaluation

Paneer was evaluated for sensorial quality using a 9 point hedonic scale (1 = dislike extremely, 2 = dislike very much, 3 = dislike moderately, 4 = dislike slightly 5 = neither like nor dislike, 6 = like slightly, 7 = like moderately, 8 = like very much, 9 = like extremely) for appearance and color, taste, texture and overall acceptability (Dongare and Syed, 2018). The sensory characteristics of prepared paneer samples were assessed by 20 trained assessors including graduates and faculty from the Department of Food Sciences at our university who know about the scaling procedures of paneer samples.

Other measurements

Water soluble extracts (WSEs) of paneer were prepared according to the procedure described by Gupta *et al.* (2013) with a few modifications, and used for further analysis. The total phenolic content (TP) of paneer or juices was determined using the method described by Reis *et al.* (2012). The TP contents were calculated as µg gallic acid equivalent (GAE)/g of paneer or µg GAE/ml of juice. Total flavonoid (TF) contents were measured *via* the spectrophotometric method according to Jia *et al.* (1999). The results were articulated as µg quercetin equivalent (QE)/g of paneer or µg QE/ml of juice. All determinations were carried out in triplicate. The ferric reducing antioxidant power (FRAP) was computed as described by Reis *et al.* (2012) with slight modifications. The results were calculated as µg ascorbic acid equivalent (AAE)/g or µg AAE/ml.

Radical scavenging activities were determined in two assays. The capability of samples to scavenge 2,2′-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS) was determined following the method of Zeghad *et al.* (2019). The ABTS radical scavenging activity was calculated as mg ascorbic acid equivalent (AAE)/100 g or mg AAE/100 ml. The potential for WSE of paneer or juices to scavenge 2,2-diphenyl-1-picrylhydrazyl radical (DPPH) was measured according to the method of Yi *et al.* (2008) with some modifications. The DPPH radical scavenging activity was calculated as mg ascorbic acid equivalent (AAE)/100 g or mg AAE/100 ml. Finally, the total antioxidant capacity (TAC) was determined as described by Prieto *et al.* (1999). The TAC was calculated as mg ascorbic acid equivalent (AAE)/100 g or mg AAE/100 mL.

Statistical analysis

Statistical analysis was performed using Minitab statistical software version 16 (Minitab Inc., State College, PA, USA) by employing two-way ANOVA and Tukey's test for pair wise comparison at a level of $P < 0.05$.

Results

pH and total soluble solids (°Brix) of coagulants

Online Supplementary Fig. S1 shows pH and °Brix of coagulants used in the preparation of goat's milk paneer. Among the coagulants, lemon juice showed the minimum pH (2.42) whereas the maximum pH was shown by Musambi juice (4.23).The citric acid solution (2%) showed higher pH values (2.92) than lemon juice but lower values compared to juices from other citrus varieties (3.43–4.23). The minimum quantity of lemon juice was used in the development of goat's milk paneer followed by citric acid solution. The maximum quantity of musambi juice was used in the preparation of goat's milk paneer. Among the natural coagulants used in the present study, lemon juice showed the minimum °Brix (8.21) whereas the maximum °Brix was shown by kinnow juice (10.94). The citric acid solution (2%) showed the minimum °Brix (1.52).

Physico-chemical characteristics of goat's milk paneer

Data regarding the physico-chemical characteristics of the goat's milk paneers are shown in Table 1. All the treatments showed significant decreasing trend of pH during storage. The pH was slightly decreased after successive storage period. There was slight variation in acidity between each treatment. All fresh paneer in the present study showed pH values in the range 5.79–5.87. All the treatments showed significant increasing trend of acidity (%) during storage. The acidity was slightly increased after successive storage period. There was slight variation in acidity between each treatment. All fresh paneer in the present study showed acidity values more or less 0.30%. Even though, there was significant effect of storage on the moisture contents of paneer but that decrement was slight after successive storage period. The decreasing trend of moisture during storage thereby led to gradual increase in dry matter of paneer with the passage of time. All fresh paneer in the present study showed more than 53% moisture contents. All the treatments showed significant increasing trend of protein (%) contents during storage. The protein contents were slightly increased after successive storage period. There was slight variation in protein contents between each treatment. All fresh paneer in the present study showed fat contents in the range 17–19%. The fat contents were slightly increased after subsequent storage period. There was slight variation in the fat contents between each treatment. All fresh paneer in the present study showed fat content in the range 18–20%. All the treatments showed very slight increasing (non-significant) trend of ash (%) contents during storage. There was slight variation in ash contents between each treatment. All fresh paneer in the present study showed ash contents values more or less 2.0%.

Microbiological quality of goat's milk paneer

Microbial count data are given in Fig. 1. The TPC values of freshly prepared control paneer were higher (2.40 $\log_{10}$ CFU/g) than all other paneer prepared through coagulation of citrus juices (except musambi juice paneer). Generally, it was observed that the TPC values of all treatments varied significantly during storage. At the end of the storage period, most of the treatments showed TPC values in the range 5.61–5.81 $\log_{10}$ CFU/g. A similar trend was also seen regarding yeast and mold counts (YMC: Fig. 1). YMC of freshly prepared control paneer were 1.45 ($\log_{10}$ CFU/g) whereas all other fresh paneer coagulated by citrus juices showed slightly lower values. The values also increased significantly during storage in all the treatments and reached up to the range of 4.46–4.86 $\log_{10}$ CFU/g. All the goat's milk paneer prepared using different coagulants showed higher TPC values than the counts of Y & M. It was found that

https://doi.org/10.1017/S0022029924000190 Published online by Cambridge University Press

Table 1. Proximal characteristics of goat's milk paneers prepared through coagulation by different citrus juices, during storage

Treatments	Days	pH	Acidity (%)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
Control paneer	0	5.87 ± 0.3a	0.35 ± 0.03p–s	55.33 ± 0.15a	17.86 ± 0.15j	18.76 ± 0.15j	2.01 ± 0.10j
	3	5.68 ± 0.1c–h	0.41 ± 0.03k–p	53.93 ± 0.33abc	18.42 ± 0.25 h–j	19.35 ± 0.15 h–j	2.07 ± 0.10hij
	6	5.57 ± 0.1 g–i	0.47 ± 0.03f–k	53.96 ± 0.33abc	18.41 ± 0.35 h–j	19.33 ± 0.25 h–j	2.07 ± 0.20hij
	9	5.52 ± 0.1hi	0.53 ± 0.03a–f	52.26 ± 0.23c–j	19.09 ± 0.21a–h	20.05 ± 0.31a–h	2.15 ± 0.20a–h
	12	5.43 ± 0.1i	0.59 ± 0.03a	52.33 ± 0.33c–j	19.06 ± 0.31a–h	20.02 ± 1.35a–h	2.14 ± 0.30a–h
Lemon juice paneer	0	5.79 ± 0.3a–e	0.33 ± 0.03q–s	53.80 ± 0.14a–d	18.48 ± 0.35 g–j	19.40 ± 0.15 g–j	2.08 ± 0.10 g–j
	3	5.73 ± 0.3a–g	0.39 ± 0.03 l–q	52.80 ± 0.23c–h	18.88 ± 0.15c–h	19.82 ± 0.11c–h	2.12 ± 0.10c–h
	6	5.65 ± 0.2d–h	0.45 ± 0.03 g–l	52.06 ± 0.43c–j	19.17 ± 0.15a–h	20.13 ± 0.21a–h	2.15 ± 0.20a–h
	9	5.62 ± 0.2e–h	0.51 ± 0.03b–g	51.33 ± 0.13f–j	19.46 ± 0.25a–e	20.44 ± 1.25a–e	2.19 ± 0.30a–e
	12	5.56 ± 0.1 g–i	0.57 ± 0.03ab	50.36 ± 0.43j	19.85 ± 0.21a	20.84 ± 1.35a	2.23 ± 0.30a
Grapefruit juice paneer	0	5.81 ± 0.3a–d	0.32 ± 0.03rs	53.73 ± 0.14a–e	18.50 ± 0.25f–j	19.43 ± 0.21f–j	2.08 ± 0.10f–j
	3	5.76 ± 0.3a–f	0.38 ± 0.03 m–r	52.63 ± 0.23c–i	18.94 ± 0.35b–h	19.89 ± 0.21b–h	2.13 ± 0.20b–h
	6	5.70 ± 0.3b–g	0.44 ± 0.03 h–m	51.56 ± 0.13d–j	19.37 ± 0.31a–g	20.34 ± 0.21a–g	2.18 ± 0.15a–g
	9	5.68 ± 0.32c–h	0.50 ± 0.04c–h	51.50 ± 0.23e–j	19.40 ± 0.31a–f	20.37 ± 1.35a–f	2.18 ± 0.12a–g
	12	5.60 ± 0.1f–i	0.56 ± 0.04a–c	50.76 ± 0.33 h–j	19.69 ± 0.31a–c	20.67 ± 0.32a–c	2.21 ± 0.25a–c
Musambi juice paneer	0	5.79 ± 0.3a–e	0.32 ± 0.03rs	53.30 ± 0.24a–g	18.68 ± 0.15d–j	19.61 ± 0.21d–j	2.10 ± 0.10d–j
	3	5.70 ± 0.3b–g	0.37 ± 0.03n–s	52.93 ± 0.33b–h	18.82 ± 0.25c–i	19.77 ± 0.21c–i	2.12 ± 0.10c–i
	6	5.703 ± 0.3b–g	0.43 ± 0.03i–n	52.23 ± 0.14c–j	19.10 ± 0.25a–h	20.06 ± 0.21a–h	2.15 ± 0.20a–h
	9	5.73 ± 0.2a–g	0.49 ± 0.04d–i	51.03 ± 0.33 h–j	19.58 ± 0.25a–c	20.56 ± 0.21a–c	2.20 ± 0.10a–d
	12	5.69 ± 0.1b–h	0.55 ± 0.04a–d	50.46 ± 0.23ij	19.81 ± 0.21ab	20.80 ± 0.32ab	2.23 ± 0.30ab
Treatments	Days	pH	Acidity (%)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
Feutrell juice paneer	0	5.86 ± 0.3ab	0.31 ± 0.03s	54.06 ± 0.34a–c	18.37 ± 0.25 h–j	19.29 ± 0.21 h–j	2.06 ± 0.10hij
	3	5.82 ± 0.3a–c	0.36 ± 0.03o–s	52.46 ± 0.43c–j	19.01 ± 0.21a–h	19.96 ± 0.11a–h	2.14 ± 0.13a–h
	6	5.77 ± 0.3a–e	0.42 ± 0.03j–o	51.16 ± 0.23 g–j	19.53 ± 0.31a–d	20.51 ± 0.31a–d	2.20 ± 0.10a–d
	9	5.73 ± 0.2a–g	0.48 ± 0.04e–j	50.46 ± 0.43 lj	19.81 ± 0.21ab	20.80 ± 0.31ab	2.23 ± 0.20ab
	12	5.71 ± 0.1a–g	0.54 ± 0.04a–e	50.96 ± 0.13 h–j	19.61 ± 0.41a–c	20.59 ± 0.22a–c	2.20 ± 0.20a–c
Kinnow juice paneer	0	5.84 ± 0.3a–c	0.32 ± 0.03rs	55.06 ± 0.33ab	17.97 ± 0.25ij	18.87 ± 0.11ij	2.02 ± 0.10ij
	3	5.80 ± 0.3a–d	0.37 ± 0.03n–s	53.46 ± 0.33a–f	18.61 ± 0.25e–j	19.54 ± 0.11e–j	2.09 ± 0.10e–j
	6	5.75 ± 0.2a–f	0.43 ± 0.03i–n	52.16 ± 0.13c–j	19.13 ± 0.21a–h	20.09 ± 0.31a–h	2.15 ± 0.20a–h
	9	5.71 ± 0.2a–g	0.49 ± 0.04d–i	51.46 ± 0.33f–j	19.41 ± 0.35a–e	20.38 ± 0.21a–e	2.18 ± 0.17a–f
	12	5.69 ± 0.2c–h	0.55 ± 0.04a–d	51.96 ± 0.33c–j	19.21 ± 0.11a–h	20.17 ± 1.25a–h	2.16 ± 0.20a–h

Means with different letters in the same column show significant ($P < 0.05$) differences between treatments and storage period.

visible fungus started to appear on the surfaces of paneer stored for more than 15 d.

Sensorial quality of goat's milk paneer

Sensory scores are given in Fig. 2 for appearance and color, flavor, texture and overall acceptability. Generally, all fresh paneer coagulated using different coagulants had higher sensory scores for all the sensory attributes. The paneer prepared using different citrus juices had higher sensory scores than control paneer at each storage period. Among all the treatments, the paneer prepared using mandarin juice gave the highest score for all sensory attributes. The paneer stored for 12 d was liked the least by the judges due to its slightly deteriorated quality.

Antioxidant potential of juice from citrus varieties

Online Supplementary Fig. S2 presents the results concerning antioxidant potential of the different goat's milk paneers. The juice from all the citrus varieties had high TP contents with the highest shown by kinnow juice (1556.67 µg GAE/ml) followed by grapefruit juice (1356.55 µg GAE/ml). Regarding TF values, the highest value (566.86 µg QE/ml) was for kinnow juice followed by grapefruit juice (466.36 µg QE/ml). The juices from other citrus varieties also showed appreciable quantities of TF.

https://doi.org/10.1017/S0022029924000190 Published online by Cambridge University Press

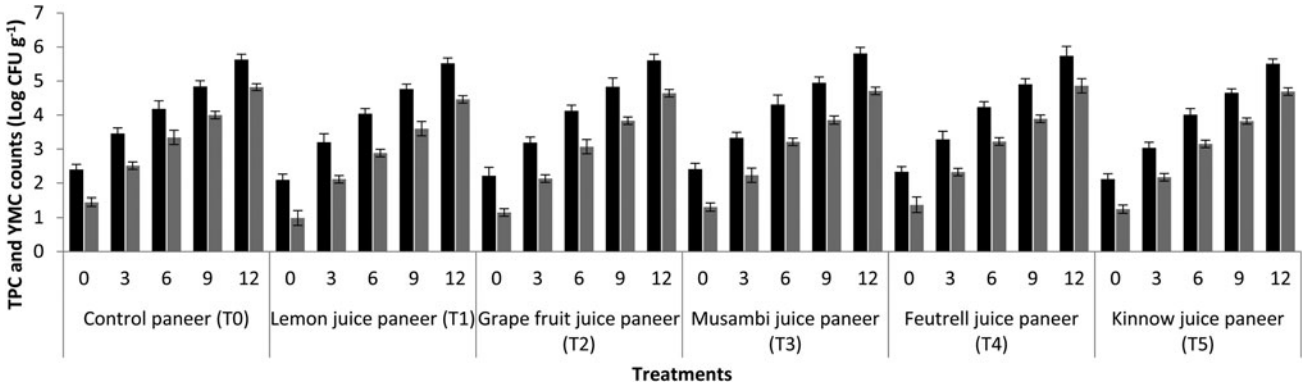

Figure 1. Total plate counts (TPC, means ± SD) (black bars) and yeast and molds (YMC, means ± SD) (gray bars) of goat's milk paneer prepared through coagulation by different citrus juices, during storage at 4°C.

Similarly, the highest value of FRAP was for kinnow juice (1310.09 µg AAE/ml) followed by mandarin (1309.05 µg AAE/ml). The juices from other citrus varieties also showed considerable FRAP values. The highest value of ABTS radical scavenging activity was for kinnow juice (224.33 mg AAE/100 ml) followed by lemon juice (199.80 mg AAE/100 ml). Good values of ABTS radical scavenging activity were also obtained by the juices from other citrus varieties. Regarding DPPH radical scavenging activity, the highest value was for mandarin juice (145.30 mg ascorbic acid equivalent (AAE)/100 ml). Good values of DPPH radical scavenging activity were also obtained by the juices from other citrus varieties. Concerning total antioxidant capacity, the highest value was for Feutrell juice (145.20 mg AAE/100 ml) followed by lemon juice (142.10 mg AAE/100 ml). Good values of TAC were also obtained by the juices from other citrus varieties.

Antioxidant potential of goat's milk paneer

Figure 3 shows the total phenolic and flavonoid contents as well as FRAP values of WSE from goat's milk paneer. The TP value of freshly prepared control paneer showed the lowest value (269.67 µg GAE/g) whereas the highest value (455.34 µg GAE/g) was for fresh paneer prepared from lemon juice. The paneer prepared using other citrus fruit juices also showed good TP contents. The TP values decreased gradually during storage in all the treatments. Regarding TF contents, the highest value was obtained by fresh paneer using kinnow juice (169.33 µg QE/g) followed by respective paneer made by grapefruit juice (144.70 µg QE/g). The lowest contents of TF were shown by the control (citric acid) paneer. The TF values also decreased gradually during storage in all the treatments. The highest value (140.88 µg AAE/g) of FRAP was for fresh paneer coagulated through lemon juice (T_1) whereas the lowest values were for control paneer. In this way, paneer prepared using kinnow juice and lemon juice showed promising results concerning TP and TF contents as well as FRAP values.

Figure 4 shows the ABTS and DPPH radical scavenging activities as well as TAC values of the paneers. In all three measures, the highest value was for fresh paneer prepared using kinnow juice followed by grapefruit juice, whereas the lowest values were shown by the control paneer. All three characteristics decreased gradually during storage in all the treatments. Overall, paneer prepared using kinnow juice and grapefruit juice showed promising results concerning DPPH and ABTS radical scavenging activities as well as TAC values.

Antioxidant potential of whey from goat's milk paneer

Figure 5 shows the results for TP and TF contents, FRAP and TAC values, DPPH and ABTS radical scavenging activity of whey collected during manufacturing of goat's milk paneer. The TP and also TF values of the control (citric acid) whey were the lowest whereas the highest values were obtained by paneer prepared using kinnow juice. The paneer prepared using juices of

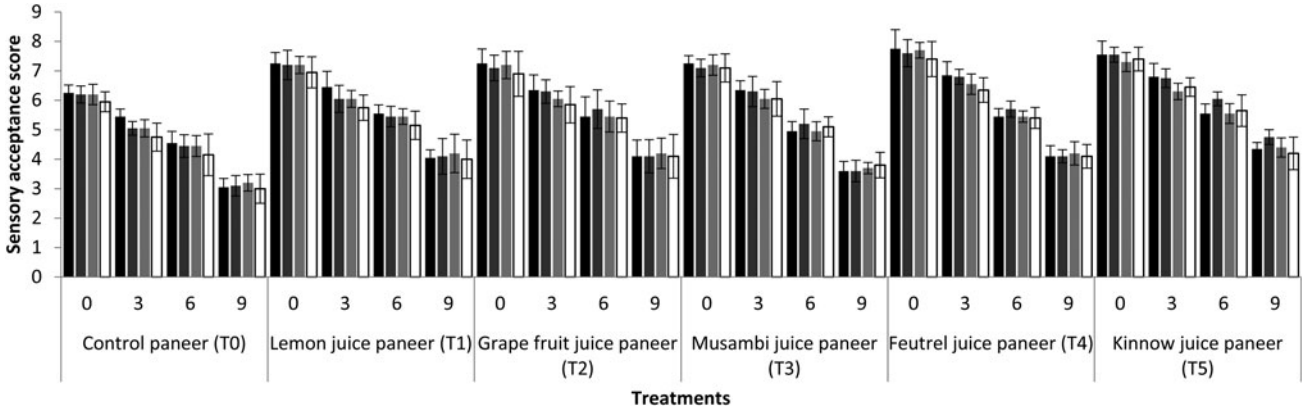

Figure 2. Sensory acceptance scores (means ± SD) regarding appearance and color (black bars), flavor (dark gray bars), texture (light gray bars) and overall acceptability (white bars) of goat's milk paneer prepared through coagulation by different citrus juices, during storage at 4°C.

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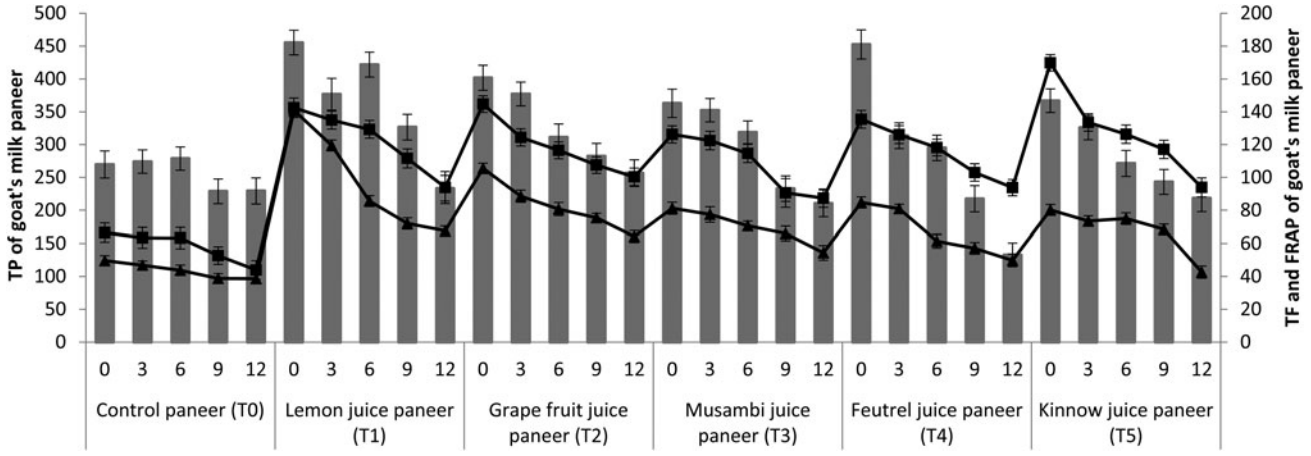

Figure 3. Total phenolics (TP, means ± SD, μg GAE/g) (gray bars), total flavonoids (TF, —■—, means ± SD, μg QE/g) and ferric reducing antioxidant power (FRAP, —▲—, means ± SD, μg AAE/g) (white bars) of goat's milk paneer prepared through coagulation by different citrus juices, during storage at 4°C.

other citrus varieties also showed considerable TP contents. Whey from paneer made with grapefruit juice had the second highest TF value. Similarly, the highest value (17.01 μg AAE/g) of FRAP was obtained by whey from paneer coagulated with kinnow juice whereas the lowest value was shown by whey from control paneer. Whey from paneer prepared using kinnow juice and grapefruit juice showed promising results concerning TP and TF contents as well as FRAP values.

Regarding ABTS and DPPH radical scavenging activities, once again whey from kinnow paneer had the highest value (3.43 and 1.63 mg AAE/100 g, respectively) and that from control paneer had the lowest. The whey collected from other paneer coagulated by different citrus juices also showed promising results for ABTS values. Lemon juice paneer also had a high DPPH activity (1.62 mg AAE/100 g). Not surprisingly the highest value of TAC was for whey from paneer prepared using kinnow juice (22.03 mg AAE/100 g) and the lowest was for control paneer whey. Whey from paneer prepared using kinnow juice showed more promising results concerning DPPH and ABTS radical scavenging activity as well as TAC values.

Discussion

The pH of the juices to be used as coagulants determined the quantity required in the preparation of paneer. The lower the pH, the lesser was the quantity of coagulants used for making paneer with the specified quantity of milk. A lower pH value of any coagulant might be due to presence of more citric acid. The musambi juice probably contained least natural citric acid because the maximum quantity of musambi juice was used in the preparation of paneer (700 ml *vs.*, for example, 65 ml of lemon juice: online Supplementary Table S1). Our pH values were similar to published reports (grapefruit juice: Ahmed *et al.* (2019); lemon juice:

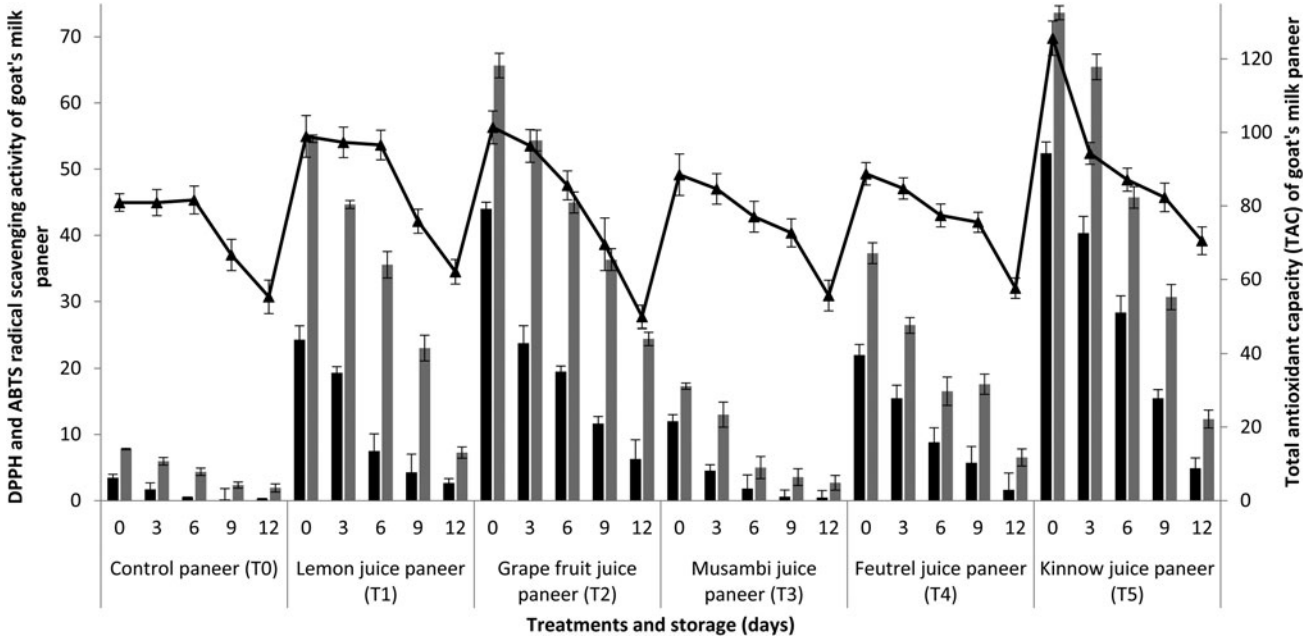

Figure 4. DPPH radical scavenging activity (means ± SD, mg AAE/100 g) (black bars), ABTS radical scavenging activity (means ± SD, mg AAE/100 g) (gray bars), and total antioxidant capacity (TAC) (—▲—, means ± SD, mg AAE/100 g) of goat's milk paneer prepared through coagulation by different citrus juices, during storage at 4°C.

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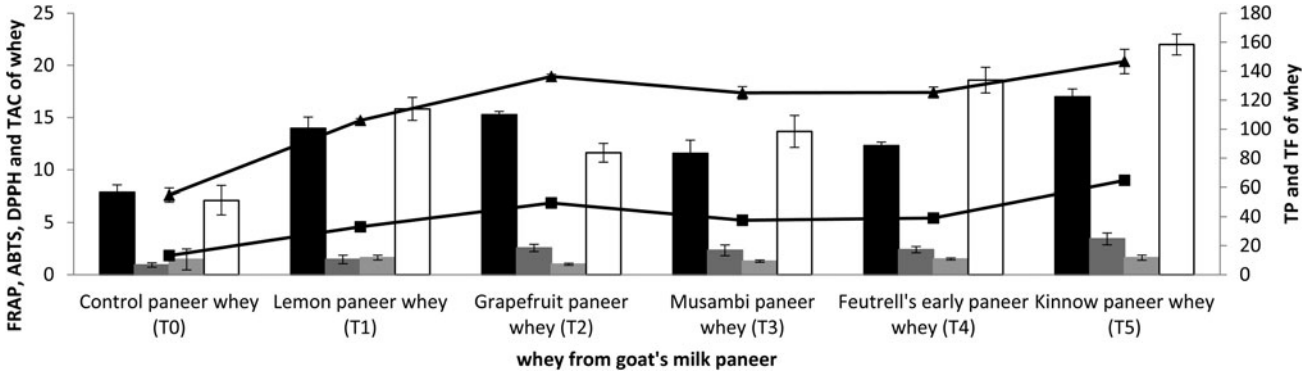

Figure 5. Total phenolics (TP, —■—, means ± SD, µg GAE/ml)), total flavonoids (TF, —▲—, means ± SD, µg QE/ml), ferric reducing antioxidant power (FRAP, means ± SD, µg AAE/ml) (black bars), ABTS radical scavenging activity (means ± SD, mg AAE/100 ml) (dark gray bars), DPPH radical scavenging activity (means ± SD, mg AAE/100 ml) (light gray bars) and total antioxidant capacity (TAC) (means ± SD, mg AAE/100 ml) (white bars) of whey from goat's milk paneer prepared through coagulation by different citrus juices.

Al-Juhaimi and Ghafoor (2013); mandarin juice: Manzoor *et al.* (2023) and Nawaz *et al.* (2020)). During our paneer manufacturing, the pH of the whole paneer mixture was adjusted to 5.70–5.80 (coagulation point) irrespective of which juice was being used. This is why different quantities of the citrus juices were used, and meant acidity values in the different fresh paneer matrices were all more or less same. Badola *et al.* (2018) observed a pH of around 5.65 at the time of milk coagulation during manufacturing of paneer which is a little bit lower than we used.

The °Brix value equates to the total soluble solids of the juice. The freshly squeezed natural juices used here would have contained sugars along with other compounds, including citric acid and other organic acids, whilst the °Brix of citric acid solution was due only to citric acid itself. As for pH, our °Brix values were similar to published data (lemon juice: Aguilar-Hernández *et al.* (2020), Al-Juhaimi and Ghafoor (2013) and Pham *et al.* (2020); grapefruit juice: Ahmed *et al.* (2019); kinnow juice: Mahawar *et al.* (2020); mandarin juice: Manzoor *et al.* (2023) and Nawaz *et al.* (2020)).

The decreasing trend of pH of paneer with storage time is in accordance with the results of Shanaziya *et al.* (2018) and Singh *et al.* (2014). This might be due to the activities of indigenous microorganisms in paneer, causing more accumulation of acids during the storage period. There was some loss of moisture contents during storage of paneer, thereby increasing the dry matter content. Again, our data are consistent with published reports (Khatkar *et al.*, 2017). As dry matter of paneer matrix increased during storage, then it positively influenced the protein, fat and ash contents.

The presence of TPC and YMC of the freshly prepared paneers might be due to presence of microbiota in goat's milk. Moreover, microbes may reinfect paneer during post manufacture conditions of handling, packaging and storage. Moreover, no starter or probiotic culture is used in the manufacturing of paneer, and it is recognized that such cultures can inhibit the proliferation of harmful microbes. For this reason, the shelf life of paneer is very low as compared to cheese. The increasing trend of TPC and YMC during storage of paneer as observed in the present study is consistent with the findings of some previous studies (Singh *et al.*, 2014; Mishra *et al.*, 2016), whilst the lower TPC and YMC counts in fresh paneer using citrus juices than in control paneer might be due to antimicrobial factors other than acidity. In this respect, lemon juice gave the best outcome.

The sensory attributes usually illustrate the quality characteristics of prepared paneer. López-Villafaña *et al.* (2023) observed sensory score values above 6.4 for freshly produced goat's milk cheese whilst our initial values were generally above this. Our observation of decreasing sensory attributes during storage agrees with earlier reports (Singh *et al.*, 2014 and Das *et al.*, 2018). The microbial counts of paneer increased to an unacceptable limit after 15 d of storage. Therefore, paneer stored for 15 d were not included in the present study for sensory evaluation.

The juices from different citrus fruits contain antioxidants and other functional compounds. For example, lemon (*Citrus limon*) juice contains appreciable quantities of vitamin C (Alfadul and Hassan, 2016), tannins, flavonoids, terpenoids and phenols (Rauf *et al.*, 2014). Grapefruit (*Citrus* × *paradisi*) juice has appreciable quantities of flavanones such as naringin and narirutin (Sicari *et al.*, 2018). Mandarins such as Feutrell's Early (*Citrus reticulata* Blanco) contains appreciable quantities of vitamin c, DPPH radical scavenging activities, total phenolics, flavonoids, flavonols and carotenoids (Nawaz *et al.*, 2020; Manzoor *et al.*, 2023). Kinnow (*Citrus reticulate* L.) juice is rich in vitamin B, vitamin C, β-carotene, minerals and other bioactive compounds (Mahawar *et al.*, 2020). The promising results of citrus fruit juices concerning TP and TF contents, FRAP values, ABTS and DPPH radical scavenging activity in the present study might be due to the presence of one or more of these compounds. The TP contents of lemon and kinnow juice obtained in the present study were slightly higher than the contents found by Al-Juhaimi and Ghafoor (2013) which might be due to geographical variations, and it should be remembered that the antioxidant activity of each citrus juice itself was many fold higher than the WSE of the goat's milk paneer prepared through coagulation of their respective juice, nevertheless the results of DPPH/ABTS and TAC of the latter were very promising. The high antioxidant potential of the juices might be attributed to the presence of high contents of phenolics as well as flavonoids. So, paneers coagulated by citrus juices may retain phenolics and flavonoids in their matrix. However, other factors might be in play as well. Casein protein fractions were reported to be responsible for the antioxidant potential of goat's milk (Mal *et al.*, 2018), as were free amino acids (Biadala and Konieczny, 2018). Hernández-Galán *et al.* (2017) also reported antioxidant potential of goat's milk owing to the presence of peptides originally present in the milk. In another study, Vázquez-Garcí *et al.* (2021) also related

https://doi.org/10.1017/S0022029924000190 Published online by Cambridge University Press

DPPH radical scavenging activity of Mexican goat cheese to the peptides originally present in goat's milk. Hence, it may be assumed that the antioxidant potential of goat's milk paneer prepared in the present study might be due to presence of different proteins, their peptides and free amino acids, as well as the contribution from the citrus juices.

Most earlier studies reported antioxidant potential of goat's milk cheese but no comprehensive study has previously been done on the antioxidant activity of goat's milk paneer. Goat's milk cheese is usually prepared using specific starter cultures (such as MO-1, LHB02 from Chr. Hansen, Denmark: Tabla et al., 2018) and rennet (50% chymosin and 50% pepsin: Baquero *et al.*, 2011) and it is these that are usually responsible for the release of bioactive peptides during production as well as during ripening. Therefore, the antioxidant potential of cheese would be different from paneer because paneer does not contain such kind of starter cultures and rennet. The total polyphenol content of goat's milk cheese obtained from the pasteurized milk of grazing goats was 300 µg GAE/g (Hilario *et al.*, 2010), similar to our data for paneer coagulated by citrus juices. They observed high concentrations of some phenolic compounds such as caffeic acid, chlorogenic acid and ferulic acid but only minute quantities of the flavonoid compounds, catequine and quercetin. Such bioactive compounds are probably present in paneer, and it may be that the decrease of antioxidant potential during storage might be due to degradation of such compounds in the paneer matrix. Most of the phenolics and flavonoids would be expected to leach into the whey of the paneer, but the higher antioxidant potential of the paneer itself (as opposed to the whey) may be expected due to the presence of casein and its fractions (peptides, amino acids) in the paneer matrix. Whey is the by-product of cheese and paneer and can be easily digested and absorbed. It contains vitamin-binding proteins and bioactive proteins, such as beta-lactoglobulin, alpha-lactalbumin, immunoglobulin, serum albumin, and lactoferrin. These components may exhibit a range of immune enhancing and antioxidant activities (Tsutsumi and Tsutsumi, 2013). Dryáková *et al.* (2010) and Mann *et al.* (2015) also reported that whey protein hydrolysates were responsible for DPPH radical scavenging activity. The whey from all the paneer (except the control paneer) had a good taste presumably due to the presence of more quantities of fruit sugars in addition to lactose. Presence of vitamin C, sugars and bioactive compounds confer improved nutritional value to such whey, which could potentially be converted into valuable infant formulations or drinks. In this respect, preparation of functional milk beverages from whey has also been reported by Mann *et al.* (2015).

In conclusion, all the paneers prepared using different citrus juices for coagulation were acceptable concerning sensory attributes. The results of all the antioxidant potential analyses were promising. Citrus fruit juice paneers had greater antioxidant potential than the control paneer, presumably due to the presence of phenolics, flavonoids and other bioactive compounds. Moreover, paneer coagulated by kinnow juice showed the maximum values regarding TF contents, TAC values and DPPH/ABTS radical scavenging activities. It may be recommended that citrus juice paneer as well as its whey should be produced by the dairy industry.

Supplementary material. The supplementary material for this article can be found at https://doi.org/10.1017/S0022029924000190

Acknowledgments. The authors acknowledge the financial support from higher education commission (HEC), Pakistan (under the project No. 10383) for carrying out research activities. This publication was made possible by grant number NC.X-267-5-12-170-1 from the National Institute of Food and Agriculture (NIFA) and the Department of Family and Consumer Sciences and the Agriculture Research Station at North Carolina Agriculture and Technical State University (Greensboro, NC, USA 27411). This work was also supported, in part, by 1890 Capacity Building Program grant no. (2020-38821-31113/project accession no. 021765).

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