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Is Ingestion of Milk-Associated Bacteria by Premature Infants Fed Raw Human Milk Controlled by Routine Bacteriologic Screening?

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Expressed human milk is often used to feed premature infants. Raw milk contains bacteria which may be a source of infection. Milk banks have developed screening programs which combine periodic quantitative milk cultures with arbitrary rules specifying limits of bacterial concentration. It is unknown whether such programs succeed in preventing infants from being fed milk containing bacteria. At the Health Sciences Centre (Winnipeg, Manitoba, Canada), milk is screened once weekly. When a woman's milk is found to have excess bacteria, it is discarded only if she is an unrelated donor (as opposed to an infant's mother). To assess the effectiveness of this screening program, we determined the frequency at which infants fed raw human milk were exposed to milk-associated bacteria and compared the bacterial contents of donor and maternal milk. From February 1986 to April 1987, all human milk fed to 98 premature infants during the first 2 weeks of feeding (n = 10,128 feeds) was cultured quantitatively. Among study infants, 100% were exposed at least once to coagulase-negative staphylococci, 41% were exposed to *Staphylococcus aureus*, and 64% were exposed to gram-negative bacilli. The proportions of feeds containing bacteria and the quantities ($\log_{10}$ CFU [mean ± standard deviation]) ingested per positive feed were: 39% and 5.9 ± 0.5 for coagulase-negative staphylococci; 2.4% and 5.1 ± 1.0 for *S. aureus*; and 5.2% and 4.8 ± 1.1 for gram-negative bacilli. There were no adverse events attributable to ingestion of milk-associated bacteria. Milk coagulase-negative staphylococcal isolates were multiply antibiotic susceptible, whereas infant isolates were antibiotic resistant. Donor milk was significantly less likely than maternal milk to contain coagulase-negative staphylococcal species in any quantity (40 versus 93% of samples, respectively [$P < 0.001$]) or in concentrations exceeding 10^8 CFU/liter (3 versus 27% of samples, respectively [$P < 0.0001$]). There was no difference between milk from either source in terms of *S. aureus* or gram-negative bacterial content (4 to 6%). These results suggest that the Health Sciences Centre screening program is effective in limiting the number of harmless coagulase-negative staphylococcal species but has no impact on the quantity of potentially pathogenic bacteria ingested by premature infants. Implications for screening donor milk are discussed.

Breast milk is considered to be the best food for premature infants, not only for its nutritional value but also for its ability to provide protection against infection (15, 18) and necrotizing enterocolitis (2, 11, 17, 23). Since infants of very low birth weight are unable to suckle directly, donor milk programs have been used to ensure a steady supply of expressed human milk (1, 21, 27). Controversy exists regarding the optimum processing method (3, 5, 10, 16). Raw milk is often used because it provides, unaltered, all the protective constituents of milk (12, 14, 19, 22). Unfortunately, it may also be a source of infection, as demonstrated by reports linking intensive care nursery outbreaks of *Salmonella kottbus* (25), *Escherichia coli* (29), *Serratia marcescens* (13), and *Klebsiella* spp. (8) to ingestion of contaminated human milk.

Human milk usually contains coagulase-negative staphylococci, alpha-hemolytic streptococci, and diphtheroids, all representing normal skin flora (5, 9, 28). Gram-negative bacilli have been found in milk from 5 to 15% of women (4–7, 9). Safe limits for bacterial concentration in human milk used to feed premature infants have never been established. Many donor milk programs have adopted arbitrary limits based, in part, on those used by the dairy industry for cow's milk (4, 5, 7, 24, 26, 30). Rather than testing every milk sample, most milk banks have adopted a screening program whereby quantitative cultures of milk samples from each donor are done on a regular basis. Whenever the bacterial concentration is found to exceed the chosen limits, all milk from a given donor is held until repeat culture results are acceptable. It is unknown whether such screening programs succeed in ensuring that premature infants will not be fed milk containing excessive bacteria. Accordingly, we initiated a prospective surveillance study to assess the effectiveness of screening procedures used by the donor milk program at the Health Sciences Centre (HSC), Winnipeg, Manitoba, Canada. A brief description of the routine screening program is necessary before outlining the specific study objectives.

HSC bacteriologic routine for screening donor milk. Quantitative cultures of milk from each donor are done at the outset of donations and then once weekly for as long as donations continue. The quantitative limits of detection are 10^0 to 10^8 CFU/liter. The limits for acceptable bacterial concentration in milk are as follows: $\leq 10^8$ CFU of coagulase-negative staphylococci per liter; $< 2 \times 10^7$ CFU of viridans group streptococci or diphtheroids per liter; $< 10^5$ CFU of *Staphylococcus aureus*, *Streptococcus* (*Enterococcus*) *faecalis*, pneumococcus, or gram-negative aerobic bacilli; and no beta-hemolytic streptococci. These were originally adapted from the standards used by the dairy industry for cow's milk. Milk which exceeds these limits is handled differently according to whether it is donor or maternal milk. Donor milk refers to expressed milk from a woman who is not the infant's mother, whereas maternal milk is from the infant's own mother. Donor milk is derived from two sourc-

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es: women already providing milk for their own premature infants (preterm donors) as well as volunteers who have delivered healthy full-term infants (term donors). Maternal milk which exceeds standards can still be used to feed a woman's own infant. In contrast, whenever excessive bacterial concentration is found in milk from a term donor, all milk from that donor is discarded and no further donations are used until repeat cultures indicate an acceptable bacterial content. The milk of a preterm donor can still be given to her own infant but not to other unrelated infants in the nursery. In all cases, repeat instruction is given in proper techniques of milk expression and cleaning equipment.

Specific study objectives. The prospective surveillance study was conducted from February 1986 to April 1987. Specific objectives were (i) to describe, in both qualitative and quantitative terms, the daily pattern of exposure to milk-associated bacteria among premature infants fed fresh expressed human milk; (ii) to relate the species and absolute numbers of bacteria ingested to potential adverse outcomes, including feeding intolerance and invasive infection; and (iii) to assess the effectiveness of the HSC routine screening program by testing the following predictions: (a) donor milk fed to premature infants in the HSC nursery should contain less bacteria than either maternal milk used in the same nursery or donor milk used in a second intensive care nursery (St. Boniface Hospital [SBH], Winnipeg, Manitoba, Canada) which lacked a routine screening program and (b) there would be no difference in bacterial content among maternal milks used in the HSC and SBH nurseries.

MATERIALS AND METHODS

Enrollment. The study was conducted for a minimum period of 1 year to allow for seasonal variation in bacterial content of expressed milk. Infants were enrolled if they weighed <2,000 g at birth and were admitted to either of the intensive care nurseries at HSC or SBH before the onset of any enteral feeding; informed consent was obtained from the legal guardian(s).

Sample collection. For the first 2 weeks of feeding, samples of all human milk fed to study infants were collected for quantitative culture. After a brief 10- to 15-min warming period, 0.5- to 1.0-ml samples of each milk feed were set aside in sterile labeled containers. Samples were left at room temperature for an additional 15 to 30 min while the infants were fed and then were either refrigerated at 4°C (Sunday through Thursday) or frozen at −4°C (Friday and Saturday). Samples were transported to the research laboratory by the study nurse on a daily basis (Monday through Friday) and cultured quantitatively on arrival. Throat and rectal swabs were obtained from all study infants once before and several times during the monitored feeding interval. Feeding diaries detailing the time, volume, and source of each feed, as well as episodes of feeding intolerance, were kept for all study infants. Feeding intolerance was defined as withholding a scheduled feeding owing to one or more of the following: vomiting, abdominal distension, diarrhea, or retention of 25% or more of the volume of a previous feed. Episodes of bacteremia among hospitalized study infants were recorded, and the isolates were saved.

Bacteriology. Milk samples were cultured by plating 0.1- and 0.001-ml portions onto both blood and MacConkey agar. Throat and rectal swabs were streaked onto similar media for semiquantitative assessment of aerobic bacterial species. Plates were incubated at 37°C overnight in room air. All discrete morphologically distinct colonies were subcultured, and species were subsequently identified by standard microbiologic techniques. API 20E strips (Analytab Products) were used for final identification of gram-negative species. All *S. aureus*, coagulase-negative staphylococci, and gram-negative species were stocked and saved by freezing at −70°C in skim milk. For milk cultures, quantitative limits of detection were 10^4 to 10^8 CFU/liter.

Antimicrobial susceptibilities. A standard agar dilution technique employing Mueller-Hinton agar was used to determine the antibiotic susceptibilities of selected bacterial isolates (20).

Analysis. For each infant, the numbers and absolute quantities of bacterial species ingested as part of the first 2 weeks of human milk feedings were determined. The association between the quantity of bacteria ingested and subsequent feeding intolerance was assessed for coagulase-negative staphylococcal species and gram-negative aerobic bacteria.

The effectiveness of the HSC screening program was assessed by testing the following null hypotheses. (i) The bacterial content of donor and maternal milk fed to HSC infants is the same. (ii) The bacterial content of donor milk fed to HSC and SBH infants is the same.

To simplify analysis, the results of the surveillance cultures were summarized by using broad categories of bacterial content as follows: any bacterial species, any gram-positive species, any coagulase-negative staphylococcal species, any *S. aureus*, and any gram-negative species. Within each category, the proportion of milk with any bacteria detected, as well as the proportion containing bacteria in excess of the upper concentration limits proscribed by the routine screening program, were determined. The mean bacterial content of maternal, term donor, and preterm donor milk was analyzed as a proportion of the total volume fed to all study infants from each source.

Standard statistical techniques were used to compare characteristics of the study infants and of the women providing milk, as well as the mean bacterial content of donor and maternal milk.

RESULTS

Study population characteristics. Expressed breast milk was provided by 96 women during the study period, of whom 66 (69%) were mothers of study infants. The mean duration of pregnancy was 30 ± 3 weeks. In addition to expressing milk for their own infants, 38 of these women provided preterm donor milk for unrelated study infants. There were 30 volunteer term donors, all of whom had uncomplicated pregnancies. There was no age difference between the two groups of women. Milk was expressed manually without a pump by 4% of mothers of study infants and 57% of volunteer donors. An electric or hand pump was used, respectively, by 94 and 17% of mothers of study infants and 13 and 33% of volunteer donors.

A total of 102 infants were enrolled in the study; 58 from HSC and 44 from SBH. There were no significant differences between the infants in the two nurseries in mean gestational age, birth weight, or duration of stay. The values (mean ± standard deviation) for the entire group for gestational age, birth weight, or duration of stay were 29.8 ± 2.8 weeks, 1,251 ± 300 g, and 69.3 ± 30.3 days, respectively. The ratio of males to females was 1:1.2. Four infants died, three (2 from HSC, 1 from SBH) before being fed and one HSC infant, who had severe congenital heart disease, after discharge home. All other infants were included in the analysis, except for one SBH infant who was switched to formula within 48 h of starting feedings.

TABLE 1. Distribution of unique bacterial species in 7,610 milk samples provided by 96 women

Species	% of all milk samples containing indicated species	% of women with species in ≥1 milk sample	Mean % of milk samples positive for indicated species from women with ≥1 positive
Gram positive			
Coagulase-negative staphylococci	77.0	95	79.1
Streptococcus viridans	5.6	43	16.0
Staphylococcus aureus	4.1	21	22.3
Nonhemolytic streptococci	1.5	12	11.0
Diphtheroids	0.2	6	3.4
Bacillus sp.	0.1	3	2.1
Group B beta-hemolytic streptococci	1.1	1	81.0
Gram negative			
Acinetobacter sp.	3.4	33	14.1
Pseudomonas fluorescens group	2.1	19	8.6
P. maltophila	<0.1	3	3.0
P. putida	<0.1	2	2.7
P. stutzeri	<0.1	1	10.4
P. cepacia	<0.1	1	0.5
Pseudomonas spp.	0.1	4	2.6
Klebsiella pneumoniae	0.4	5	6.5
K. oxytoca	0.7	7	6.6
K. ozaenae	0.2	1	54.2
Escherichia coli	0.2	6	3.8
E. hermannii	0.4	2	14.2
Enterobacter cloacae	1.0	12	5.8
Enterobacter agglomerans	0.1	6	2.5
Serratia liquefaciens	0.1	4	7.7
S. marcescens	<0.1	2	7.1
Serratia sp.	<0.1	1	14.3
Chromobacterium species	0.1	4	4.0
Moraxella spp.	0.2	4	4.1
Flavobacterium meningosepticum	0.1	1	37.5
F. odoratum	0.1	2	3.4
Cedacea lepagei	<0.1	2	1.6
CDC groups (VE1, VE2, 17, and 41)	<0.1	4	2.2
Yersinia enterocolitica	<0.1	1	1.2

Exposure to bacteria among premature infants fed raw human milk. Samples from 10,128 separate milk feedings were sampled for quantitative culture just before being fed to the study infants. These samples represented 84% of all milk feeds given to the study infants during the first 2 weeks of feeding. The proportional distribution of bacteria among all feeds was: no detectable growth in 19.1%, only gram-positive bacteria in 74.3%, only gram-negative bacteria in 1.1%, and mixed gram-positive and gram-negative species in 5.7%. The proportion of all infant feedings in which the concentration of bacteria exceeded the limits proscribed by the donor milk program was 25% for gram-positive bacteria and 4.7% for gram-negative bacteria.

A specific donor source could be identified for 75% (7,610) of human milk feedings. The distribution of unique bacterial species among all women expressing milk and all milk samples tested is shown in Table 1. As expected, the majority of milk samples (77%) contained coagulase-negative staphylococci. The prevalence of all other species ranged from 0.01 to 5.6% of samples tested. Gram-negative bacteria were present in ≥1 milk sample from 51 (53%) of 96 women providing milk. Of women with at least one milk sample containing gram-negative bacteria, 16 (31%) had only a single species recorded, 15 (29%) had two species, 7 (14%) had three species, 6 (12%) had four species, 4 (8%) had five species, 2 (4%) had six species, and 1 had nine unique species recovered. There was no discernible temporal pattern for gram-negative content in milk from any donors. The means and ranges of concentrations for the most prevalent bacterial species in individual feedings are shown in Table 2.

Events related to ingestion of milk-associated bacteria. Table 3 summarizes the frequency with which infants ingested milk-associated coagulase-negative staphylococci, *S. aureus*, or gram-negative aerobic species during the monitored feeding interval. There were surprisingly few, if any, adverse events that could be directly related to ingestion of bacteria in raw breast milk. During the study interval, an episode of feeding intolerance followed 2% of the feeds. The quantity ($\log_{10}$ CFU; mean ± standard deviation) of coagulase-negative staphylococci or gram-negative bacilli ingested in the feeds immediately preceding an episode of feeding intolerance (5.6 ± 1.1 for coagulase-negative staphylococci; 4.8 ± 1.2 for gram-negative bacilli) was not significantly different from that ingested in the feeding before tolerated feeds (5.5 ± 0.9 for coagulase-negative staphylococci; 4.8 ± 1.2 for gram-negative bacilli). During the study interval,

TABLE 2. Bacterial species isolated from raw breast milk fed to premature infants

Bacterial species	No. of samples[a] containing indicated species	Log_{10} CFU/liter in positive samples[b]		% of samples exceeding limits[c]
		Mean ± SD	Range	
Coagulase-negative staphylococci	5,847	7.2 ± 0.9	4–>8	28
Streptococcus viridans	423	5.6 ± 0.9	4–>8	3
Staphylococcus aureus	307	6.8 ± 0.8	4–>8	94
Nonhemolytic streptococci	117	6.5 ± 1.0	4–>8	25
Group B beta-hemolytic streptococci	85	7.0 ± 0.5	6–>8	100
Acinetobacter sp.	232	6.5 ± 1.1	4–>8	89
Pseudomonas fluorescens group	116	6.1 ± 1.2	4–>8	83
Klebsiella pneumoniae	27	6.4 ± 0.8	4–7	89
K. oxytoca	54	5.1 ± 0.7	4–7.5	91
K. ozaenae	13	7.5 ± 0.9	6–>8	100
Escherichia coli	14	5.5 ± 0.8	4–6.3	72
E. hermannii	31	5.0 ± 0.2	5–6	100
Enterobacter cloacae	74	5.3 ± 0.8	4–7.5	76
E. agglomerans	11	4.7 ± 0.6	4–6	45

[a] Each sample represents one feeding given to one infant from a discernible maternal or donor source (total of 7,610 samples).
[b] For milk samples containing $>10^8$ CFU of a given species per liter, an arbitrary value of 8.1 was used to calculate the mean.
[c] The limits are as proscribed by the HSC donor milk program (see text).

bacteremia was documented in 10 infants. Coagulase-negative staphylococci were recovered in eight episodes, a nontypeable *Haemophilus influenzae* was recovered in one episode, and *Klebsiella oxytoca* was recovered in another. Timing and comparative antimicrobial susceptibility testing suggested that none of these episodes could be directly related to exposure to bacteria present in raw milk. The episode of nontypeable *H. influenzae* occurred in a 1-day-old infant before onset of oral feedings. The infant with *K. oxytoca* bacteremia had documented rectal colonization by *K. oxytoca* before being fed breast milk. All eight bacteremic isolates of coagulase-negative staphylococci were multiply antibiotic resistant (defined as resistant to gentamicin, oxacillin, and at least one other antibiotic using NCCLS [20] guidelines), whereas all the milk isolates fed to the infants before the bacteremic episode were multiply antibiotic susceptible.

TABLE 3. Frequency and intensity of ingestion of milk-associated bacteria by 98 premature infants during the first 2 weeks of oral feeds

Bacterial species	% of babies with >1 ingestion	% total feeds containing indicated species	Mean log_{10} CFU (SD) ingested per feed
Coagulase-negative staphylococci	100	39.0	5.9 (0.5)
S. aureus	41	2.4	5.1 (1.0)
Any gram-negative aerobe[a]	64	5.2	4.8 (1.1)

[a] The specific gram-negative species present in expressed milk are shown in Table 1. The values for unique gram-negative species ranged from 2 to 12% babies with >1 ingestion, 0.1 to 0.6% total feeds containing indicated species, and 2.5 to 4.8 for mean log_{10} CFU ingested per feed.

Data linking ingestion of milk-associated bacteria to subsequent gastrointestinal colonization among study infants are being presented in greater detail as a separate communication. A brief summary of the results is provided because they have implications regarding the need for screening. Multiply antibiotic resistant coagulase-negative staphylococcal species colonized the majority of infants before initiation of feeding, whereas <10% of a sample of 433 milk coagulase-negative staphylococcal isolates were multiply resistant. The sample was chosen to include 10% of all coagulase-negative staphylococcal isolates recovered from the milk of each of 91 donors. Selection was not random but designed to include isolates recovered from each woman throughout the donation interval. These results suggest that colonization of study infants by milk-associated coagulase-negative staphylococcal species, if it occurred, had no clinical significance. The results for milk-associated gram-negative bacilli were less reassuring, in that colonization of the gastrointestinal tract by 11 bacterial species may have occurred after ingestion of the same species in 48% of 62 babies exposed (B. Law, J. Lertzman, B. Urias, L. Romance, and D. Robson, Program Abstr. 28th Intersci. Conf. Antimicrob. Agents Chemother., abstr. no. 681, 1988).

Effectiveness of the routine HSC milk screening program. The comparative distribution of bacteria in maternal milk versus term and preterm donor milk when calculated as a proportion of the total volume fed from each source is shown in Fig. 1. The total volumes of milk ingested during the first 2 weeks of feeding by infants in the HSC and SBH nurseries were 98 and 57 liters, respectively. The proportions of total milk fed which came from a maternal, term donor, or preterm donor source were 59, 26, and 5%, respectively, for HSC infants and 54, 2, and 13%, respectively, for SBH infants. A specific source could not be assigned for 10 and 31% of the volumes fed to HSC and SBH infants, respectively. These included feedings in which unknown proportions of donor and maternal milk were mixed or in which frozen milk from donors not enrolled in the study was used.

The bacterial content of HSC donor milk was significantly less than that of HSC maternal milk in terms of the proportionate volume containing any bacteria (50% for all donors and 95% for mothers [$P < 0.0001$]) as well as the proportionate volume with a concentration exceeding the upper limits proscribed by the routine screening program (12% for all donors and 32% for mothers [$P \leq 0.0005$]) (Fig. 1A). The same trend was observed when comparing HSC preterm donor milk to SBH preterm donor milk (for any bacteria, 36% for HSC and 81% for SBH [$P \leq 0.0001$]; for excess bacteria, 12% for HSC and 34% for SBH [$P = 0.02$]). The bacterial contents of HSC and SBH maternal milk were the same for any quantity of bacteria (95% for HSC and 96% for SBH [$P > 0.5$]) but differed for the proportionate volume which exceeded the limits (32% for HSC and 52% for SBH [$P = 0.01$]).

The differences between HSC donor milk and milk from all other sources could be accounted for by differences in the content of coagulase-negative staphylococci (Fig. 1B). In contrast, there were no significant differences between donor and maternal milk from either nursery with respect to content of *S. aureus* (Fig. 1C) or gram-negative aerobic species (Fig. 1D). Among HSC donor milk the prevalence of gram-negative species was lower in milk from preterm donors than from term donors (1.2 versus 6.1% [$P < 0.05$]).

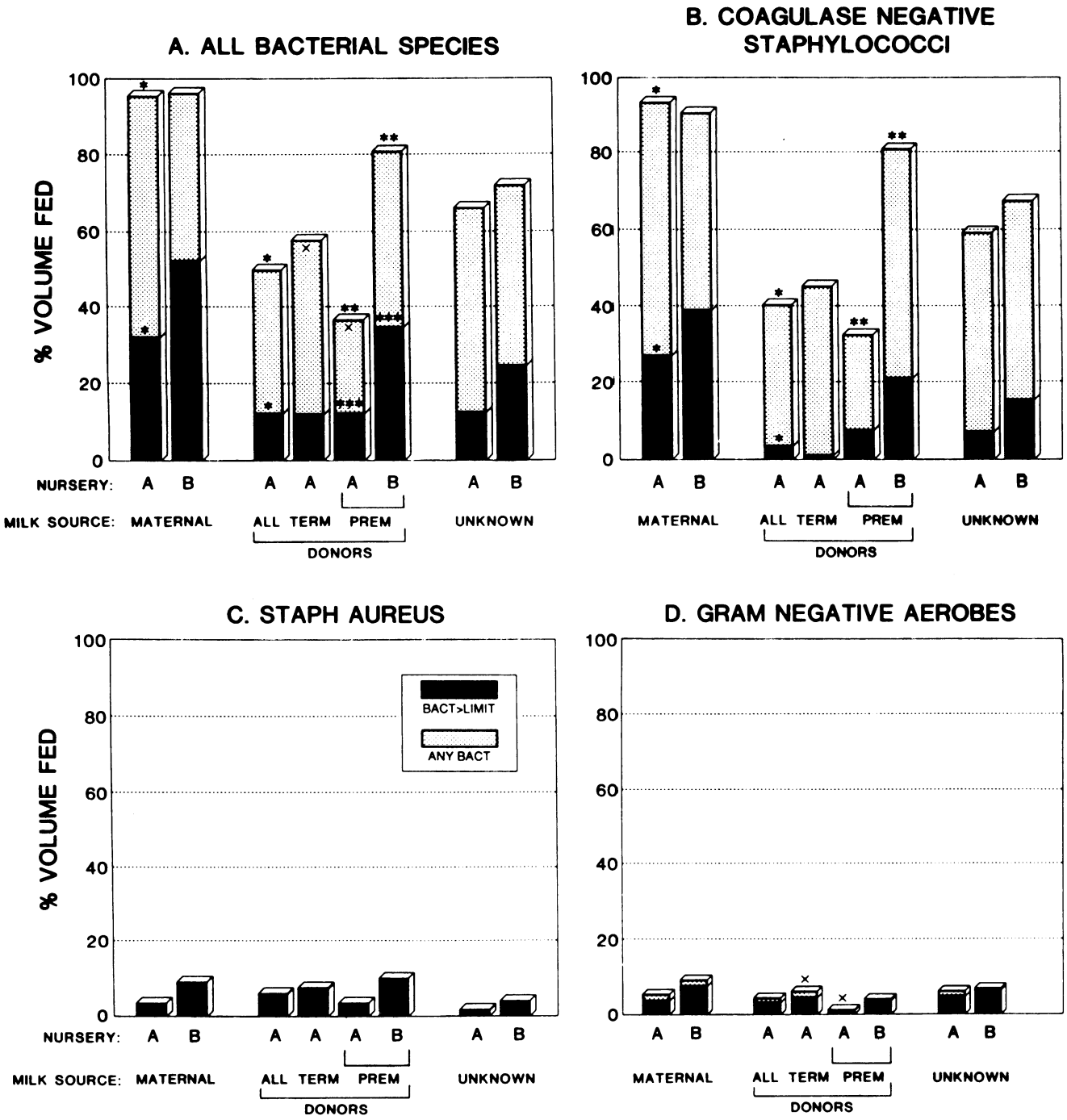

FIG. 1. Comparison of the bacterial contents of raw breast milk ingested by HSC (A) and SBH (B) infants according to whether the milk originated from an infant's mother, an unrelated donor, or an unspecified source. For HSC donors, results are given for the combined group, as well as for term and preterm donors. There were no term donors at SBH. Results are expressed as the mean proportion (percentage) of the total volume of milk fed from a given source which contained the bacterial species as indicated in any quantity or in concentrations which exceeded the arbitrary limits as outlined in the text. Symbols: *, $P < 0.0005$ for HSC donors versus HSC mothers; ×, $P < 0.01$ for term donors versus preterm donors at HSC; **, $P < 0.0001$ and ***, $P < 0.02$ for preterm donors at HSC versus preterm donors at SBH.

The study design did not allow for further analysis to explain this difference.

DISCUSSION

These data clearly document that premature infants fed raw expressed human milk are frequently exposed to large numbers of milk-associated coagulase-negative staphylococci whereas the exposure to *S. aureus* and gram-negative aerobic bacteria is less frequent and of lower magnitude. Surprisingly, we were unable to document any adverse events that could be directly related to ingestion of bacteria in raw breast milk. Nearly all of the ingested milk coagulase-

negative staphylococcal isolates were multiply antibiotic susceptible, whereas the majority of study infants were colonized with multiply resistant coagulase-negative staphylococci before being fed for the first time (B. Law et al., Abstr. 55th Conjoint Meet. Infect. Dis., p. A-1, 1987). Feeding intolerance could not be attributed to ingestion of either coagulase-negative staphylococci or gram-negative aerobes. Bacteremia also could not be attributed to ingestion of coagulase-negative staphylococci or gram-negative aerobes, but the number of infants studied was small. The potential for invasive gram-negative infection was demonstrated by the observation that colonization of the gastrointestinal tract may have occurred after exposure to milk-associated gram-negative bacilli in as many as 48% of study infants. These observations provide a basis for the previously reported intensive care nursery outbreaks of gram-negative sepsis in which contaminated milk was implicated as a source.

The routine screening program was found to be only partially successful in limiting the bacterial content of raw donor milk used to feed premature infants. The system appeared to work for bacteria which are frequently present in milk, namely coagulase-negative staphylococci, but was ineffective for less prevalent species, such as *S. aureus* and gram-negative aerobic bacilli.

Two potentially confounding influences in this study must be addressed. For the routine screening program, milk was transported from the nursery refrigerator directly to the microbiology laboratory. In contrast, samples for the surveillance study were handled by the nurses as part of the feeding preparation and were held out of the refrigerator for as long as 1 to 2 h while the infant was being fed. This delay in culturing was necessitated by practical aspects of sample collection in an intensive care setting. We justified this aspect of methodology on the basis of evidence that raw milk is bacteriostatic and that the concentration of bacteria in the milk does not change over 6 h at room temperature (24). If contamination during handling or growth of bacteria during a delay in processing were significant, the impact should have been equal on both maternal and donor samples. Furthermore, the occurrence of exogenous contamination would not invalidate conclusions regarding screening program effectiveness. Whether bacteria are present at the time milk is expressed or contaminate it during subsequent handling, the cost of periodic screening cannot be justified if periodic screening does not reduce the chance that infants will be exposed to milk-associated pathogens.

Variation in methods of expression may have influenced the observed differences in bacterial content between maternal and donor source milk. All but one of the mothers of study infants used electrical pumps to express milk, whereas the majority of volunteer donors used non-pump-assisted manual expression. The use of pumps is more frequently associated with the introduction of gram-negative contaminants than is manual expression (19). Since preterm donors were more likely to use electrical pumps than term donors, one would have expected gram-negative contaminants more frequently in the former group when in fact, the opposite was observed.

In conclusion, the content of coagulase-negative staphylococci in raw donor milk fed to premature infants can be reduced by periodic screening, but no clinical advantage is gained. In contrast, screening has no impact on milk-related exposure of infants to aerobic gram-negative species or *S. aureus*. There was no temporal pattern to the intermittent recovery of pathogens among milk samples from a given individual. Thus, the only way to eliminate these chance occurrences would be to remove a sample from each separate expression and perform a quantitative culture while storing the rest of the donated milk at 4°C. The usual maximum refrigerator shelf life for raw milk of 48 to 72 h would preclude this practice. Freezing most of the sample while awaiting quantitative culture results of a test sample has been advocated but is costly and impractical (7). Heat treatment of milk will eradicate bacteria but does not eliminate the possibility of subsequent contamination due to handling of the milk before feeding the infant. Furthermore, it has been demonstrated that pasteurized milk supports bacterial growth more readily than does raw milk (14). Finally, the fact that large quantities of bacteria, especially coagulase-negative staphylococcal species, were ingested daily by most study infants with no apparent ill effect suggests that processing milk to eliminate such bacteria is unnecessary.

On the basis of our results, we have discontinued the HSC donor milk bacteriologic screening program. Educating both donors and mothers of premature infants in proper techniques of expressing, handling, and transporting milk is continued, as is screening to ensure that all donors are seronegative for cytomegalovirus, hepatitis B, and human immunodeficiency virus. Milk donations are interrupted whenever there is clinical evidence of mastitis. Milk is cultured only when it is suspected to be a source of neonatal sepsis. Our results do not support attempts to define a safe upper limit for bacterial concentration in raw expressed milk. For units using human milk to feed premature infants, the decision to be made should be whether to use fresh, frozen, or pasteurized milk, rather than how to screen or what limits to apply regarding bacterial content. The risk of sepsis after ingestion of contaminated human milk is unknown but probably very low. Whether the risk is acceptable in the North American intensive care nursery setting is unknown and will remain so until a large multicenter prospective study is carried out comparing the risks and benefits of feeding premature infants human milk versus sterile formula. In the meantime, the data presented here do not support adoption of routine bacteriologic screening programs for intensive care units using raw milk to feed premature infants.

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