Do the Characteristics of Human Milk Change With Storage, Warming, and Standing?

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Background

Because of the beneficial effects, all premature infants receive expressed human milk in the NICU (1). Data is needed to support optimal collection and storage conditions for this milk. Previously, our lab reported that expressed human milk can be stored at refrigerator temperature for 96 hours without affecting its integrity (2). The study found that there was a small, but significant decline in milk pH throughout the 96 hours of storage (2). It is known that expressed human milk can be maintained at room temperature for 6 - 8 hours before the bacterial colony counts rise (3,4). A recent observation is that freezer storage, thawing, and warming results only in minimal changes in milk characteristics, but subsequent standing at room temperature for 4 hours increases bacterial colony counts (5). This observation is pertinent to care in the NICU because after storage, thawing and warming, 4 hours may elapse in a usual tube-feeding session. To verify observed changes in *processed* milk after maintenance at room temperature, we compared those results to fresh milk or refrigerated/warmed milk maintained at room temperature for 4 hours.

Objectives

- 1. To determine if there is a difference in bacterial colony counts and pH levels when freshly expressed human milk is maintained at room temperature for 4 hours compared to the same milk stored at 4°C x1 hour, warmed, and then maintained at room temperature for 4 hours.
- 2. To compare the above changes with those reported from frozen milk that was stored frozen and then thawed, warmed, and maintained at room temperature.

Hypothesis

There is no difference in bacterial colony counts and pH levels among samples of freshly expressed human milk that is either stored at room temperature, or stored at 4°C before being warmed and maintained at room temperature, or milk that has been stored frozen and then warmed, thawed, and maintained at room temperature.

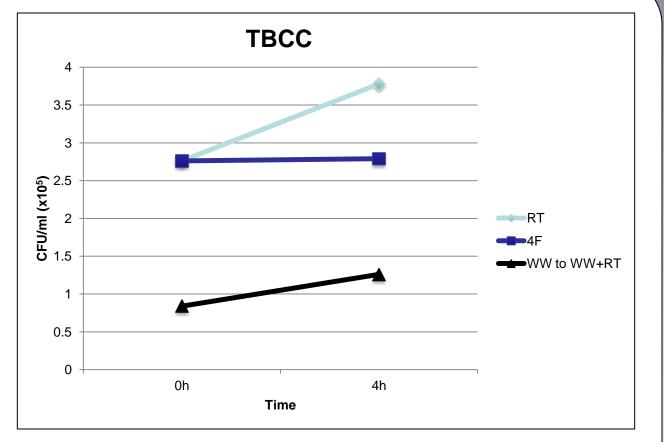
Methods

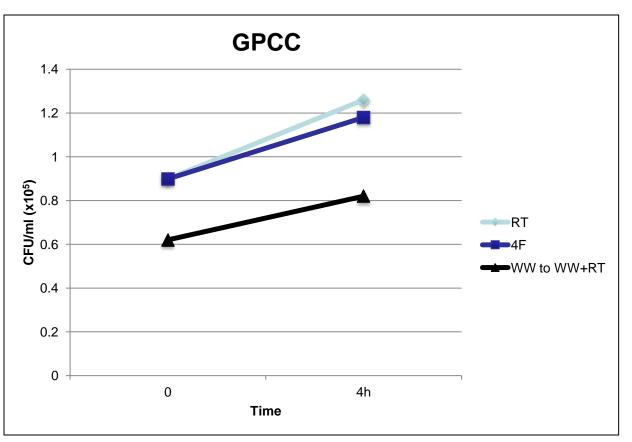
20 mL of random, fresh, expressed human milk samples (n=13) were collected from mothers in the NICU and were divided into two 10 mL aliquots. At time zero the pH levels and bacteria colony counts (TBCC) were measured. One 10 mL aliquot was stored at room temperature (RT) for 4 h. After the four hours the pH levels and TBCC were measured. The second aliquot was stored at 4°C for 1 h, warmed in a waterless warmer, and stored at RT for 4 h and pH level and TBCC were measured. TBCC included Gram positive (GPCC) and Gram negative (GNCC) colony counts assessed by growth on Trypticase Soy, Colombia CNA, and MacConkey Agar, (Fisher Scientific). pH was measured with the Fisher Scientific Accumet pH meter 915.

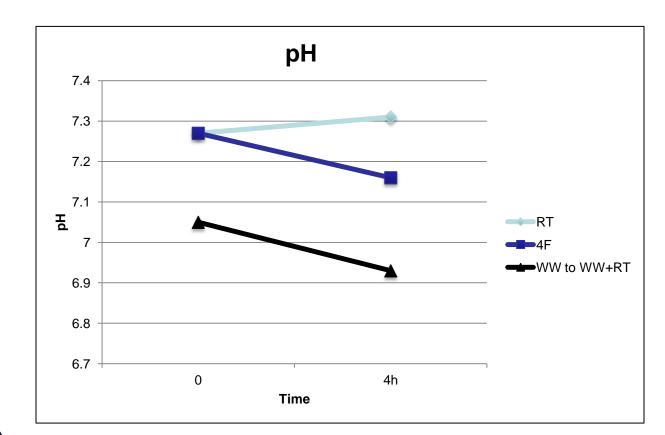
Results

Graph Key:

- RT = This sample is freshly expressed human milk maintained at room temperature (RT) for 4 h (aliquot #1).
- 4F = This sample is freshly expressed human milk that was stored at 4°C for 1 h, warmed in a waterless warmer, and then maintained at RT for 4 h (aliquot #2).
- WW = This separate sample was stored frozen, thawed, and warmed in a waterless warmer (comparison study sample#1).
- WW+RT = This sample was stored frozen, thawed, warmed in a waterless warmer, and then maintained at RT for 4 h (comparison study sample #2).
- 1. There were no differences in TBCC between fresh milk at time 0 and 4 h for sample RT and time 0 and 4 h for sample 4F. There were no differences in TBCC between samples RT and 4F in this study and samples WW and WW+RT from the comparison study.
- 2. There were no differences in GPCC between fresh milk at time 0 and 4 h for sample RT and time 0 and 4 h for sample 4F. There were no differences in GPCC between samples RT and 4F in this study and samples WW and WW+RT from the comparison study.
- 3. There were insufficient GNCC to determine any changes.
- 4. There were no differences in pH between fresh milk at time 0 and 4 h for sample RT and time 0 and 4 h for sample 4F. The pH was significantly greater in fresh milk at time 0 and 4 h for sample RT and time 0 and 4 h for sample 4F vs the comparison study (samples WW and WW+RT).







Conclusions

Bacterial colony counts and pH do not change significantly when fresh human milk is maintained at room temperature, even after being refrigerated and warmed.

Bacterial colony counts are similar when fresh milk is compared to milk that had been stored frozen and then thawed, warmed, and maintained at room temperature.

Milk pH is lower when milk has been stored frozen and then thawed, warmed, and maintained at room temperature compared with fresh milk maintained at room temperature. The differences in pH remain unclear and suggest that perturbations in milk with processing need to be explored before definitive protocols can be established.

References

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