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Modification of milk resulting in low potassium and minimal electrolyte changes with minimal changes in taste

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Research Article

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Abstract

The objective of this research communication was to produce low potassium milk in which other electrolyte changes and changes in taste were minimized. To reduce potassium concentrations, several studies have reported batch methods of directly mixing milk or formula with sodium polystyrene sulfonate, which can exchange cations such as potassium for sodium. However, they also reported increases in sodium content, decreases in calcium and magnesium content, and changes in taste, because sodium polystyrene sulfonate exchanged other substances such as calcium and magnesium for sodium. In the present study, a method of dialyzing whole cow's milk using both sodium polystyrene sulfonate and a small amount of water through cellophane membranes was developed. A batch method for comparison was also performed. Each milk sample was evaluated biochemically and analyzed for taste and aroma in a sensory analysis. We showed that the potassium concentration in the dialyzed milk was reduced to 38% of that in unreacted milk. It was also shown that changes in sodium (increased) as well as calcium and magnesium (decreased) in the dialyzed milk were less than half of those in the batch method milk. Sensory analysis showed that minimal changes occurred in the taste of the dialyzed milk.

Hyperkalemia is a potentially life-threatening condition caused by reduced renal excretion or excessive intake of potassium. In patients with impaired renal function, especially with a glomerular filtration rate below 15 ml/min, a slight increase in potassium intake can cause severe hyperkalemia (Lehnhardt and Kemper, 2011). Milk, which contains 39–44 mM of potassium (Bunchman *et al*., 1991; Ranjith *et al*., 1999; Lehnhardt and Kemper, 2011), is one of the high potassium drinks that young patients under potassium restriction have reported to miss the most (Picq *et al*., 2014). To reduce potassium concentrations, several studies have reported a batch method using ion-exchange resin of sodium polystyrene sulfonate for the pretreatment of milk or formula (Bunchman *et al*., 1991; Picq *et al*., 2014; Taylor *et al*., 2015), but also reported increases in sodium content, decreases in calcium and magnesium content, and changes in taste. This excessive increase in sodium content was apparently related to exchanges of calcium, magnesium, and proteins for sodium by sodium polystyrene sulfonate (Bunchman *et al*., 1991).

Other studies evaluating milk dialyzed against water showed that sodium, potassium, and chloride were significantly reduced, but calcium, magnesium, protein, and fat in the milk remained unchanged (Roadhouse and Koestler, 1929; Berlyne and Epstein, 1971). However, it was also shown that the taste of the dialyzed milk was tasteless and watery (Roadhouse and Koestler, 1929).

The objective of the present study was to produce low potassium milk in which other electrolyte changes were minimized and taste was conserved. A method of dialyzing whole milk with both sodium polystyrene sulfonate and a small amount of deionised water, which is the dialysate of this method (online Supplementary Fig. S1), was developed. This dialyzed milk was analyzed for its composition, taste, and aroma.

Materials and methods

Milk

A brand of homogenized-whole-cow's milk was commercially purchased. In the present study, weight measurements were utilized in the preparations of the milk, deionised water, and sodium polystyrene sulfonate for accuracy and convenience. A stirrer (CT-MINI 2-4990-01™; AsOne Corporation, Osaka City, Osaka Prefecture, Japan) was used for stirring the milk. The rotational speed of the stirrer was controlled at 60% of its maximum output to prevent the stirring bar from idling.

Sample preparation

Two methods for reducing potassium were evaluated; a dialysis method (DM) and a batch method (BM). Twelve sets each of dialyzed DM milk, the dialysate of DM, milk prepared using BM and unreacted milk were prepared for biochemical assays. Milk in the same set was obtained from the same carton or container of milk. DM and BM were repeated as many times as the number of sets. Another set of each milk type was also prepared for sensory analysis. In addition, another four sets of the dialyzed DM milk and unreacted milk were prepared to measure freezing point depressions. These freezing points were measured using a TL1-A™ (ThermoProbe, Inc., Pearl, Mississippi, USA) while the milk was stirred by CT-MINI 2-4990-01™.

Dialysis method

As shown in online Supplementary Fig. S1, 150.0 g of milk was dialyzed with 7.5 g of sodium polystyrene sulfonate (AmberLite™ IR120B, DuPont de Nemours, Inc., Wilmington, Delaware, USA) and 15.0 g of deionised water through a cellophane membrane. The dialysis reaction time was set to 300 min after which both the dialyzed DM milk and the dialysate were decanted and analyzed. Detailed materials and experimental procedures are discussed in the online Supplementary file (online Supplementary Figs. S1–S7).

Batch method for comparison

In BM we directly mixed milk with sodium polystyrene sulfonate as has been adopted in many studies (Bunchman *et al.*, 1991; Picq *et al.*, 2014; Taylor *et al.*, 2015). In the present study, 7.5 g of sodium polystyrene sulfonate was added to 150.0 g of milk. This mixture was stirred for 30 min and then allowed to settle for 30 min. The supernatant was decanted and used for analyses for its composition, taste, and aroma.

Biochemical assays

Twelve sets each of milk samples and dialysates were analyzed for potassium, sodium, chloride, magnesium, calcium, inorganic phosphorus, glucose, and pH. Potassium, sodium, and chloride analyses were carried out using electrode methods (Zannier *et al.*, 2002). Calcium analyses used the Arsenazo III method (Burkitt *et al.*, 2007). Inorganic phosphorus analyses were carried out by the molybdic-acid direct method (Burkitt *et al.*, 2007). Glucose analyses were performed using the enzymatic method (Neese *et al.*, 1976). The pH was measured using a digital pH meter (NISA-008™).

Magnesium analyses were carried out by the xylidyl blue method (Corina *et al.*, 2015), where the maximum measurement range was 3.3 mM. Therefore, when magnesium sample concentrations were ≥ 3.3 mM, the sample was diluted 10-fold, re-analyzed, and the result was multiplied by 10.

Sensory analysis

In sensory analysis, differences in taste and aroma in the dialyzed DM milk from the unreacted milk were evaluated. The differences in the BM milk from the unreacted milk were also evaluated. The evaluation of milk was conducted by six external paid and trained assessors (four females, two males; three aged 30s, two aged 40s, and one aged 50s) using a scoring method according to JIS Z 9080: 2004 (Japanese Standards Association, 2004). All assessors signed informed consent forms. Six attributes of milk were assessed: comprehensive evaluation, characteristic aroma of milk, sweetness, richness, sourness, and saltiness. The characteristic aroma of milk was evaluated by sniffing. The attribute of comprehensive evaluation of milk was scored with a favorability scale from −3 to 3; where −3 = very unfavorable, 0 = same as the unreacted milk, and 3 = very favorable. The other attributes were scored with an intensity scale from −3 to 3; where −3 = very weak, 0 = same as the unreacted milk, and 3 = very strong. Before assessment, the samples were kept at an air-conditioned room temperature of 25°C.

Statistical analysis

Two-sided paired *t*-tests were used to compare biochemical data from each milk procedure with these of the unreacted milk. All analyses were carried out using R (Version 4.0.2). A *P*-value < 0.05 was considered to be statistically significant.

Results

All experiments were conducted in a refrigerator with the temperature kept at 3–4°C. The resulting dialysate of DM remained clear after every reaction (online Supplementary Fig. S8). The dialyzed DM milk, which weighed 150.0 g before the reaction, weighed 148.8 g on average (95% Confidence Interval [CI] = 148.1–149.4) after the reaction.

Table 1 shows the biochemical data from each milk procedure. The amount of potassium in the dialyzed DM milk and the BM milk reduced to 38% (95% CI = 37–39) and 40% (95% CI = 39–42) on average of the level found in unreacted milk, respectively. When compared with the unreacted milk: the mean concentration of the sodium content in the dialyzed DM milk (25.6 mM, 95% CI = 24.3–26.9) increased to less than half of that in the BM milk (52.6 mM, 95% CI = 51.0–54.2, $P < 0.001$), the mean concentration of calcium content in the dialyzed DM milk (2.8 mM, 95% CI = 1.9 to 3.6) decreased to less than one-fourth of that in the BM milk (12.9 mM, 95% CI = 11.9 to 13.9, $P < 0.001$) and the mean concentration of magnesium content in the dialyzed DM milk (0.8 mM, 95% CI = 0.6 to 1.0) decreased to less than half of that in the BM milk (2.0 mM, 95% CI = 1.8 to 2.2, $P < 0.001$). The freezing points of the dialyzed DM milk and the unreacted milk were −0.52 (95% CI = −0.54 to −0.49) and −0.55 (95% CI = −0.58 to −0.53) on average, respectively.

Figure 1 shows the mean data for each attribute in the sensory analysis. Data from each attribute in the unreacted milk were adjusted to zero. Mean scores of comprehensive evaluations, the characteristic aroma of milk, and sweetness for the dialyzed DM milk (mean score: −0.8, −1.7, and −0.7, respectively) and the BM milk (mean score: −2.2, −0.7, and −0.3, respectively) were lower than those for the unreacted milk. Mean scores of richness for the dialyzed DM milk (mean score: 0) and the BM milk (mean score: 0) were the same as unreacted milk. Mean scores of sourness and saltiness for the dialyzed DM milk (mean score: 0.2 and 1.0, respectively) and BM milk (mean score: 0.8 and 2.2, respectively) were higher than those for the unreacted milk.

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Table 1. Milk and dialysate data

Test substance	Unreacted milk (*n* = 12)	Milk by batch method (BM) (*n* = 12)	Dialyzed milk by dialysis method (DM) (*n* = 12)	Dialysate of dialysis method (*n* = 12)
Potassium (mM)	36.2 ± 1.1	14.7 ± 0.8*	13.7 ± 0.8*	9.9 ± 0.5*
Sodium (mM)	17.3 ± 2.2	69.8 ± 2.9*	42.8 ± 1.4*	58.8 ± 1.9*
Chloride (mM)	28.0 ± 2.1	28.3 ± 3.3	25.4 ± 1.8*	26.7 ± 2.5*
Magnesium (mM)	3.8 ± 0.3	1.8 ± 0.1*	3.1 ± 0.2*	0.4 ± 0*
Calcium (mM)	28.0 ± 1.3	15.2 ± 1.3*	25.3 ± 0.8*	0.6 ± 0.1*
Inorganic phosphorus (mM)	25.6 ± 1.3	25.6 ± 0.9	25.1 ± 1.0	10.2 ± 0.6*
Glucose (mM)	1.7 ± 0.1	1.9 ± 0.1*	1.6 ± 0.1	0.3 ± 0.2*
pH (units)	6.62 ± 0.08	7.05 ± 0.11*	6.67 ± 0.07	6.91 ± 0.05*

Data presented are means ± standard deviations.
$^*P < 0.05$ compared with unreacted milk.

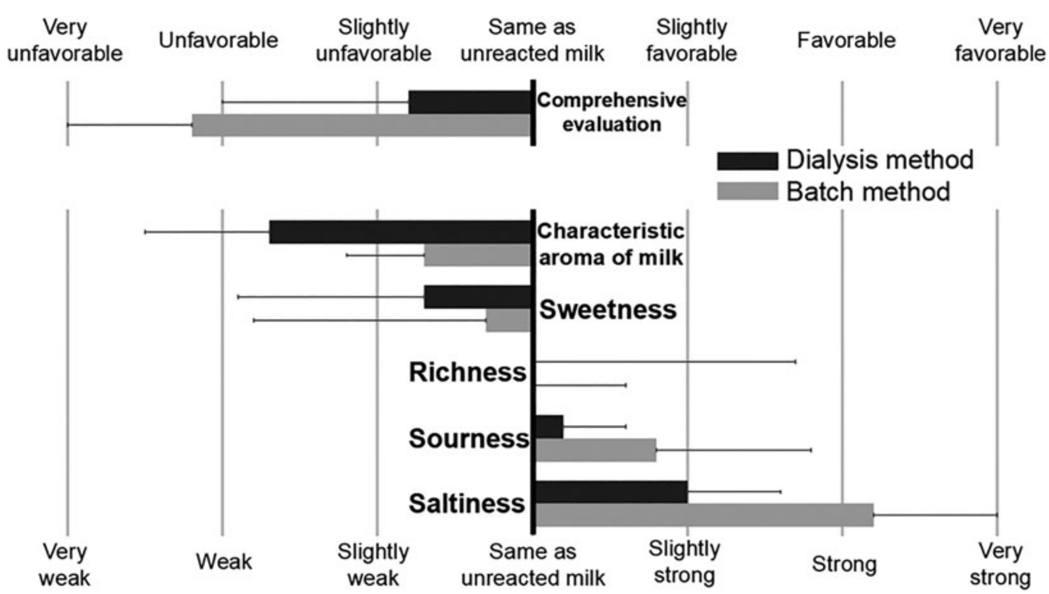

Fig. 1. Results of the sensory analysis. Data for each attribute in the unreacted milk were adjusted to zero. Mean data for each attribute in the dialyzed milk (DM) and the batch method milk (BM) are presented; error bars indicate standard deviations.

Discussion

Using the dialysis method, the potassium concentration in the dialyzed DM milk was reduced to 38% of that in unreacted milk. The increase in sodium content and the decreases in calcium and magnesium content in the dialyzed DM milk were less than half of those in the BM milk. Sensory analysis indicated that taste changes in the dialyzed DM milk were minimized. First, potassium in the milk passed through the cellophane membranes and moved into the dialysate during dialysis by the principle of diffusion (Wing and Magowan, 1975). Potassium in the dialysate was exchanged for sodium by the sodium polystyrene sulfonate and the dialysate's potassium concentration was reduced. This induced additional movement of potassium from the remaining milk passing through the cellophane membranes into the dialysate because the difference between the potassium concentrations in the remaining milk's content and the dialysate was maintained. As a result, the potassium concentration in the dialyzed DM milk was reduced.

Second, cellophane membranes minimized the increase in sodium content and the decreases in calcium and magnesium content in the dialyzed DM milk. Since the pore diameter of the cellophane membranes is regarded as approximately 5 nm (Wing and Magowan, 1975), small substances such as potassium, sodium, and chloride with diameters of 0.2–0.36 nm can pass through the cellophane membranes. However, large substances such as casein micelles, which consist of several proteins, with a diameter of 150–182 nm (Roy *et al.*, 2020) and fat globules with a diameter of 2800–4600 nm (Roy *et al.*, 2020) cannot cross the membranes. This was also supported by the result that the dialysate remained clear after the reaction (online Supplementary Fig. S8) because light scattering by casein micelles and fat globules can cause milk to appear opaque. Additionally, the majority of calcium ions (93%) and magnesium ions (84%) combine with

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casein, colloidal calcium phosphate, citrate, and so on in milk (Oh and Deeth, 2017), making it difficult for these substances to pass through cellophane membranes. Therefore, the cellophane membranes did not allow the majority of calcium and magnesium, as well as casein, to pass through and also prevented them from being exchanged for sodium by the sodium polystyrene sulfonate. Moreover, the excessive increase in sodium content is considered to be due to exchanges of calcium, magnesium, and proteins for sodium by sodium polystyrene sulfonate (Bunchman *et al*., 1991). Since these reactions were prevented by the cellophane membranes, the increase in sodium content in the dialyzed DM milk was also minimized.

Third, in addition to the cellophane membranes, the small volume of dialysate contributed to the conservation of taste. Nanofiltration membranes with pore sizes of 1–10 nm, which include the pore size ranges of cellophane membranes (Wing and Magowan, 1975), can concentrate the taste components of milk (Komatsu *et al*., 2005). However, the study which discussed dialyzing milk with the same amount of water showed that the taste turned tasteless and watery (Roadhouse and Koestler, 1929). This reveals that significant decreases in small substances in milk can also cause deterioration in taste despite the conservation of large substances. Therefore, in the present study, the weight of dialysate water was restricted to only one-tenth of the weight of milk. In this condition, if a small substance in milk which does not react with sodium polystyrene sulfonate fully diffuses into the dialysate, this substance may decrease by no more than approximately 10%. Chloride, where the concentration decreased by 9.2% (95% CI = 7.8–10.6) on average, is one such substance. In addition, the freezing point is in direct proportion to the molarity of the solute in milk (Shipe, 1959). The molarity of the solute in the dialyzed DM milk decreased by 6.3% (95% CI = −0.5–13.1) on average compared with that in the unreacted milk. Since at least a 20% increase in concentration of some taste substances is required before a person can distinguish a change in taste (Kobayashi *et al*., 2010), this approximate 10% change in the dialyzed DM milk may be permissible. For these reasons, the use of cellophane membranes and a small volume of dialysate water contributed to the conservation of taste.

In the sensory analysis for the dialyzed DM milk, the increase in sodium content affected not only saltiness, but other attributes. Fujikawa and Kawamura (2013), using 108 assessors, conducted a sensory analysis between unreacted milk and milk with the addition of 18 mM sodium chloride; attributes in the milk with sodium chloride compared with the unreacted milk were scored with scales from −3 to 3, similar to the present study. Interestingly, the mean scores in the dialyzed DM milk in the present study were similar to their results in the milk with sodium chloride: comprehensive evaluations −0.71, aroma of milk −0.22, sweetness −0.14, richness 0.03, sourness 0.35, saltiness 0.85. From this, it is thought that the result trends in the sensory analysis for the dialyzed DM milk were affected by the increase in sodium content. However, further analysis for aroma components will be needed to elucidate the reason the characteristic aroma of milk was weakened to a relatively large extent in the dialyzed DM milk.

A contradiction between pH and the sourness scores was found in the dialyzed DM milk and the BM milk. Similar to potassium, hydrogen ion was exchanged for sodium by the sodium polystyrene sulfonate. Therefore, a slight increase in pH in the dialyzed DM milk and an increase in pH in the BM milk were observed, as was also reported by Taylor *et al*. (2015). Nevertheless, increases of the sourness scores in both the dialyzed DM milk and the BM milk were reported in the sensory analysis. A hypothesis for this contradiction is that saltiness enhanced sourness due to taste–taste interactions (Keast and Breslin, 2003).

The present study did not investigate the condition of casein micelles in the dialyzed DM milk. Ranjith *et al*. (1999) argued that removal of calcium from colloidal calcium phosphate using ion-exchange resin induces disintegration of the casein micelle structure, although they reported that the concentration of protein in skim milk did not change after the reaction with ion-exchange resin. On the other hand, Fox and Hearn (1978) reported that casein micelles appeared to remain intact after dialysis, which was accompanied by a slight decrease in calcium content. Further investigation regarding the condition of casein micelles in dialyzed DM milk will be required.

In conclusion, the present study showed that our dialysis method employing both sodium polystyrene sulfonate and a small amount of water through cellophane membranes succeeded in producing low potassium milk where the increase in sodium content, the decreases in calcium and magnesium contents and the changes in taste were minimized.

Supplementary material. The supplementary material for this article can be found at https://doi.org/10.1017/S0022029922000358

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