Effect of pasteurisation on the mannose-binding lectin activity and the concentration of soluble CD14 in human milk

Madam,

Human milk is suitable for meeting many needs of the newborn.¹ However, during the early period of postnatal life, the neurological immaturity and often associated respiratory distress of preterm infants prevent them from suckling on their mothers' breasts effectively. They are initially fed expressed human milk by nasogastric tube or bottle. However, it was shown that between 60% and 80% of samples of expressed breast milk can be contaminated with bacterial pathogens or high amounts of skin contaminants.² Furthermore, it was shown that bacterial contamination of expressed human milk by breast pumps in neonatal intensive care units is not uncommon and has resulted in outbreaks of bacteraemia or fatal cases of necrotising enterocolitis.³ In order to prevent bacterial contamination, expressed human milk can be pasteurised. The use of pasteurised expressed breast milk raises some significant questions concerning the effects of storage, handling, and heat processing on the unique immunological components of the milk.⁴ In order to evaluate the effect of pasteurisation on the immunological quality of expressed breast milk, we aimed to investigate the impact of heat treatment on two bioactive factors present in human milk: mannose-binding lectin (MBL) and soluble CD14 (sCD14). Between July 2007 and March 2008, 38 human milk samples were collected from 38 lactating women whose babies were admitted to the neonatal intensive care unit of the University Hospitals Leuven, Belgium. The study was approved by the local Ethics Boards of the Catholic University of Leuven. Each woman received oral and written instructions about hygienic expression by an electric breast pump and about proper handling and transport of the milk. Stage of lactation, birth weight and gestational age were recorded. Each sample was divided in two aliquots. One aliquot was pasteurised at 62.5 °C for 30 min, and the other aliquot was kept raw. Samples were then centrifuged (2500 g for 10 min) to separate the lipid and aqueous fractions. Enzyme-linked immunosorbent assays (ELISAs) were performed on the aqueous fraction. Measurement of MBL activity was done using the *Wieslab*™ Complement system MBL pathway (DiaSorin SA/NV, Brussels, Belgium). A quantitative colorimetric ELISA kit was used to assess the concentration of sCD14

Figure 1 Mannose-binding lectin (MBL) activity (A) and soluble CD14 (sCD14) concentration (B) in non-pasteurised and pasteurised expressed human milk.

(R&D Systems Europe Ltd, Abingdon, UK). MBL activity and the concentration of sCD14 were reported by mean and SD. Paired comparisons (before/after pasteurisation) were performed using Wilcoxon–Mann–Whitney *U*-test.

Thirty-eight milk samples were harvested from mothers who delivered at a median of 38 weeks (range: 32–41); the babies had a median birth weight of 3055 g (range: 1400–4250). According to the stage of lactation, all but two samples consisted of transitional milk (postpartum days 6–14). Results of sCD14 concentrations and the MBL activity in breast milk before and after pasteurisation are shown in Figure 1. Although MBL activity was low, we did not find a significant difference between MBL activity in pasteurised (12.94% ± 2.1) vs non-pasteurised milk (12.47% ± 1.98). Comparison between the sCD14 concentration in pasteurised and non-pasteurised breast milk showed that the concentrations were significantly lower ($P < 0.01$) in pasteurised breast milk (0.28 µg/mL ± 0.11) than in non-pasteurised breast milk (2.29 µg/mL ± 0.32).

Human milk protects the infant from pathogens in the environment through both specific antibody-targeted mechanisms (secretory IgA, IgM, and IgG) and several distinct, broad-spectrum mechanisms, including the bactericidal effects of lactoferrin

and lysozyme.⁴ However, as a safeguard against the transmission of certain viral and bacterial pathogens, pasteurisation of human donor milk is now proposed by national guidelines in the UK.⁵ Although pasteurisation assures the microbiological safety of mother's milk, heat can destroy important nutritional biomolecules. Several studies showed that pasteurisation significantly reduced mean total protein, lysozyme and IgA concentrations.⁴ As heat can influence the structure as well as the functionality of proteins, we chose to measure MBL activity instead of MBL concentration. We were unable to show any difference between the pasteurised and non-pasteurised samples.

As sCD14 is actively secreted by differentiated mammary epithelial cells, a possible role in protection of the neonatal gut was considered.⁶ Our data show that pasteurisation significantly reduces the amount of sCD14 in breast milk. Together with other studies, which show that pasteurisation can decrease important immunological components, this suggests that pasteurisation of breast milk could lead to more infectious episodes in comparison to raw milk. This was indeed described by Schanler *et al.*, who found that infants fed with pasteurised human milk had more episodes of late-onset sepsis and necrotising enterocolitis than children fed with raw human milk.⁷ Additional research is needed to clarify the mechanisms and clinical relevance of the many bioactive factors in milk and the impact of heat treatment on these specific protective benefits.

Conflict of interest

None declared.

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Reducing blood culture contamination

Madam,

It has long been recognised that contamination of blood cultures is a problem for the clinician, in terms of the significance of result and decision to treat.^{1,2} Studies have suggested that rates for contamination of blood cultures of 2–3% are achievable.³

One approach to reducing contamination rates is by improving the technique of taking blood cultures. This has been shown to reduce contamination rates in one study although statistical significance was not reported; moreover, any detrimental impact on morbidity and mortality by