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Can Shaker Water Bath be used as Low Cost Pasteurization Device in Human Milk Banks?

Ranajit Mukherjee[1], Dinesh Munian[2], Abhishek Basu[3]

[1]Associate Professor, Department of Neonatology, IPGME&R – Kolkata.
[2]Assistant Professor, Department of Neonatology, IPGME&R – Kolkata.
[3]Administrator, Human Milk Bank, Department of Neonatology, IPGME&R – Kolkata.

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ABSTRACT

Background: Pasteurization is the most crucial step for donor milk processing in human milk banks and Holder pasteurization is the most widely accepted method. Automated instruments are available for pasteurization but they are costly. Shaker water bath is also used for pasteurization which is a cheaper device. In this study we checked the quality of pasteurization carried out in shaker water bath. Objective: Evaluation of biological safety of pasteurized human Milk by Shaker water bath and to see if pasteurization using shaker water bath alters macro-nutrient values of milk using human milk analyzer. **Methods:** (a) Bacteriologic quality of post pasteurized milk of 80 pasteurization cycles by shaker water bath. (b) Macro-nutrient values of pre and post pasteurized milk samples from shaker water bath are measured from 20 different cycles using Human Milk Analyzer (MIRIS AB, Sweden). These 20 samples were selected using computer generated random table. **Results:** Bacteriologic testof pre pasteurized sampleswere comparable and colony count of all samples were within acceptable limit (Gram negative bacilli <104, Gram positive cocci <104). None of the post pasteurized samples showed any growth. Shaker water bath pasteurization did not alter macro nutrient values (protein: 1.56±0.23g/dl, fat: 3.20±0.35g/dl, energy: 64.00±3.08Kcal/dl). **Conclusion:** Shaker water bath is a low cost pasteurization device for Holder pasteurization of human milk; there is no alteration of milk quality and is bacteriologically safe as well.

Keywords: Holder method, human milk analyzer, human milk bank, shaker water bath.

INTRODUCTION

Pasteurization of donated milk in Human Milk Bank (HMB) is the most important aspect of donor milk processing. Various methods of pasteurization are known or available but the Holder method (HoP, heating milk at 62.5^{0}C for 30 minutes) is the most widely accepted method. All available milk banking protocols recommend the pasteurized milk be rapidly cooled to 4^{0}C to prevent regrowth of micro-organisms.[1,2]

There are machines that incorporate consecutive heating and cooling cycles referred here as Automated Pasteuriser (AP), an example being Sterifeed S90; but these machines have high cost. An alternative less costlier device for HoP is Shaker Water Bath (SWB), also used in various milk banks.[3]

Can we pasteurize human milk at low cost using SWB maintaining international milk banking standards? To answer this question we checked bacteriologic quality of pasteurized milk using SWB.

Name & Address of Corresponding Author
Dinesh Munian,
Assistant Professor,
Department of Neonatology,
IPGME&R – Kolkata.

We have also studied changes in macro-nutrient values in pasteurized milk when using SWB.

MATERIALS AND METHODS

This descriptive study was undertaken in Human Milk Bank of Department of Neonatology, IPGMER, Kolkata, a teaching hospital in eastern India. This study was approved by ethical committee of IPGME&R, Kolkata. Data obtained in the period February to June 2016 is analyzed to check the microbiological quality and macronutrient values of pasteurized milk. Pasteurization is carried out in SWB.

Donors who are screened negative (HIV I & II, Hepatitis B & C, Syphilis), express their breast milk using hospital grade electric breast pumps. The pre pasteurization sample is not acceptable if total bacterial count is >105CFU/ml or Gram negative bacilli >104CFU/ml or Gram positive cocci >104CFU/ml.[1,2]

Donations of multiple donors are pooled under a laminar flow hood. Pre autoclaved conical flasks of capacity 1 liter are used for pooling and the process is carried out by a trained technician maintaining strict asepsis. From this pooled milk, a sample is taken for pre-pasteurization bacteriologic test. Next this pooled milk is poured in polypropylene bottles of 130ml capacity with a distribution of 100ml in each bottle. The bottles are labeled, capped and sealed with bottle sealer.

Pasteurization Equipment:

SWB made by Remi Elektrotechnik Ltd of Thane, India with model number RSB12 isused as pasteurizer. The bath chamber houses flexible clamps which can hold 12 bottles. The bath temperature and frequency of shaking can be controlled by regulators. The temperature of the bottle content during pasteurization is monitored by a temperature data logger.

Table 1: Description of AP and SWB

Description	AP	SWB
Model and make	S90, Sterifeed	RSB12, Remi Elektrotechnik Ltd
Cost (approx.)	INR 15,00,000	INR 50,000
Water Consumption per cycle	85 litres	7.5 litres
Water Pressure required	50-900 kN/m2, 7-130 lbs/sq	No need
Electrical specifications	230V/50Hz/1ph 2000W, FLA 9.2A	220V, 50Hz, Max. Amp. 3.4, Single phase

Figure 1: Shaker water bath

Pasteurization process and bacteriologic tests

This study assesses 80 pasteurization cycles in SWB. Screened negative express milk is stored in -200C freezer after labelling (donor ID, date of collection etc.). On day of pasteurization, the frozen milk bottles are brought out from the freezer and thawed in water bath. The water bath is preheated to 370C and then the frozen bottles are placed in it. Thawing cycle ends when the milk becomes liquid and it requires approximately half an hour.

The SWB is preheated to 62.50C and the bottles are placed in it. One extra bottle filled with 100ml water is also placed in the bath with which a temperature probe (data logger) is attached to monitor the temperature of bottle content during pasteurization. In SWB, the cooling cycle is arranged separately by immediately immersing the pasteurized bottles into a different chamber containing ice slurry with chilled water. Here also the temperature data logger is used to monitor the temperature of bottle content. After cooling, the indwelling temperature probe ensures that the milk reaches 40C. After cooling, a sample is taken for bacteriologic testing from one bottle of each cycle and the bottles are stored in -200C freezer.Both microbiologic samples are taken using a loop and directly inoculated into CLED agar and blood agar plates. Colony count of any type of growth is being assessed as per standard microbiological procedure.

Analysis of milk components

To check if pasteurization in SWB alters the macro-nutrient values of milk, milk samples are analyzed before and after pasteurization from 20 different batches out of 80 cycles. From each batch a bottle (containing pooled milk) is chosen and earmarked by random selection from which milk samples are collected for both pre and post pasteurization analysis.3ml of milk is collected using a disposable 5ml syringe for each analysis. The pre-pasteurization sample is collected during bottling of milk and the post-pasteurization sample is taken after completion of pasteurization. Macro-nutrient values of milk are analyzed using HMA made by MIRIS AB, Sweden. This HMA uses the principals of mid-infrared transmission spectroscopy. Before testing, all the milk samples are warmed to 400C in water bath to get optimum results as recommended by the manufacturer of HMA.

RESULTS

Bacteriologic test of pre and post pasteurization milk samples are done for 80 cycles of SWB. Out of 80 samples 73 samples contain GPC less than 10^2CFU/ml and 7 samples contain show GPC count between $10^3$to 10^4CFU/ml. For GNB, 60 samples contain less than 10^2CFU/ml and 20 samples show GNB between $10^3$to 10^5CFU/ml. None of the post pasteurization samples show any growth. [Table 2]

Table 2: Bacteriologic test results

HoP device	Pre HoP				Post HoP	
	GPC		GNB		GPC	GNB
	CFU/ml	No.	CFU/ml	No.		
SWB (80 cycles)	Nil	7	Nil	18	No growth	No growth
	< 10 - <102	66	< 10 - <102	42		
	< 103 - <104	7	< 103 - < 104	18		
	> 104	0	< 105	2		
GPC - Gram Positive Cocci, GNB - Gram Negative Bacilli, CFU - Colony Forming Unit						

Macronutrient values in pre and post Pasteurization (using SWB) milk samples measured in MIRIS HMA. p value shows no significant change in fat, protein or energy content. [Table 3]

Table 3: Macro-nutrient content in pre & post HoP milk (HoP is done in SWB), N = 20

Macro-nutrient values	Pre HoP	Post HoP	p value
Fat (g/dl)	3.24 ± 0.36	3.20 ± 0.35	0.90
Protein (g/dl)	1.54 ± 0.21	1.56 ± 0.23	0.85
Carbohydrate (g/dl)	6.42 ± 0.12	6.43 ± 0.13	0.67
Total solid (g/dl)	11.78 ± 0.33	11.79 ± 0.37	0.46

DISCUSSION

Our study shows SWB pasteurized milk is bacteriologically safe and there is no growth from any pasteurization sample as like other automatedpasteurizer machines. It is evident that AP is costlier, consumes more power and water; and so running cost is high. The only limitation of SWB is that the cooling following pasteurization has to be arranged separately.

We did not found any significant change in macronutrient values after pasteurization in SWB.A few studies have reported reduction in fat and energy after HoP.[4,5] García-Lara et al using MIRIS HMA showed a decrease in fat content following repeated freezing and thawing of pasteurized milk.[4] They emphasized that pasteurization itself is not related to reduction of nutrient values. In our study using MIRIS, we did not find any difference in fat before and after pasteurization unlike García-Lara et al probably because our samples were analyzed immediately after samples were taken, frozen samples were not tested. However Vieira et al using creamatocrit technique reported significant diminution in energy content of pasteurized human milk.[5] Peila C et al in a review in 2016 concluded that evidence is not sufficient to show that feeding pasteurized milk to newborn babies (compared to raw human milk) result in significant change in energy intake.[6]

Effect of pasteurization on protein has been reported in literature with conflicting findings. Vieira et al using IR analyzer showed a reduction in mean protein concentration (-3.9%) following pasteurization; a similar result is shown by Ley et al using biochemical analysis of milk.[7,8] In our study, we did not find any difference in protein content which is in agreement with Peilaet al.

In future we plan to look other variables like age of mother, gestational age of the baby and interval between childbirth and date of expression in days to look for any correlation between them and macronutrient values in raw as well as pasteurized human milk.

CONCLUSION

SWB is a low cost pasteurization device for HoP of human milk; there is no alteration of milk quality and is bacteriologically safe as well. Quality improvement in neonatal care in developing countries is expected to increase intensive care facilities and a parallel increase in demand of donor human milk for survival with better neurodevelopmental outcomes. There is a pressing need of setting up milk banks with low budgets to provide optimal quality milk for LBW newborns. Indigenously made machines and equipments at lower prices are needed; they will also ensure prompt maintenance services.

Further experiences from other centers will bolster our conclusion.

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