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Microbial, physico-chemical and sensory characteristics of mango juice enriched probiotic dairy drinks

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ABSTRACT

This study aimed to determine whether mango juice can improve the viability of probiotics in a fermented dairy-based beverage whilst maintaining its quality characteristics. Formulations containing *Lactobacillus acidophilus* La-5 culture, whole cow's milk and varying concentrations of mango juice (0%, 10%, 20%, 30% and 40% (w/w)) were produced and stored for five weeks at 4°C. Results showed that probiotic viability was enhanced with the addition of 10% mango juice. Additionally, this formulation improved probiotics tolerance when exposed to *in-vitro* gastrointestinal digestion. According to the sensory analysis, beverage sensory scores improved as levels of mango juice increased from 20% to 40%.

Keywords: Probiotics, Dairy processing, Fermented milk, Gastrointestinal health, *Lactobacillus acidophilus*, Sensory analysis

INTRODUCTION

Probiotics are live microorganisms that, when administered in adequate amounts, confer a health benefit on the host (Hill *et al.* 2014). The probiotic market is experiencing rapid growth in both food and nutrition supplement industries. Food manufacturers are attracted to the usage of probiotics due to the projected market growth, high margins and growing consumer interest in functional foods (Bimbo *et al.* 2017). In 2012, the probiotic market was estimated to be worth \$26 million and by 2017 it had increased to approximately \$1.7 billion (Betz *et al.* 2015). Over the last ten years, more than 500 new probiotic food and beverages have entered the marketplace (Ramos *et al.* 2018). Such growth in the probiotic market has been attributed to changing consumer needs with demand moving towards healthy, convenient and portable food and beverages to suit a fast paced lifestyle (Allgeyer, Miller and Lee 2010). There is increasing evidence supporting the use of probiotics in treating health conditions such as coeliac disease, obesity and gastrointestinal conditions including irritable bowel syndrome (Ranadheera *et al.* 2017). Probiotics have also been associated with improved digestive health, prevention of urogenital infections, enhanced immune function, anti-allergy and anti-cancer activity, reductions in blood pressure and cholesterol levels (Joseph 2018) and could be beneficial in maintain oral health (Nadelman *et al.* 2018). Fermented foods have been consumed since the beginning of modern civilisation and are considered an ideal non-invasive method of delivering beneficial probiotic bacteria to the gut. Foods are fermented for many different reasons such as to extend the shelf-life of a product, improve the sensory profile and improve the nutritional composition. Dairy based probiotic food and beverages are currently widespread in the market. This could be associated with consumers believing these products are a more credible source of active ingredients than other non-dairy functional foods (Bimbo *et al.* 2017).

Currently, the greatest difficulty for manufacturers is maintaining probiotic viability during production, storage and their eventual digestion (Thomas 2016; Champagne, da Cruz and Dang 2018). To confer a health benefit to the host a minimum therapeutic level (10^6 - 10^7 cfu/g or ml of carrier food product) capable of manipulating biological function must be maintained throughout the shelf-life of the product (Ranadheera *et al.* 2012). The dairy matrix has inherently been reported to improve probiotic tolerance during digestion when compared with non-dairy products. This is a result of reducing probiotic contact with the harsh condition of the gastrointestinal tract and providing a buffering capacity *via* the milk and milk-fat contents (Ranadheera *et al.* 2017).

Dairy products such as yogurt (Kurtuldu and Ozcan 2018), cheese (Murtaza *et al.* 2017; Tomar 2019) and ice cream (Ayar *et al.* 2018) are considered as traditional food products that have been used in probiotic delivery. The addition of fruit juice and pulps to probiotic dairy products appears promising in improving probiotic viability, though results are seen to differ depending on the fruit and strain of bacteria used (Ranadheera *et al.* 2012). In some studies the addition of fruit juice and pulps is seen to modify the pH of the dairy product and introduce antimicrobial compounds such as organic acids and flavour compounds which could be harmful to the probiotic cultures (Shori 2016). On the other hand, improved viability has also been observed as the probiotics can obtain additional nutrients from the juice for cell synthesis, similar to the pathway used during fruit juice fermentation (Sarao and Arora 2017). In the past, yoghurts enriched with fruit juices and pulps have been associated with increased acceptance by consumers, as the sour tastes developed during fermentation are masked (Januário *et al.* 2017).

Recently there appears to be an increased interest by food manufacturers in the production of functional fruit and milk beverages. These beverages are relatively simple to produce, have increased consumer acceptance, and the beverages are perceived as healthy and refreshing which conform with current food trends (Lima Tribst *et al.* 2009; Islam *et al.* 2018). Mango is a popular tropical fruit worldwide; their popularity may be attributed to their appealing sensory properties and nutritional composition (Kim, Lounds-singleton and Talcott 2009). As mango are a seasonal fruit, when the supply is in surplus the excess fruits are often processed into purees or juices for use in jam, yoghurt, ice cream and beverages to avoid wastage (Kaushik 2015). Mangos are a good source of vitamins (A, C and E), dietary fibre and the mineral magnesium and potassium. They also contain the phytochemicals phenolic acid, mangiferin, carotenoids and gallotannins (Burton-Freeman, Sandhu and Edirisinghe 2017), which could provide health benefits to humans beyond that of micro- and macro-nutrients (Fazilah *et al.* 2018). Phenolic acid and carotenoids have been associated with antioxidant activity which is important in extending the shelf life of dairy products by preventing lipid oxidation. However, mango antioxidant activity can be affected by the cold storage (Atallah 2015;

Fazilah *et al.* 2018). Phytochemicals have also been reported to improve the viability of gut bacteria and their adhesion to intestinal epithelia cells (Parkar, Trower and Stevenson 2013).

The objective of this study was to develop a novel probiotic (*L. acidophilus*) dairy drink enriched with mango juice. Hence, the viability of *L. acidophilus*, *in vitro* gastrointestinal survival of *L. acidophilus* in mango dairy drinks, the major physio-chemical properties (titratable acidity, pH, colour and viscosity) during five weeks of storage at 4°C and the sensory properties of these products were analysed.

MATERIALS AND METHODS

Beverage formulation

To activate the freeze-dried probiotic, 0.5 g of *Lactobacillus acidophilus* La-5 culture (Christian Hansen, Australia) was mixed with 1 L of pasteurised full cream cow milk (Coles, Australia) and incubated at 37°C for 30 minutes in order to achieve at least 10⁶ cfu/ml. The milk was divided into five 200 ml portions. In order to maintain the consistency among the batches, canned mango slices (Coles Band, Coles Supermarkets Pvt Ltd, Australia, 13.6% sugar) were used to prepare mango juice in this study. The mango slices were drained of syrup and blended to a nectar-like consistency and pasteurised at 70°C for 20 minutes.

Five product formulations were trialled containing 0%, 10%, 20%, 30% and 40% mango juice (w/w) in milk (Table 1). A total quantity of 1400g per beverage formulation was produced per each trial and the whole experiment was repeated twice. Beverages were incubated/fermented at 37°C for 12 hours and stored at 4°C in plastic jars for five weeks for microbial and physico-chemical analysis. The samples for consumer acceptance were stored at 4°C in sterile plastic and glass bottles for three days before analysis was conducted.

Probiotic viability during storage

The probiotic viability of each beverage was evaluated weekly using De Man-Rogosa-Sharpe (MRS) agar (Edwards, Narellan, NSW, Australia) and spread plating techniques (*n*=3). Plates were incubated anaerobically at 37°C for 48 hours in containers with anaerobic sachets (Thermo Fisher Scientific, Scoresby, VIC, Australia), and counts were expressed as log cfu/ml as per Ranadheera *et al.* (2012).

Probiotic viability during *in vitro* gastrointestinal tolerance assay

Simulated gastric and small intestinal juice tolerance of *L. acidophilus* La-5 in the probiotic beverages was assessed at one week of storage as per Ranadheera *et al.* (2014). In brief, gastric juices were

prepared by suspending pepsin (Thermo Fisher Scientific, Scoresby, VIC, Australia) (3g/L) in 0.5% (w/v) NaCl solution. The pH was adjusted to 2.0 using concentrated HCl. Intestinal juices were prepared by suspending pancreatin (Thermo Fisher Scientific, Scoresby, VIC, Australia) (1 g/L) in 0.5% (w/v) NaCl solution with 0.3% bovine bile salts (Sigma-Aldrich, Castle Hill, NSW, Australia). The pH was adjusted to 8.0 using 0.1N NaOH.

A 1 ml beverage sample was transferred into screw cap tubes (Sarstedt Australia Pty Ltd., Mawson Lakes, SA, Australia) containing either 9 ml of gastric or small intestinal juices. The mixture was homogenized well for one minute using a vortex mixer (Scientific Equipment Manufacturers, SA, Australia). Gastric juice samples were incubated at 37°C for a total of 180 minutes and aliquots of 1 ml were removed from the tubes at 0, 60 and 180 minutes and plated onto MRS agar using the serial dilution and spread plating techniques. Intestinal juice samples were incubated at 37°C for a total of 240 minutes and aliquots of 1ml were removed from tubes at 0, 60 and 240 minutes and plated as above. Plates were incubated anaerobically at 37 °C for 48 hours in containers with anaerobic sachets and counts were expressed as log cfu/mL.

Physico-chemical analysis

All physico-chemical measures were performed weekly during the study period in duplicates. The pH was measured using a HANNA Instruments digital pH meter (HANNA Instruments, Smithfield, USA). Viscosity was determined using a Brookfield viscometer at room temperature (Brookfield, Middleboro, USA). A size 2 spindle was used for all samples, spindle speed varied between samples. Colour was assessed using a Konica Minolta colorimeter (Konica Minolta CR-410 Chroma Meter, Japan), and L*, a* and b* values were recorded. As per the manufacturer, the L value indicated the brightness (100 lighter and 0 darker) of the beverage, whilst the a* value indicated the red/green (positive being redder and negative being greener) and the b* value indicated the yellow/blue (positive being yellower and negative being bluer). Titratable acidity (TA) was analysed as per Ranadheera *et al.* (2016), by titrating 9 g of sample with 0.1N NaOH solution using phenolphthalein (Chem-supply, Gillman, SA, Australia) as the indicator.

Sensory analysis

The sensory evaluation was conducted following approval by the Veterinary and Agricultural Sciences Human Ethics Advisory Committee of the University of Melbourne, Australia (Ethics ID: 1852110.1).

Sensory analysis was performed within one week of production. The sensory panel included 63 healthy consumers (36 female, 25 male and 2 not stated) within 20-47 of age, were recruited by advertising at the University of Melbourne online notice board, who were not allergic to milk.

Participants were asked to evaluate the five beverage formulations based on overall liking, colour, aroma, appearance, flavour and mouthfeel/texture using a 9 point hedonic scale; dislike extremely = 1, dislike very much = 2, dislike moderately = 3, dislike slightly = 4, neither like nor dislike = 5, like slightly = 6, like moderately = 7, like very much = 8 and like extremely = 9 (Ranadheera *et al.* 2012).

Statistical analysis

Data analysis was performed using Minitab (version 17) statistical software (Minitab Inc., USA). Probiotic viability and physico-chemical data were analysed using two-way repeated measure ANOVA with storage time and mango juice levels as factors. Sensory data was analysed using the ANOVA general linear model and Bonferroni post-hoc test. All tests were conducted at a 95% confidence level.

RESULTS AND DISCUSSION

Probiotic viability during storage

As predicted, there was a decreasing trending in *L. acidophilus* viability over the five-week storage period at 4°C (Figure 1). The beverage containing 10% mango juice was found to maintain the highest viability during the storage period. The beverage still contained sufficient counts of *L. acidophilus* (7.72 log cfu/mL) to be termed a probiotic product after five weeks of storage. On the contrary, the addition of 40% mango appeared to have a significant ($p < 0.05$) negative influence on probiotic viability. At the end of storage period, probiotic viability in 40% mango drink (4.08 log cfu/ml) were no longer at the recommended therapeutic levels (Ranadheera *et al.* 2017). Similar trends were observed for the formulations containing 0%, 20% and 30% mango juice at the end of storage and no longer contained sufficient probiotic counts.

In order to confer a health benefit to the host, probiotic viability must be maintained at 10^6 - 10^7 cfu/mL until the beverage expiry date (Ranadheera *et al.* 2017). The results of this study appear to agree with current literature, whereby the addition of fruit juice to a probiotic beverage can enhance or reduce probiotic viability depending on the formulations of beverages (Shori 2016; Sarao and Arora 2017; Islam *et al.* 2018). Results revealed that probiotic viability across five weeks of storage was enhanced with the addition of 10% mango juice compared with the control (0% mango). Milk naturally contains the essential growth factors required for probiotic survival, though concentrations may not be optimal. Therefore, the addition of mango juice may provide additional nutrients to support cell synthesis and enhance growth (Shori 2016; Sarao and Arora 2017). However, when 20% or more mango juice was added to the beverage, probiotic viability during storage was reduced which may be a result of the decrease in pH or the addition of antimicrobial compounds present in the

mango juice or both (Shori 2016). Therefore, in terms of probiotic viability maintaining, there may be an optimum fruit juice level and more research of the sort is needed to establish such guidelines. Since consumers are more likely to consume dairy products with high fruit juice levels (Ranadheera *et al.* 2012), it would be interesting to investigate various strategies such as microencapsulation of probiotic cells to improve their viability in dairy food formulations with higher levels of fruit juice concentrations. If viability can be maintained, the natural fruit concentrations can be increased in such formulations providing consumers with a product which brings them close to meeting the recommended therapeutic guidelines (Ranadheera *et al.* 2017).

Probiotic viability during *in vitro* gastrointestinal digestion

Probiotic viability during *in vitro* gastric transit phase was significantly reduced ($p < 0.05$) in all beverages with or without added mango juice (Table 2). The addition of 10% mango juice resulted in greatest survival rate of *L. acidophilus* amongst the beverage formulations with a count of 4.15 log cfu/mL at the end of 180 min. Alternatively, the addition of 20% or more mango juice resulted in poor acid tolerance and reduced probiotic viability.

As seen in Table 3, within one minute of exposure to small intestinal juice *L. acidophilus* counts decreased in all samples by approximately 1-2 log cycles. A significant reduction, almost 50% loss in viability was apparent within one minute in the 40% mango juice formulations ($p < 0.05$). At the end of the *in vitro* intestinal transit (after 240 minutes), formulations containing 0% and 10% mango juice still contained sufficient counts of probiotics. The beverage formulation containing 20% mango juice or more did not contain satisfactory numbers of probiotics at the end of 240 minutes.

In order to provide the health benefits for the consumer, probiotics should survive the gastrointestinal transit and colonize in the gut. When exposed to gastric and small intestinal juice *in vitro* the addition of 10% mango was observed to have a protective influence on viability. This may be due to its ability in maintaining higher viability levels during beverage formulations and refrigerated storage. The decrease in viability experienced by formulations containing 20% or more mango may be a result of the lower milk concentration within these beverages. Dairy products have been found to exert a protective effect on probiotic viability during digestion by reducing bacterial exposure to the gastrointestinal environment (Ranadheera *et al.* 2017). High fat dairy foods have also been found to enhance probiotic viability and bile acid tolerance. Adding mango at concentrations $>10\%$ may generate diluting effect on fat contents and thus the protective effect of milk fat is reduced (Ranadheera *et al.* 2017). The lower viability in higher mango formulations may also due to its ability in lowering the probiotic tolerance to environmental stress. Probiotic viability in dairy beverages has been associated with the probiotic strain used, the presence of dissolved oxygen and hydrogen

peroxide, pH, the concentration of metabolites (lactic acid), storage temperature, other ingredients in the formulation and buffering capacity (Shori 2016). However, *in vivo* and clinical studies are needed (Martins *et al.* 2018) to fully elucidate gastrointestinal survival of *L. acidophilus* in this product.

Physico-chemical analysis

The beverage formulations *via* adding mango juice were detected to inherently vary in pH. Data showed a positive correlation between the amounts of added mango juice and the decline in pH value (Figure 2). Storage time was only found to have a significant influence on the pH of the formulation containing 0% mango ($p<0.05$). As shown in Figure 3, titratable acidity was significantly influenced by beverage formulation ($p<0.05$). In terms of TA, the formulations containing 0%, 10% and 20% mango were found to be significantly influenced by storage time ($p<0.05$). The beverages containing 30% and 40% mango maintained a relatively stable TA over the storage period.

Several studies have reported an association of probiotic viability with the intrinsic characteristics of the beverages such as acidity (Pereira *et al.* 2018). Milk is known to have a pH of approximately 6.5-6.7, whereas mango has a pH of 3.5-4.0 (Kaushik, Nadella and Rao 2015). As predicted, when formulations contained additional mango the pH appeared to decrease. This was associated with the lower viability of probiotics in beverages with higher levels of mango juice in the present study (Figure 1). Beverage pH was also decreased gradually over the study period as a result of the production of lactic acid by *L. acidophilus* which is known as post-fermentation. Similar trends in pH were reported by Ertem and Cakmakci (2018) in a fermented yoghurt containing *L. acidophilus*. Post-fermentation acidity development is detrimental for probiotics and was also observed by Ranadheera *et al.* (2012) in stirred fruit yoghurt containing ABY-1 culture (*Streptococcus thermophilus*, *L. delbrueckii subsp. bulgaricus*, *L. acidophilus* LA-5 and *B. animalis subsp. lactis* BB-12).

The decrease in pH during storage appeared to be positively correlated with the increasing TA; whereby as lactic acid is produced the pH decreases and the TA increases. Beverage formulations containing 0% and 10% mango appeared to experience a greater increase in TA than formulations containing 20%, 30% and 40% mango juice. This could be a result of maintaining probiotic viability in relatively higher numbers in formulations containing 0% and 10% mango and consequentially an increase in rate of lactic acid production during storage. Higher levels of initial TA and lower pH in mango containing beverages in this study could also be associated with the presence of malic and citric acid within the mango juice (Medlicott and Thompson 1985).

There was a significant positive relationship between viscosity and the amount of added mango juice ($p<0.05$) (Figure 4). A significant difference was noted between the formulation containing 40%

mango with the rest of the beverages and the viscosity of the formulation containing 30% mango was also significantly different to the 0%, 10% and 20% mango juice containing beverages.

The beverage containing 40% mango measured 6560 mPas initially whilst formulations containing 0%-30% mango measured 11-615 mPas. The inherently thick viscosity of the 40% formulation may be a result of the total soluble solids content of mango juice (Islam *et al.* 2018). In yoghurt, casein, calcium and phosphorus within milk is known to produce the gel-like structure (Kanurić *et al.* 2018) and addition of mango juice may have supported to strengthen the structure. Similar viscosity trends were observed by Islam *et al.* (2018) during the development of a probiotic (*Lactobacillus* spp.) dairy beverage enriched with 5% and 10% mango juice.

As per the manufacturer's instructions, colour change was determined through the differences in L*, a* and b* values (Konica Minolta CR-410 Chroma Meter, Japan). Beverage formulation had a significant influence on the beverage L*, a* and b* values ($p=0.000$) (Table 4). L* and b* values were seen to decrease as formulated mango concentration increased, whilst the a* values increased. Storage time was only found to be significant for the a* value of formulations containing 20% and 30% mango ($p<0.05$).

Sensory analysis of beverages

The participants originated from 25 different countries, with majority stated their nationality as Australian (n=10), Chinese (n=13) and Indian (n=8). It was found that 79% of participants consumed dairy products daily (n=50). A total of 6% of respondents consumed probiotics at least daily, 41% weekly, 13% monthly, 17% yearly and 22% stated they had never consumed probiotic beverages. Additionally, it was found that none of the respondents consumed mango or mango flavoured beverages daily, 14% consumed them weekly, 49% monthly, 30% yearly and 6% stated they never consumed these types of products.

As seen in Table 5, beverage formulation was found to have a significant impact on consumer liking in all categories (overall liking, colour, aroma, appearance, flavour and mouthfeel) ($p<0.05$). The sensory scores also indicate that the addition of mango juice was positively associated with increased consumer liking across all categories, as 40% mango achieved the highest average score across all categories. The 10% mango formulation surprisingly received the lowest mean score in the overall liking, flavour and mouthfeel categories (although did not differ significantly from the 0% formulation), otherwise there was generally a correlation between increased liking and mango concentration.

Sensory flavour scores were generally increased with a higher mango juice concentration. The improved scores are likely to be a result of the mango providing sweetness and fruity flavours which mask the sour notes developed during fermentation. A study by Thompson *et al.* (2007) found that natural strawberry flavour and aroma, and sweet taste were key factors influencing consumer liking of strawberry drinkable yoghurts which corresponds with the finding of this study. Another study by Allgeyer, Miller and Lee (2010) found that consumers preferred a yoghurt drink which was neither too sweet or too sour. The lower sensory scores achieved by the 10% mango formulation in overall, mouthfeel and flavour categories may be a result of mango being too faint to be appealing. The addition of mango juice also introduced a stringy pulp which appears to be acceptable in the high mango formulation, but unappealing at low concentrations (hence the mouthfeel scores). The 40% mango formulation also achieved the highest mouthfeel and appearance scores by the consumers. This formulation was the most viscous which appeared to appeal to the consumers, which may be due to the beverage texture being likened to a smoothie.

The colour of the beverage is thought to be a result of the instinct characteristics of the mango and milk when combined in different concentrations. When colour measurements (Table 4) were viewed in conjunction with the colour sensory scores (Table 5,) it appeared that liking was correlated with a lower L* value (darker colour), and higher a* (redder) and b* (more yellow) values, resulting in the intense colour of the 40% formulation which received the highest sensory acceptability for colour. Alternatively, 0% mango had the highest L* value indicating a lighter sample, and the lowest a* and b values. This sample received lowest sensory scores for the colour attribute. Thick consistency of the 40% formulation as indicated by viscosity data (Figure 4) may be more appealing for the consumers and this could be a reason to achieve highest mouthfeel values for the 40% formulation. Apparently, acidic and sour nature in the same formulation (Figures 2 and 3) may have also contributed to higher consumer acceptability of tested sensory attributes. However, sensory evaluations with the help of trained panellists would be highly beneficial (Silva *et al.* 2018) in the process of commercialization of this product.

CONCLUSIONS

The addition of mango juice to a fermented probiotic beverage has potential in improving the quality characteristics of the final product. The addition of 10% mango juice appeared to improve viability over storage, though when added in higher concentrations a reduction in viability was evident. Similar trend was observed for the *in vitro* gastrointestinal survival of probiotics. However, a higher concentration of mango juice (40%) was preferred by consumers than at the lower levels. Results suggest that commercialisation of such a product must find a balance between consumer liking and maintaining therapeutic amounts of probiotics in these types of products.

CONFLICT OF INTEREST

There is no conflict of interest.

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- Figure 1.** Viable counts of *L. acidophilus* in fermented dairy drinks enriched with various concentrations of mango juice during 5 weeks of storage at 4 °C (Mean±SD)
- Figure 2.** pH of mango juice enriched probiotic dairy drink over 5 weeks of storage at 4 °C (n=4) Mean±SD
- Figure 3.** TA of mango juice enriched probiotic dairy drink over 5 weeks of storage at 4°C (n=4) Mean±SD

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465 **Figure 4.** Viscosity of mango juice enriched probiotic dairy drink over 5 weeks of storage at 4°C

466 (n=4) Mean±SD

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Table 1. Mango juice enriched probiotic dairy drink formulations

Mango (%)	Cultured milk (g)	Uncultured milk (g)	Mango juice (g)
0	200	1200	0
10	200	1060	140
20	200	920	280
30	200	780	420
40	200	640	560

Table 2. Effect of simulated gastric juice (pH 2.0) on the viability of *L. acidophilus* in mango juice enriched probiotic dairy drink (viable counts shown as log cfu/mL)

Formulation	0 min	1 min	60 min	180 min
0%	7.22 ± 0.14 ^A	6.05 ± 0.18 ^{aA}	4.88 ± 0.17 ^{aA}	<1
10%	8.02 ± 0.03 ^B	6.09 ± 1.54 ^{aA}	6.07 ± 0.23 ^{aB}	4.15 ± 0.21 ^{aA}
20%	7.47 ± 0.02 ^A	6.04 ± 0.30 ^{aA}	3.06 ± 0.23 ^{aC}	<1
30%	7.27 ± 0.08 ^A	6.60 ± 0.00 ^{aB}	<1	<1
40%	7.60 ± 0.33 ^A	6.84 ± 0.00 ^{aB}	2.00 ± 0.00 ^{aD}	<1

Mean values (±SD).

^{A,B,C,D} Values in the same row having different superscripts differ significantly.

^a Indicates a significant difference (p<0.05) in viable counts compared with the control (0 minutes/before the gastric tolerance assay).

Table 3. Effect of simulated small intestinal juice (pH 8.0) on the viability of *L. acidophilus* in mango juice enriched probiotic dairy drink (viable counts shown as log cfu/mL)

Formulation	0 min	1 min	60 min	240 min
0%	7.22 ± 0.14 ^A	6.32 ± 0.56 ^{aA}	6.27 ± 0.27 ^{aA}	6.42 ± 0.15 ^{aA}
10%	8.02 ± 0.03 ^B	6.91 ± 0.09 ^{aA}	7.00 ± 0.00 ^{aB}	6.50 ± 0.14 ^{aA}
20%	7.47 ± 0.02 ^A	5.94 ± 0.06 ^{aB}	6.11 ± 0.24 ^{aA}	4.45 ± 0.64 ^{aB}
30%	7.27 ± 0.08 ^A	5.16 ± 0.34 ^{aC}	<1	<1
40%	7.60 ± 0.33 ^A	4.29 ± 0.02 ^{aD}	<1	<1

Mean values (±SD).

^{A,B,C,D} Values in the same row having different superscripts differ significantly.

^a Indicates a significant difference ($p < 0.05$) in viable counts compared with the control (0 minutes/before the intestinal tolerance assay).

Table 4. Weekly colour difference expressed as L*, a* and b* values over 5 weeks of storage (n=4).

Formulation		WEEK 0	WEEK 1	WEEK 2	WEEK 3	WEEK 4	WEEK 5
0%	L*	57.10 ± 1.45	58.79 ± 0.35	58.13 ± 0.46	57.64 ± 0.20	54.77 ± 1.29	53.22 ± 2.30
	a*	-1.87 ± 0.18	-1.98 ± 0.07	-1.83 ± 0.05	-1.75 ± 0.12	-1.88 ± 0.06	-1.82 ± 0.13
	b*	3.92 ± 0.25	4.53 ± 0.63	4.84 ± 0.67	4.70 ± 0.51	4.82 ± 0.61	4.23 ± 0.29
10%	L*	55.46 ± 1.41	55.83 ± 1.59	56.13 ± 1.42	56.06 ± 0.58	56.23 ± 0.56	56.31 ± 0.90
	a*	-1.63 ± 0.11	-1.72 ± 0.03	-1.64 ± 0.02	-1.65 ± 0.11	-1.59 ± 0.08	-1.65 ± 0.04
	b*	10.48 ± 1.18	8.55 ± 0.68	11.21 ± 0.36	10.68 ± 0.45	10.07 ± 0.89	9.91 ± 0.76
20%	L*	53.03 ± 1.28	54.62 ± 1.28	54.95 ± 1.00	54.29 ± 0.43	54.46 ± 0.46	54.20 ± 0.58
	a*	-1.60 ± 0.13	-1.54 ± 0.01	-1.43 ± 0.01	-1.42 ± 0.03	-1.36 ± 0.01	-1.25 ± 0.03
	b*	13.70 ± 1.71	14.69 ± 0.03	14.45 ± 0.12	13.87 ± 0.19	14.07 ± 0.03	14.29 ± 0.18
30%	L*	50.59 ± 0.94	52.47 ± 1.08	53.37 ± 1.08	53.71 ± 0.63	52.99 ± 0.63	52.34 ± 0.28
	a*	-1.29 ± 0.12	-1.19 ± 0.02	-1.16 ± 0.05	-1.19 ± 0.04	-1.03 ± 0.01	-0.95 ± 0.01
	b*	15.98 ± 1.98	16.98 ± 0.41	16.61 ± 0.14	16.64 ± 0.35	16.47 ± 0.42	15.90 ± 0.59
40%	L*	48.90 ± 1.26	51.78 ± 1.03	51.87 ± 1.03	51.84 ± 0.59	51.15 ± 0.85	50.98 ± 0.33
	a*	-0.81 ± 0.02	-0.69 ± 0.15	-0.70 ± 0.12	-0.69 ± 0.05	-0.58 ± 0.02	-0.52 ± 0.08
	b*	16.97 ± 2.35	18.58 ± 0.56	17.96 ± 0.45	17.56 ± 0.73	17.10 ± 0.62	16.51 ± 0.88

Mean (±SE)

The L value indicated the brightness (100 lighter and 0 darker) of the beverage, whilst the a* value indicated the red/green (positive being redder and negative being greener) and the b* value indicated the yellow/blue (positive being yellower and negative being bluer).

Table 5. Mean scores of tasting panellists (n=63) for sensory properties of mango juice enriched dairy drink (0%, 10%, 20%, 30% and 40%) after one week of storage.

Formulation	Overall	Colour	Aroma	Appearance	Flavour	Mouthfeel
0%	4.63 ± 0.25 ^{BC}	5.27 ± 0.25 ^C	4.89 ± 0.20 ^C	5.24 ± 0.24 ^D	4.54 ± 0.25 ^{BC}	4.89 ± 0.26 ^{BC}
10%	4.55 ± 0.20 ^C	5.43 ± 0.22 ^{BC}	5.16 ± 0.17 ^C	5.60 ± 0.18 ^{CD}	4.08 ± 0.22 ^C	4.59 ± 0.22 ^C
20%	5.37 ± 0.20 ^B	5.98 ± 0.17 ^B	5.44 ± 0.14 ^{BC}	6.06 ± 0.18 ^{BC}	5.08 ± 0.22 ^B	5.38 ± 0.22 ^B
30%	6.19 ± 0.20 ^A	6.89 ± 0.13 ^A	5.98 ± 0.18 ^{AB}	6.41 ± 0.19 ^B	6.00 ± 0.21 ^A	6.16 ± 0.20 ^A
40%	6.66 ± 0.20 ^A	7.48 ± 0.14 ^A	6.25 ± 0.18 ^A	7.11 ± 0.16 ^A	6.70 ± 0.22 ^A	6.49 ± 0.21 ^A

Mean (±SE)

^{A,B,C,D} Values in the same column indicate significantly difference (p < 0.05)

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