Relationships Between Symptoms and Changes in Breast Physiology During Lactation Mastitis

CATHERINE M. FETHERSTON,[1] C.T. LAI,[2] and PETER E. HARTMANN[2]

ABSTRACT

Objective: The objective was to investigate changes in milk composition that reflect variations in breast permeability, milk synthesis, and immune response in women before, during, and after mastitis

Methods: Mothers ($n = 26$) were followed prospectively from day 5 postpartum to the end of their lactation. Milk from each breast, blood, 24-hour urine samples, and data on breast and systemic pathologies were collected at reference intervals during the first 3 months postpartum, daily during the occurrence of any breast inflammation, and 7 days after resolution of symptoms, and was analyzed using mixed-model analysis (repeated measures).

Results: There was a significant difference in sodium ($p < 0.001$), chloride ($p < 0.001$), serum albumin ($p < 0.02$) and lactose ($p < 0.003$) in the breast with mastitis when compared with both the contralateral asymptomatic breast and "healthy" breasts. Inflammation of the whole breast was a significant predictor for a decreased glucose ($p < 0.01$) and hyperacute systemic symptoms predicted a decrease in milk glucose ($p < 0.03$) and an increased lactoferrin ($p < 0.05$) and sIgA ($p < 0.03$).

Conclusions: There is an increased breast permeability, reduced milk synthesis, and increased concentration of the immune components sIgA and lactoferrin with increasing severity of breast and systemic symptoms. The changes observed in milk composition during periods of increased breast permeability cannot be solely explained by the current theory of permeability of the paracellular pathway and further research in this area is required.

INTRODUCTION

INFLAMMATION OF THE BREAST during lactation is experienced in varying degrees of severity ranging from mild transient symptoms of inflammation, or blockage of the breast, to mastitis that is generally characterized by a more severe illness accompanied by systemic symptoms.[1] Approximately one in five women can expect to suffer at least one episode during their lactation,[1–5] yet there is little understanding of the physiologic and pathologic processes occurring before, during, and after mastitis.

Changes previously observed in the biochemical composition in milk from breasts with mastitis include increases in the concentrations of sodium,[6–11] sodium potassium ratio,[11] chloride,[8] serum albumin,[8,10] immune components,[9] and cytokines,[11,12] along with a decrease in lactose,[6–9] and glucose.[8] It is not

---

[1]Attadale Private Hospital, Perth, Western Australia, Australia.
[2]School of Biomedical, Biomolecular, and Chemical Sciences, The University of Western Australia, Crawley, Western Australia, Australia.

known if these changes occur in the presence of all the varying clinical forms of mastitis or to what degree the natural course of change and recovery occurs.

The aim of this study was to observe women before, during, and after the occurrence of mastitis to describe changes in milk composition that reflect variations in breast permeability, milk synthesis, and immune response.

MATERIALS AND METHODS

Sample collection, recruitment, and demographics of subjects have been reported in detail elsewhere;[13] however, in brief: The research conducted used a convenience sample of lactating women ($n = 26$), assessed to be at risk for developing mastitis, who were then followed prospectively from the fifth day postpartum to the end of their lactation. Reference samples of blood and milk were collected from women at intervals during the first 3 months postpartum to obtain data on the normal range of a set of potential biochemical markers for mastitis. Included in this range of markers were sodium (Na), chloride (Cl), lactose, glucose, serum albumin, lactoferrin, and secretory immunoglobulin A (sIgA) in milk and lactose in blood and urine. C-reactive protein (CRP) in milk and blood also was measured on days 5 and 14 postpartum. Women were interviewed at collection times to identify any coexisting breast (e.g., nipple trauma) or systemic pathologies. If women suffered with mastitis at any time during their lactation, further samples were collected daily during the course of their mastitis and at follow-up 7 days after resolution of symptoms. Women with mastitis completed a symptom log to provide descriptive data on the characteristics and management surrounding the onset and duration of their mastitis.

The study was approved by The University of Western Australia, Human Research Ethics Committee.

Operational definitions

Breast inflammation. Breast inflammation was defined as a warm to hot, red, and tender/ painful area of the breast. It was further categorized as localized, segmental, or total breast inflammation according to the area of inflammation present.

Systemic symptoms. Systemic symptoms were defined as *mild* (feels unwell, tires easily, afebrile [body temperature (T°) <37.5°C]); *acute* (unwell, T° >37.4°C and <38.5°C, myalgia, needs to go to bed); and *hyperacute* (T° >38.4°C, myalgia, headache or vomiting, rigors).

The symptoms experienced were then used to apply the following diagnostic criteria. Blocked ducts were defined as tenderness, erythema, and/or nodular area(s) of the breast present for less than 24 hours and without accompanying systemic symptoms.

Mastitis is defined as any inflammation of the breast present for more than 24 hours, and accompanied by any degree of systemic illness.

Biochemical analysis

Methods for the biochemical analysis conducted in this study along with recoveries and coefficients of variance (excluding sIgA and lactoferrin) have been reported by the authors in detail.[13] However, briefly: All analysis in milk was undertaken on samples that were defatted after collection (except chloride) and then stored at −20°C until analysis. The concentration of lactose and glucose in milk was determined by spectrophotometric assay[14] and lactose in urine and blood by bioluminescent assays.[15]

The enzyme-linked immunosorbent assay (ELISA) for serum albumin in milk was adapted from the method of Schwerer et al.[16] and is outlined in Cregan et al.[17] Determination of the sodium concentration in defatted milk samples was undertaken using a Corning 435 Flame Photometer® (Essex, UK) and chloride concentration in whole milk was determined using a combination chloride ion electrode (ORION Model 96-17B®) and a direct concentration read out specific meter (ORION 490A+®).

Measurement of sIgA and lactoferrin was undertaken using ELISA.[18] The recovery of a known amount of sIgA and lactoferrin added

to milk samples was $100.5 \pm 3.0\%$ ($n = 11$) and $96.7 \pm 5.9\%$ ($n = 5$), respectively. The interassay coefficients of variation were 3.8% ($n = 10$) and 4.9% ($n = 10$), respectively.

Statistical analysis

Descriptive statistics used means and standard deviations or medians and interquartile ranges, as appropriate, depending on normality. Hypothesis testing of all outcome measures was based on analysis of variance with repeated measures within a mixed model. Models were used where individual women were treated as random effects and comparison groups (e.g., mastitis/no mastitis) as fixed effects.

In the analysis of outcomes that occurred at various stages of lactation, appropriate adjustments were made, using stage of lactation (day postpartum) as one of the covariates in analysis of variance models. Estimate effects were presented as means and 95% confidence intervals (95% CI). Any p-values < 0.05 were considered significant. All data were analyzed using the mixed model module in SPSS 11.0 for Mac OSX.

RESULTS

Biochemical composition of milk from "healthy" breasts

Breast milk samples were collected at five reference time points, on days 5, 14, 30, 60, and 90 postpartum, and analyzed for composition of lactose, glucose, sodium, chloride, serum albumin, sIgA, and lactoferrin. Sample numbers varied at each collection time point because some mothers provided limited participation and others suffered mastitis or other breast symptoms at the time of collection; therefore excluding them from the "healthy breast" sample.

Mixed model analysis, adjusted for the presence of any breast symptoms and stage of lactation, showed no significant difference between concentrations of milk components in the right and left breasts for sodium ($p < 0.29$; df:1,184; F = 1.1), chloride ($p < 0.15$; df:1,184; F = 2.1), glucose ($p < 0.26$; df:1,184; F = 1.3)

lactose ($p < 0.54$; df:1,181; $F = 0.4$), serum albumin ($p < 0.140$; df:1,110; F = 2.2), lactoferrin ($p < 0.51$; df:1,182; F = 0.4), and sIgA ($p < 0.75$; df:1,182; F = 0.1). The combined results for "healthy" left and right breasts at each reference collection time point are shown in Table 1.

Changes in milk composition associated with events other than mastitis

Variation from the cohort's normal milk composition was observed in both women with breast symptoms other than mastitis and women with no breast symptoms. There was a marked increase in sodium and chloride and a decrease in lactose in milk in asymptomatic breasts at days 5 and 14. These changes were observed in women with birth at or before 38 weeks gestation ($n = 4$), Cesarean birth ($n = 3$), and increased CRP in blood above the normal mean for that time postpartum ($n = 5$). Women identified with low supply ($n = 1$), self-perceived oversupply ($n = 3$), and nipple trauma ($n = 7$); all showed an association with an increase in milk sodium ($p < 0.001$; $p < 0.001$, and $p < 0.004$, respectively) and a decrease in milk lactose ($p < 0.05$; $p < 0.015$; and $p < 0.001$, respectively). Changes also were observed in the concentrations of chloride, glucose, lactoferrin, and sIgA in milk, as well as the 24-hour excretion of lactose in urine (Table 2).

Incidence of mastitis in the "at-risk" sample

Applying the operational definitions previously outlined, 14 of the 26 at-risk mothers suffered a total of 22 episodes of mastitis (including two bilateral). Range of occurrence was day 5 to day 400 postpartum, median day 49 (20, 187). Seven mothers experienced 13 episodes of blocked duct(s) with range of occurrence from days 14 to 310, median 60 (25, 102).

Blocked duct(s)

One mother (M8), who had undetectable concentrations of sIgA in her breast milk and was subsequently diagnosed with IgA deficiency during the course of the study, contributed over 50% of episodes of blocked duct(s) ($n = 7$). Her data were omitted from the analysis and will be reported elsewhere. The re-

TABLE 1. REFERENCE VALUES FOR BREAST MILK FROM "HEALTHY" RIGHT AND LEFT BREASTS COMBINED (N)
COLLECTED AT REFERENCE TIME POINTS DURING THE FIRST 3 MONTHS POSTPARTUM

Milk component	Day 5 (n = 26)	Day 14 (n = 25)	Day 30 (n = 32)	Day 60 (n = 33)	Day 90 (n = 40)
Lactose (mmol/L)	164 ± 21.2	171 ± 19.6	178 ± 18.2	175 ± 16	175 ± 18.8
Glucose (mmol/L)	1.27 ± 0.68	1.4 ± 0.47	1.67 ± 0.46	1.7 ± 0.51	1.6 ± 0.54
Sodium (mmol/L)	21.4 ± 8.0	13.4 ± 4.8	12.6 ± 2.3	13.3 ± 4.9	12.6 ± 4.3
Chloride (mmol/L)	31.2 ± 8.6	21.1 ± 3.4	18.5 ± 3.7	19.7 ± 5.6	19.9 ± 5.3
sIgA (g/L)	1.65 ± 0.55	1.3 ± 0.54	1.0 ± 0.4	1.0 ± 0.38	1.1 ± 0.5
Lactoferrin (g/L)	5.8 ± 2.8	3.8 ± 2.9	2.9 ± 1.8	2.2 ± 1.2	2.6 ± 1.2
Serum albumin (g/L)	0.51 ± 0.72	0.52 ± 0.19	0.51 ± 0.20	0.35 ± 0.01	0.35 ± 0.14
Urinary lactose excretion (mmol/ 24 hours)	4.9 ± 3.2 (n = 13)	3.4 ± 1.1 (n = 12)	2.9 ± 1.15 (n = 16)	2.85 ± 1.67 (n = 16)	2.65 ± 1.46 (n = 20)

maining six mothers experienced one episode each of blocked duct(s), and although milk composition in the affected breast was variable, the median was within normal range (Table 3), as was median lactose excretion/24 hours at 3.0 (2.2, 5.1) mmol/24 hours. Variability in milk composition was noted to be associated with three mothers who had coexisting symptoms of nipple trauma ($n = 2$) and oversupply ($n = 1$).

Mastitis symptoms

At the onset of mastitis 71% ($n = 17$) of breasts affected ($n = 24$) presented with segmental inflammation, 13% ($n = 4$) with local-ized inflammation, and 11% ($n = 3$) with inflammation of the whole breast.

Duration of mastitis episodes ranged from 1 to 5 days, with a median of 2 days (1, 3). Breast symptoms for mastitis episodes ranged from 1 to 5 days (2.2 ± 1.1), whereas duration for systemic symptoms was from 1 to 4 days (2.0 ± 1). At the onset of mastitis episodes, 38% ($n = 8$) of mothers suffered mild systemic symptoms, 38% ($n = 8$) acute symptoms, and 28% ($n = 6$) hyperacute symptoms.

The severity of both systemic and breast symptoms was greatest at the commencement of the mastitis episode and decreased with time. The severity of breast inflammation did

TABLE 2. INDICATORS FOR CHANGES IN MILK COMPOSITION DURING THE FIRST THREE MONTHS POSTPARTUM

Breast condition	Sodium	Chloride	Lactose	Glucose	Lactoferrin	sIgA	Excretion of lactose in urine/24 h
Nipple trauma (n = 17)	Increased ($p \leq 0.004$)	Increased N/S	Decreased ($p \leq 0.001$)	Decreased ($p \leq 0.031$)	Decreased N/S	Increased N/S	Increased N/S
Perceived oversupply (n = 19)	Increased ($p \leq 0.001$)	Increased ($p \leq 0.001$)	Decreased ($p \leq 0.015$)	Decreased N/S	Increased ($p \leq 0.001$)	Increased N/S	Increased ($p \leq 0.001$)
Low supply (n = 4)	Increased ($p \leq 0.001$)	Increased ($p \leq 0.001$)	Decreased ($p \leq 0.05$)	Decreased ($p \leq 0.001$)	Increased N/S	Increased ($p \leq 0.04$)	Increased ($p \leq 0.034$)

Adjusted for coexisting pathologies and stage of lactation. Abbreviations and symbols: n = number of samples; N/S = not significant.

TABLE 3. ESTIMATED MEAN (95% CI) FROM THE MIXED MODEL ANALYSIS FOR CONCENTRATION OF MILK COMPONENTS
IN THE MASTITIS BREAST COMPARED WITH THE CONTRALATERAL ASYMPTOMATIC
BREAST, "HEALTHY" ASYMPTOMATIC BREASTS, AND WOMEN WITH BLOCKED DUCT(S)

Milk component	Mastitis breast estimated mean (95% CI)	Contralateral asymptomatic breast estimated mean (95% CI)	Statistical analysis (mixed model mastitis/no mastitis) adjusted for stage of lactation and coexisting pathologies	Blocked duct/s breast median (25, 75) (n = 6)	No mastitis "healthy breasts" estimated mean (95% CI)
Sodium (mmol/L)	21.8 (16.8; 26.9) (n = 80)	14.8 (9.7, 20.0) (n = 63)	p ≤ 0.001; df: 1,127; F = 23.9	13 (8, 18)	14 (12, 16) (n = 155)
Chloride (mmol/L)	30 (23.9, 36.8) (n = 80)	22 (15.7, 28.6) (n = 63)	p ≤ 0.001; df: 1,127; F = 23.6	22 (19, 26)	21 (19, 23) (n = 155)
Serum albumin (g/L)	0.74 (0.54, 0.92) (n = 79)	0.60 (0.41, 0.79) (n = 62)	p ≤ 0.02; df: 1,126; F = 5.6	0.5 (0.4, 0.8)	0.5 (0.36, 0) (n = 154)
Lactose (mmol/L)	159 (151, 166) (n = 80)	168 (161, 175) (n = 63)	p ≤ 0.003; df: 1,128; F = 8.9	170 (158, 189)	174 (169, 1) (n = 155)
Glucose (mmol/L)	1.39 (1.1, 1.69) (n = 79)	1.5 (1.2, 1.8) (n = 62)	p ≤ 0.21; df: 1,126; F = 1.6	1.5 (1.3, 2)	1.6 (1.4, 1) (n = 154)
Lactoferrin (g/L)	3.45 (2.2, 4.7) (n = 79)	2.68 (1.41, 3.95) (n = 62)	p ≤ 0.069; df: 1,128; F = 3.4	2.4 (1.2, 3.4)	3.2 (2.5, (n = 155)
sIgA (g/L)	1.22 (1.02, 1.43) (n = 79)	1.19 (0.99, 1.4) (n = 62)	p ≤ 0.068; df: 1,127; F = 0.164	1.2 (0.8, 1.6)	1.25 (1.1, 1 (n = 154)

not predict the severity of systemic symptoms ($p < 0.23$, df:2,71; F = 1.5).

Changes in milk composition during mastitis

The mixed model analysis showed a significant difference in the concentrations of sodium, chloride, serum albumin, and lactose in the breast with mastitis when compared with both the contralateral asymptomatic breast and "healthy" breasts from women without either mastitis or coexisting breast symptoms (Table 3). Although there was a trend for a decrease in glucose and an increase in lactoferrin and sIgA in the mastitis breast, the difference was not significant in the mastitis/no mastitis model. The 24-hour urinary excretion of lactose during mastitis has been reported in detail.[13] It was significantly higher during mastitis ($p < 0.001$; df:1,117; F = 26.6), peaked at the commencement of the mastitis at a median of 7.5 mmol/24 hours (6.3, 12.4), and decreased over time until there was no significant difference at the time of follow-up (7 days after resolution of symptoms) when compared with the mothers with no mastitis ($p < 0.25$; df:1,48; F = 1.3).

There were no significant differences in the breast with mastitis at follow-up when compared with "healthy" breasts for sodium ($p < 0.12$; df:1,168; F = 2.4), chloride ($p < 0.16$; df:1,157; F = 0.16), serum albumin ($p < 0.29$; df:1,173; F = 1.1), lactose ($p < 0.16$, df:1,153; F = 1.9), glucose ($p < 0.91$; df:1,164; F = 0.01), lactoferrin ($p < 0.93$; df: 1,157; F = 0.006), and sIgA ($p < 0.55$; df:1,162; F = 0.36). All analyses were adjusted for coexisting symptoms and stage of lactation.

Variation in milk composition changes during mastitis

Results from two mothers (M1 and M17) are presented as outlier examples of the varying course, and degree of change, in milk composition between individuals who experienced similar severity and duration of symptoms. The most extreme changes in milk composition were experienced by M17 (see Table 4), who reported oversupply problems in the weeks before suffering with mastitis. These results reflect the findings from the mixed models analyses (see Table 3), with a return to normal

TABLE 4. MILK COMPOSITION IN THE BREAST WITH MASTITIS OF M17 FOR EACH DAY OF HER MASTITIS EPISODE WITH FOLLOW-UP COMPARED WITH THE DAY 30 REFERENCE RESULTS FROM "HEALTHY" BREASTS

M17/ day postpartum	Sodium (mmol/L)	Chloride (mmol/L)	Lactose (mmol/L)	Glucose (mmol/L)	Serum albumin (g/L)	Lactoferrin (g/L)	sIgA (g/L)
day 61	77	100	82	1.54	2.54	3.2	0.81
day 62	50	71	135	1.51	2.25	2.3	1.07
day 63	27	55	175	1.54	2.0	2.1	0.9
Follow-up day 70	9	18	184	1.3	1.28	4.8	1.2
Day 30 reference results from "healthy" breasts	12 ± 2	18.5 ± 4	178 ± 18	1.6 ± 0.5	0.5 ± 0.2	2.9 ± 1.8	1.0 ± 0.4

concentrations at follow-up with the exception of serum albumin and lactoferrin, which were both increased in comparison with reference day 30 results from "healthy" breasts. The excretion of lactose on day 1 of the episode was very elevated at 12.5 mmol/24 hours compared with her reference day 90 sample, which was within normal range at 2.2 mmol/24 hours. In contrast M1, who had a history of mastitis, experienced minor changes in milk composition at the onset of symptoms followed by an overnight recovery of the concentrations of sodium, chloride, lactose, and serum albumin to within normal range despite going on to experience symptoms for another 3 days.

Changes in milk composition in relation to severity of symptoms

Mixed model analyses were undertaken to examine whether either the severity of breast symptoms (localized, segmental, or whole breast inflammation) or the severity of systemic symptoms (nil, mild, acute, or hyperacute) could predict changes in milk composition.

Although the concentration of sodium and chloride in the breast with mastitis increased according to severity of symptoms suffered, the differences were not significant.

Acute systemic symptoms predicted a significant decrease in milk lactose ($p < 0.037$; df:3,69; F = 2.9) compared with patients in whom systemic symptoms had resolved but breast inflammation remained.

The statistical models showed inflammation of the whole breast ($p < 0.01$; df:2,65; F = 5.0) and hyperacute systemic symptoms ($p < 0.034$; df:3,69; F = 3) were both significant predictors for a decrease in milk glucose compared with less acute symptoms (Fig. 1), where glucose remained fairly stable.

Although lactoferrin and sIgA followed a varied course, in regard to breast and systemic symptoms, the concentration of both defense components showed a trend toward increasing with severity of symptoms experienced. There was a marked increase in lactoferrin with the more extreme symptoms of whole breast inflammation, and both lactoferrin ($p < 0.05$; df2,71; F = 2.4) and sIgA ($p < 0.028$; df:2,67; F = 3.9) were significantly increased with hyperacute systemic symptoms.

Sodium, chloride, lactose, and serum albumin remained increased even when mothers had recovered from systemic symptoms but still experienced breast inflammation.

Mixed model analyses showed that neither milk composition at day 5 postpartum nor at the reference sampling days before mastitis could predict mastitis occurrence.

DISCUSSION

The prospective design used in this study has enabled a detailed observation of breastfeeding women before, during, and after their experience of mastitis. Recruiting breastfeeding
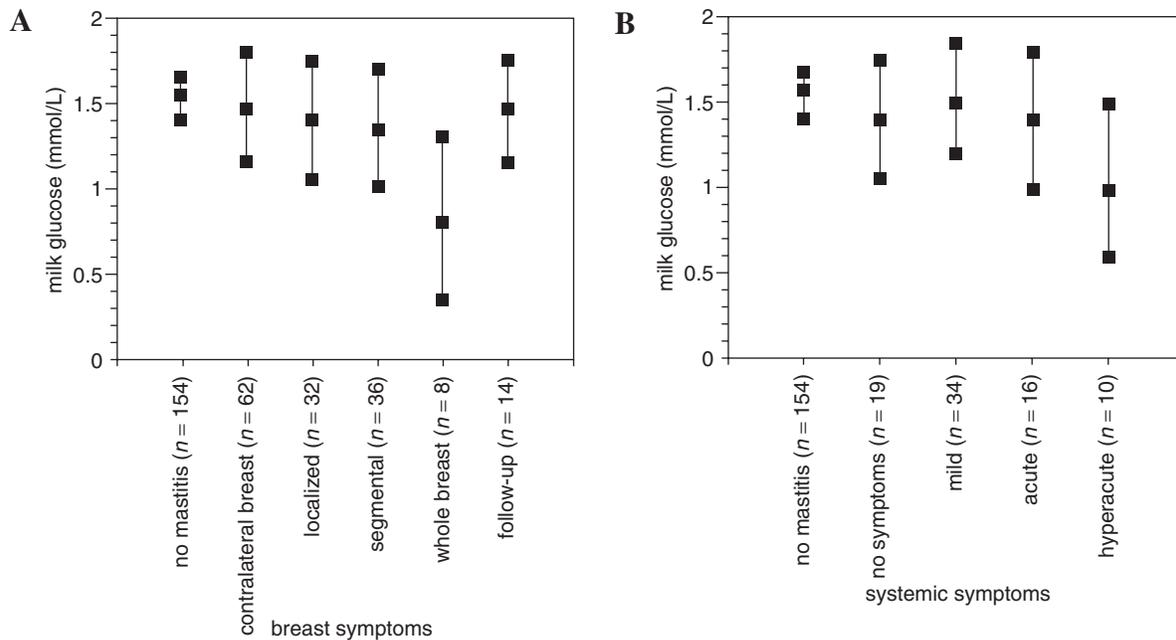
FIG. 1. The statistical models showed inflammation of the whole breast **(A)** and hyperacute systemic symptoms **(B)**, were both significant predictors for a decrease in milk glucose compared with less acute symptoms, where glucose remained fairly stable.

women with known risk factors for mastitis, in particular women with a past history of mastitis and/or nipple trauma and attachment difficulties in the early postpartum period, provided a very high incidence of mastitis with a wide range of severity of symptoms. Subsequently, this proved an efficient method for the prospective observation of women who develop mastitis during lactation. It also demonstrated the importance of counseling mothers with known risk factors for mastitis in strategies that can be undertaken to prevent this significant problem.

Although the results for milk composition from healthy breasts in the first 3 months postpartum have been controlled for any breast-feeding disorders, detectable through examination and interview, there are some marked differences in concentrations in some milk components compared with previously published findings.[19,20] Despite the general trends being similar, lactose concentrations at all reference time points within the first 3 months postpartum were lower, and conversely sodium and chloride concentrations were higher. The relatively low stable excretion rate for urinary lactose indicates that these variations are

not a result of increased paracellular pathway permeability. This suggests that the higher than normal concentrations of milk sodium and chloride observed in "healthy breasts" may be a normal physiologic response to a lower concentration of lactose, ensuring that the osmolality of milk remains isotonic with blood. This has important implications for the assumption that a raised sodium concentration is a result of subclinical mastitis.

The term subclinical mastitis has been applied to lactating women who present with increased sodium after day 3 postpartum in the absence of weaning.[21] However, this definition may obscure other potential causative factors. The authors' study identified other factors that may be associated with changes in the concentration of sodium and other milk components over the first 3 months postpartum, such as: preterm birth, nipple trauma, self-perceived oversupply, low supply, and an increased acute phase response (raised CRP). The latter supports other findings, in which an increase in the Na/K ratio in milk from both breasts was observed in women with systemic infection.[22]

The small sample of women diagnosed with blocked duct(s), showed no significant change

in milk composition, although there was a wide variation in some milk components. This is in contrast to breast inflammation that lasted longer than 24 hours and was accompanied by systemic symptoms (defined as mastitis), in which significant changes in sodium, chloride, lactose, and serum albumin in milk were observed. The apparent transitory nature and lack of significant change in milk composition during blocked duct(s) is reassuring, particularly as this is a very commonly reported problem among breastfeeding women.

The increase in the concentrations of sodium, chloride, and serum albumin and the decrease in lactose in the milk from the breast with mastitis is similar to that found in other studies.[6–11] The 24-hour excretion of lactose in urine also was significantly increased, confirming that these changes were likely to have occurred as a result of an increase in the permeability of the paracellular pathway. However, concentrations of glucose and serum albumin in milk remained raised even after sodium, chloride, and lactose had returned to normal, suggesting that the changes in milk composition were more complex than is currently explained by the increased permeability of the paracellular pathway breast model during mastitis. Also, the concentration of glucose in breast milk remained low at lactogenesis II (day 5) and stable during mastitis, except during severe symptoms, even in the presence of extreme changes in sodium, chloride, and lactose (see Tables 3 and 4). This raises many questions about the biological structure of the tight junctions within the breast and changes that might occur in response to pathology.

Although there were no significant changes in the general mastitis/no mastitis models for glucose, lactoferrin, and sIgA, the models differentiating between the degree of severity of inflammation and systemic symptoms were significantly predictive as severity of symptoms increased. The significant decrease in concentration of glucose during whole breast inflammation and hyperacute systemic symptoms indicates an inhibition of glucose uptake into the cell, rapid depletion because of increased metabolism resulting from loss of lactose, or both. The percentage of lactose excreted in urine did not account for the decrease in lac-

tose concentration within the breast.[13] Therefore, it is concluded that in addition to loss of lactose through the paracellular pathway there also was a decrease in lactose synthesis. This may result from both the decrease in available glucose, especially during extreme symptoms, and damage or death of lactocytes because of inflammation.

The significant increases in the concentrations of sIgA and lactoferrin only in the presence of whole breast and hyperacute systemic symptoms may partially explain the varied findings from different researchers about lactoferrin during mastitis. Buescher and Hair[12] did not find an increase in lactoferrin in milk during mastitis despite an increase in TNF-α concentrations. This is contrary to a report[11] of increased lactoferrin concentrations in women with mastitis (1.23 g/L) compared with women without mastitis (0.56 g/L). However, the concentrations reported in this study[11] during mastitis were not higher than the concentrations observed in the "healthy" cohort from the authors' study (2.9 g/L), the pooled results from Jensen[19] (1.65 to 1.94 g/L), or results from The Gambia[9] (1.87 g/L). The concentration of lactoferrin in milk appears quite variable, suggesting it may not be a reliable marker for mastitis.

Previous research has observed changes in the contralateral asymptomatic breast suggestive of subclinical inflammatory changes.[10] Despite an increased variation around the mean similar to the changes seen in the breast with mastitis, when controlled for stage of lactation and other coexisting pathologies, this study found no significant differences in the contralateral asymptomatic breast when compared with "healthy" breasts.

Milk from the breast with mastitis was analyzed at follow-up 7 days after the resolution of symptoms, and found to have returned to within normal range for most mothers. This confirms that the breast affected by mastitis made a rapid return to normal function in most cases, and supports other findings[9] that there was no significant difference in the breast with mastitis compared with the unaffected breast at follow-up of 5 weeks after initial presentation.

Milk composition has been found to be a predisposing factor for mastitis, with a lower con-

centration of sIgA observed in the milk of women who developed mastitis in The Gambia.[9] Although no significant predictors in milk composition were identified either at day 5 or at the sample collected before the occurrence of mastitis, it is of interest that the mother with no sIgA in her milk suffered with recurrent episodes of transient localized breast inflammation and apparent blockage.

CONCLUSION

Changes in milk composition observed in this study indicate there is an increased breast permeability, reduced milk synthesis, and increased concentration of the immune components sIgA and lactoferrin with increasing severity of breast and systemic symptoms. The changes observed in milk composition during periods of increased breast permeability cannot be solely explained by the current theory of permeability of the paracellular pathway, and further research in this area is required. Despite the acute illness and degree of breast inflammation often associated with mastitis, it is remarkable to find that the changes observed in milk composition are only temporary, and in most cases recovery of the breast to its normal state is achieved within 1 week of the resolution of mastitis symptoms.

As there appears to be a wide range of factors that might influence changes in some milk components, it is important that a mother's health status, and in particular breast health, should be taken into consideration when establishing a normal range for breast milk composition. A better understanding of how breast and systemic pathologies affect milk composition and breast physiology will be gained if a possible causative association for breast milk changes are further investigated and defined.

ACKNOWLEDGMENTS

REFERENCES

1. Fetherston C. Characteristics of lactation mastitis in a Western Australian cohort. *Breastfeeding Rev* 1997;5:5–11.
2. Riordin JM, Nichols FH. A descriptive study of lactation mastitis in long term breastfeeding women. *J Hum Lact* 1990;6:53–58.
3. Kinlay J, O'Connell DL, Kinlay S. Incidence of mastitis in breastfeeding women during the six months after delivery: A prospective study. *Med J Aust* 1998;169:310–312.
4. Vogel A, Hutchison BL, Mitchell EA. Mastitis in the first year postpartum. *Birth* 1999;26:218–225.
5. Foxman B, D'Arcy H, Gillespie B et al. Lactation mastitis: occurrence and medical management among 946 breastfeeding women in the United States. *Am J Epidemiol* 2002;155:103–114.
6. Ramadon MA, Salah MM, Eid SZ et al. Effect of breast infection on the composition of human milk. *J Reprod Med* 1972;9:81–83.
7. Conner AE. Elevated levels of sodium and chloride in milk from mastitic breast. *Pediatrics* 1979;63:910–911.
8. Prosser CG, Hartmann PE. Comparison of mammary gland function during the ovulatory menstrual cycle and acute breast inflammation in women. *Aust J Exp Biol Med Sci* 1983;61:277–286.
9. Prentice A, Prentice AM Lamb WH. Mastitis in rural Gambian mothers and the protection of the breast by milk antimicrobial factors. *Trans Royal Soc Trop Med Hyg* 1985;29:90–95.
10. Abakada AO, Hartmann PE, Grubb WB. Sodium and serum albumin in milk: biochemical markers for human lactational mastopathy. *Proc Austral Biochem Soc* 1990;22:SP52.
11. Semba RD, Kumwenda N, Taha TE, et al. Mastitis and immunological factors in breast milk of lactating women in Malawi. *Clin Diag Lab Immunol* 1999;6:671–674.
12. Buescher SE, Hair PS. Human milk anti-inflammatory component contents during acute mastitis. *Cell Immunol* 2001;210:87–95.
13. Fetherston CM, Lai CT, Mitoulas LR, et al. Excretion of lactose in urine as a measure of increased permeability of the lactating breast during inflammation. *Acta Obstet Gynecol Scand* 2006;85:20–25.
14. Arthur PG, Smith M, Hartmann PE. Milk lactose, citrate and glucose as markers of lactogenesis in normal and diabetic women. *J Paed Gast Nutr* 1989;9:488–496.
15. Arthur PG, Kent J, Hartmann P. Microanalysis of the metabolic intermediates of lactose synthesis in human milk and plasma using bioluminescent methods. *Anal Biochem* 1989;176:449–456.
16. Schwerer B, Bach M, Bernheimer H. ELISA for determination of albumin in the nanogram: Assay in cerebrospinal fluid and comparison with radial immunodiffusion. *Clin Chim Acta* 1987;163:237–244.
17. Cregan M. Mitoulas LR, Hartman PE. Milk prolactin, feed volume and duration between feeds in women

breastfeeding their full term infants over a 24 h period. *Exp Physiol* 2002;87:207–214.

18. Tijssen, P. *Practice and Theory of Enzyme Immunoassays*. Amsterdam, Elsevier, 1985.

19. Jensen RG, ed. *Handbook of Milk Composition*. San Diego, Academic Press, 1995.

20. Kulski JK, Hartmann PE. Changes in human milk composition during the initiation of lactation. *AJEBAK* 1981;59:101–114.

21. Filteau SM, Lietz G, Mulokozi G, et al. Milk cytokines and subclinical breast inflammation in Tanzanian women: Effects of dietary red palm oil or sunflower oil supplementation. *Immunology* 1999;97: 595–600.

22. Georgeson JC, Ahmed Y, Filteau S, et al. Longitudinal changes in milk sodium/potassium ratio in women with serious infection in the postpartum period. In: Davis MK, Isaacs CE, Hanson L, Wright AL eds. *Integrating Population Outcomes, Biological Mechanisms and Research Methods in the Study of Human Milk Lactation.* New York, Kluwer/Plenum, 2002.

Address reprint requests to:
Catherine M. Fetherston, Ph.D.
Attadale Private Hospital
21 Hislop Road
Attadale 6156
Perth, Western Australia, Australia

E-mail: cfethers@gmail.com

This article has been cited by:

1. Janet L. Engstrom , Paula P. Meier , Briana Jegier , Judy E. Motykowski , Joyce L. Zuleger . 2007. Comparison of Milk Output from the Right and Left Breasts During Simultaneous Pumping in Mothers of Very Low Birthweight Infants. *Breastfeeding Medicine* **2**:2, 83-91. [Abstract] [PDF] [PDF Plus]

2. Peter E. Hartmann . 2007. The Lactating Breast: An Overview from Down Under. *Breastfeeding Medicine* **2**:1, 3-9. [Abstract] [PDF] [PDF Plus]

3. Wei Wei Pang, Peter E. Hartmann. 2007. Initiation of Human Lactation: Secretory Differentiation and Secretory Activation. *Journal of Mammary Gland Biology and Neoplasia* **12**:4, 211. [CrossRef]