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Breast Milk Stem Cells: Current Science and Implications for Preterm Infants

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Breastmilk Stem Cells: Current Science and Implications for Preterm Infants

Abstract

Background. The benefits of breastmilk are well described, yet the mechanistic details related to how breastmilk protects against acute and chronic diseases, and optimizes neurodevelopment remain largely unknown. Recently, breastmilk was found to contain stem cells that are thought to be involved in infant development.

Purpose. The purpose of this review was to synthesize all available research involving the characterization of breastmilk stem cells to provide a basis of understanding for what is known and what still needs further exploration.

Methods/Search Strategy. The literature search was conducted between August and October 2015 using CINAHL, PubMed, and reference list searching. Nine studies addressed characterization of human breastmilk stem cells.

Findings/Results. Five research teams in four countries have published studies on breastmilk stem cells. Current research has focused on characterizing stem cells in full-term breastmilk. The amount, phenotype, and expression of breastmilk stem cells is known to vary between mothers and they have been able to differentiate into all three germ layers (expressing pluripotent characteristics).

Implications for Practice. There is much to learn about breastmilk stem cells. Given the potential impact of this research, healthcare professionals should be aware of their presence and ongoing research to determine benefits for infants.

Implications for Research. Extensive research is needed to further characterize stem cells in breastmilk (full-term and preterm), throughout the stages of lactation, and most importantly, their role in the health of infants, and potential for use in regenerative therapies.

Key Words (5-10): Breastmilk, Breast milk, Stem Cells, Development, Infant feeding, Pluripotency, Multipotency, Preterm Infants, Neonatal Intensive Care Unit

Background

The benefits of breastmilk are well described, yet the mechanistic details related to how breastmilk protects against chronic diseases and optimizes neurodevelopment remain unknown. Recently, breastmilk was found to contain stem cells which may be a source for cellular interactions or secreted products that foster childhood health. As such, they have been hypothesized to be involved in infant growth and development.[1-3] The presence of breastmilk stem cells provides an opportunity to consider new pathways for describing the mechanisms that underlie the significance of breastmilk in early life, and the potential to explain how breastmilk may positively influence childhood and adult health, especially in the preterm infant. See **Table 1** for definitions of words used throughout this manuscript.

Breastmilk is Different for Full-term and Preterm Infants

Breastmilk is recognized as the ideal feeding type for all infants due to its known nutritional and immunological properties.[4] As research methods advance, our knowledge of the science and mechanisms supporting the benefits of breastmilk continues to grow. Researchers have recognized that breastmilk is individualized for each mother-infant dyad to meet specific needs of the infant based on a variety of factors such as infant illness, gestation, post-natal age and gender.[1,5-7] For example, the breastmilk of mothers whose infants have active infections is known to contain increased numbers of infection-fighting white blood cells.[6] Mothers whose infants are born prematurely are also known to have breastmilk with different composition (e.g. micro and macronutrients) than mothers of full-term infants.[8-11] Additionally, lactation research in Holstein cattle have found that more milk is produced for female calves than males.[5] Considering the hypothesis that breastmilk stem cells are involved in infant growth and

development, and the known variability in other aspects of breastmilk composition, suggests that there may be differences in the stem cell content between full-term and preterm milk.

Overview of Stem Cells

Human embryonic stem cells (hESCs) are derived from blastocysts (a 3 to 5 day old embryo).[12] These stem cells are the most versatile because they can form any type of cell and tissue within the body due to their normal function to grow from embryo to fetus (ectoderm, mesoderm, and endoderm are the three germ layers).[12] Once stem cells are directed to a particular lineage, our body uses them to regenerate cells and tissues as part of normal growth and repair. At this stage they typically cannot deviate from their assigned cell type. For example, adult cells are typically only able to replicate into the specific cell or tissue lineage they are from (e.g. skin cells can only replicate into other skin cells). However, scientists have found ways to induce pluripotency (ability to replicate into other cell lines) among some adult cells for possible use in regenerative medicine.[12]

Stem Cells in Breastmilk

Breastmilk stem cells were first described in 2007 by a team of scientists in Australia.[13] This initial study analyzed breastmilk from mothers of full-term infants and found that nestin positive stem cells were present.[13] Nestin is a well-known marker for neural, bone marrow, pancreatic, and epithelial stem cells.[13-18] Further research indicated that breastmilk stem cells exhibit characteristics of pluripotent stem cells.[19] This finding indicates that like hESCs, they have the potential to differentiate into all of the cells and tissues in different body organs.[1,19]

Stem cell research from sources other than breastmilk provides a variety of uses in regenerative medicine. Currently, stem cells are used in cell replacement therapies, cell renewal, and many ongoing research trials that seek to expand the use of stem cells in the prevention and

treatment of disease.[20] The potential of stem cells is immense and finding them within breastmilk promises a new mechanism of action to consider when exploring how to garner and allocate their beneficial properties. Many of the teams who have studied breastmilk stem cells have agreed that these cells may have the potential to be used in regenerative medicine and bioengineering.[2,13,19,21-25] Researchers hypothesize that following ingestion, breastmilk stem cells interact with the infant to promote tissue homeostasis, development, and overall regeneration.[2] Breastfeeding allows maternal cells (e.g. leukocytes) to spread throughout the infant's body in a process known as microchimerism.[26,27] This mechanism prompted scientists to explore similar actions of breastmilk stem cells, and using mouse models they found that ingested breastmilk stem cells survive the gastrointestinal tract and spread throughout the body where they are then found as differentiated cells within different organs of the mouse pup.[28,29] This finding has the potential to explain some of the short and long-term effects attributed to being breastfed as an infant but further investigation is needed to substantiate this linkage. Further understanding of breastmilk stem cells shows promise in at least two different areas. The first of these is the potential that breastmilk stem cells are actively involved in supporting infant growth and development (both short and long term) through mechanisms that are not yet fully understood. For example, it is not fully understood how breastmilk ingestion in early life can decrease disease later in life (clear mechanistic linkages do not exist). These mechanisms might be explained through breastmilk stem cells. The second is the possibility that breastmilk stem cells could be used in regenerative medicine to replenish and restore damaged tissues. These findings could be particularly important for preterm infants, whose vulnerabilities (e.g. underdeveloped organs and high risk for morbidities) make their needs particularly great.

Laboratory Techniques

Breastmilk stem cells have been analyzed using various methods. The methods to characterize breastmilk stem cells provide information on the cell phenotype and expression, amount of cells present, and the viability of cells. Some of the most common methods of analysis include immunofluorescence, quantitative reverse transcription polymerase chain reaction (qRT-PCR), and flow cytometry. Each of these methods works to identify whether certain markers are expressed in a cell population and use different mechanisms to tag these genes of interest (**See Table 1**). Typically breastmilk is collected fresh and then brought directly to the laboratory for processing. Scientists can then examine the freshly expressed breastmilk for the presence of specific genes, which are likely most representative of the cells that are ingested by the infant if the breastmilk had been used in enteral feeding. The breastmilk can also be cultured, which is a process of extracting cells from breastmilk and placing them in a culture dish with specific growth medium to encourage cellular growth. Many scientists will grow the cells until they are confluent, which means that they fill the plate they are growing on, and will then proceed with analyzing these cells to determine the presence of different stem and other cell characteristics.

Purpose

As the scientific evidence accumulates regarding breastmilk stem cells, scientists have hypothesized about their potential to influence infant health. There are published reviews on the implications of breastmilk stem cells,[1,3] however, despite the mounting literature, no known publications provide a concise accumulation of all original articles on human breastmilk stem cell research. The purpose of this review was to synthesize all available original research focused on characterizing human breastmilk stem cells.

Methods/Search Strategy

The literature search was conducted between August and October 2015 and no restrictions were placed on year of publication. The electronic databases of PubMed and the Cumulative Index for Nursing and Allied Health Literature (CINAHL) were searched for potential articles using the search terms of *Human milk, Breastmilk, Stem Cells,* and limited to research on human breastmilk.

Study Selection and Data Collection Process

Articles were evaluated to determine if they analyzed human breastmilk for the presence of stem cells. The outlined search strategy resulted in 12 articles from CINAHL and 29 articles from PubMed (see **Figure 1**) for a total of 41 publications examined for the review**.** After title and abstract review 29 articles were eliminated for not meeting inclusion criteria (original research on breastmilk stem cells) – leaving 12 articles for full manuscript review. Thorough evaluation of those 12 studies resulted in the elimination of three studies due to their lack of analyzing human breastmilk samples for stem cells (they were focused on other aspects of breastmilk cells, and transplant tolerance after receiving breastmilk). The remaining nine studies were extensively reviewed for findings on characterization of breastmilk stem cells. The reference lists of included articles were reviewed for additional eligible studies, but no new studies were identified through this method. Additionally, we present our research focused on the preterm infant, a first in this line of research.

Findings/Results

Nine studies were reviewed that specifically addressed breastmilk stem cells and are summarized in **Table 2**. The years that studies were published ranged from 2007 until 2015 (**Figure 2**), and the existing research evolved primarily from four large research teams in four

countries (Australia, Singapore, Iran, India). One additional study of our team in the United States is also presented in Table 2 because of its focus on the preterm infant.

Breastmilk Contains Stem Cells

Cregan et al. were the first to investigate the presence of stem cells in breastmilk in 2007.[13] These scientists evaluated breastmilk for the presence of a mammary stem cell marker, mature epithelial markers, and nestin (a multipotent stem cell marker). Their findings indicated that breastmilk contains nestin positive cells which indicates that there are multipotent stem cells in breastmilk. This groundbreaking work sparked an interest in breastmilk stem cell research across the world.

Other teams worked to define the characteristics of breastmilk stem cells and Fan and colleagues also described these cells' potential for self-renewal and differentiation through the identification of various stem cell marker types .[21] Indumathi and colleagues found that there were varying amounts of different stem cells within breastmilk[23]. Additionally, their findings differed from other teams in that their samples had limited numbers of mesenchymal stem cells (a type of multipotent stem cell derived from the mesoderm that can differentiate to a variety of cell lines such as those found in tissue of the umbilical cord[12]), and a large number of hematopoietic stem cells, which are the cells that produce all blood cell lines.

Breastmilk Stem Cells Can Differentiate into all Three Germ Layers

Once it was established that stem cells were present in breastmilk, researchers began to characterize these cells. Patki et al. were the first team to successfully differentiate breastmilk stem cells into different cell lines.[30] They were able to differentiate these stem cells into adipogenic (fat), chondrogenic (cartilage), and osteogenic (bone) lineages. They also began to characterize the presence of stem cells in breastmilk and found that 10-15% of cells in breastmilk

are stem-like and that this number increases significantly after time in culture. These researchers confirmed that breastmilk stem cells in their study were mesenchymal, and as such, have the potential to be differentiated and used in regenerative medicine. These scientists also suggested that breastfeeding, by its very nature, is a stem cell therapy. Specifically, they hypothesized that breastmilk stem cells are ingested by the infant and have the ability to interact with the infant's normal cells, influencing their development. See **Figure 3** for our model which helps show the theorized method of stem cell dispersion throughout the body after ingestion.

Additionally, Hosseini and colleagues' (2014) findings support these results when they found that breastmilk stem cells expressed both mesenchymal and embryonic stem cells markers, and were differentiated into the three neural lineages (Oligodendrocytes, astrocytes, and neurons), which provides promise for further understanding on how breastmilk positively affects neurodevelopment.[31] This research team also described the presence of hESC markers that were common among other research study findings (e.g. TRA-1-60, NANOG, OCT4), but unlike some of the prior studies, they did not find expression of a marker commonly associated with hESCs (SSEA1/4).[24] This research team was also able to differentiate the breastmilk stem cells into both osteogenic (bone) and adipogenic (fat) lineages.

The team from the University of Western Australia (Hassiotou e al., 2012) also found that breastmilk stem cells express similar genes to that of pluripotent human embryonic stem cells (hESCs). [19] Additionally, they were able to differentiate the stem cells into the three germ layers, which is characteristic of pluripotency. Yet, despite these findings, they were unable to form tumors in immunocompromised mice (teratoma assays), which is the standard to prove pluripotency[19]. However, the authors recognize that studies of other adult cells with pluripotent features sometimes do not form teratomas, but are still able to differentiate into damaged tissue.

They propose that this characteristic is positive and indicates that these cells have good potential for use in regenerative medicine (due to the fact they will not form tumors).

Breastmilk Stem Cells Have Increased Levels of Pluripotent Genes Compared to the Resting Breast

Hassiotou and colleagues has also specifically focused on breast cancer and examined breastmilk cells as a comparison of stem cell gene expression.[32] Both breastmilk stem cells and breast tumors had up regulated expression of pluripotency genes when compared to the resting (non-lactating) breast. Markers associated with pluripotent characteristics of hESCs were highest in a mother who was pregnant when she provided breastmilk samples. They also found variation in expression levels between mothers and proposed that the variation may be due to the stage of lactation.

Breastmilk Gene Expression Varies by Infant and Maternal Characteristics

In the most recent study, researchers began to examine the variation in breastmilk stem cells between mothers.[33] Twigger and colleagues found that the gene expression of stem cell markers is different dependent on some maternal and infant characteristics. One difference was in the expression of some markers between mothers with infants born at earlier gestational ages when compared to mothers whose infants were born at a later gestational age. Researchers hypothesized this finding might help explain why some mothers of preterm infants have low milk supply (i.e. decreased expression of stem cell markers may mean reduced function of the lactating breast).[33] Additionally, increased maternal BMI was correlated with lower expression of a stem cell marker that is associated epithelial tissue (i.e. lactocytes, possibly explaining low milk supply with maternal obesity). Larger changes in maternal bra cup size from pre-pregnancy were correlated with higher expression of epithelial cell markers. This study was the first to

address the differences in stem cell gene expression between women, and also was the first to identify any specifics on the infants (which included gestational age at birth and infant gender). This study was the first to include mothers of preterm infants, However, it was not clear at what stage of lactation breastmilk collection occurred in relation to preterm birth (median infant age at time of collection was 30.5 weeks *after* birth—i.e. the infant was over 7 months old at time of breastmilk collection), or specifically how many mothers with preterm infants were included (age range of infants was 27.1 - 43.4 weeks gestation with a median of 39.3).[33]

Across all of these studies, there were differences in total cell counts in each mother's breastmilk, and in expression of stem cell and cell markers.[13,19,21,23,32,33] Not all studies identified the sample size or at what stage of lactation women were at when breastmilk collection occurred. Additionally, even though sample sizes seemed adequately large, it is important to note that for some of the analysis methods, only a very small number were analyzed (i.e. as noted in Table 2 with methods such as flow cytometry). As suggested by Hassiotou et al, the differential expression of stem cell genes may be related to stage of lactation and being so, this information is important for future studies to address and report.[32] Additionally, besides the publication by Twigger et al[33], none of the studies report characteristics of the women who provided breastmilk, or characteristics of their breastfeeding infant.

Stem Cells in Preterm Breastmilk

Our team is currently the first known team in the world to investigate breastmilk stem cells from mothers of hospitalized preterm infants.[34] Current research is focused on identifying differences in breastmilk for preterm and full-term infants within the first few weeks of birth. We have established feasibility of recruitment within the intensive care setting and collected specific health information from the infants and mothers to address the gaps in what has previously been

reported on the participants in this field of research. Current data analyses support that preterm breastmilk stem cells are variable between mothers similar to the variability in the full-term mother-infant dyad.[35]

Discussion

Implications for Practice

The benefits of breastmilk are well reported in the literature; however, there is much to learn about breastmilk stem cells. As new evidence develops on the presence of stem cells within breastmilk, it will be important for healthcare professionals to be aware of their presence and to understand their potential impact on infant health. Preterm infants are born before achieving full gestation and typical growth after delivery is different from growth that would have occurred during gestation. In addition to the already known benefits of breastmilk, breastmilk stem cells may be contributing to improved growth and development in preterm infants.

Implications for Research

Breastmilk stem cells were first identified in 2007, and there remains a vast need for extensive research to further characterize their presence in all breastmilk (full-term and preterm), throughout the stages of lactation, and most importantly, their role in promoting and protecting the health of all infants. Due to the paucity of research in this area, there is much to be done in the coming years. Our research team is currently addressing the gap in literature focused specifically on the preterm infant and beginning to address questions related to the influence of breastmilk stem cells on infant health. Exciting work from the University of Western Australia shows through mouse models that breastmilk stem cells survive the neonatal gut and can be found as differentiated cells throughout the mouse pup's body.[28] The mechanism surrounding this expansion is still unknown and has the potential to advance scientific understanding and

facilitate future research endeavors focused on improving infant health. As more information is uncovered on breastmilk stem cells, we can begin to answer newly formed questions: When breastmilk stem cells are ingested, how are they involved with growth and development? More specifically, are they able to target vulnerable areas of the body (i.e. for the preterm infant, the gastrointestinal system, lungs, and brain)? How do NICU practices (e.g. refrigeration/ freezing/ warming) impact breastmilk stem cell viability and impact? Can breastmilk stem cells be used as a source for stem cell therapy (in the infant/in others)? Additionally, the beneficial properties of pluripotent stem cells are often used in the laboratory for a variety of research. As mentioned previously, the human embryonic stem cell is the most versatile type, yet there is controversy of their use. An easily accessible pluripotent-characteristic stem cell from breastmilk may promise potential avenues for all research involving stem cells.

Conclusion

The discovery of breastmilk stem cells has launched a new direction for breastmilk research which has begun to explain in greater detail the beneficial properties of breastmilk. The potential of breastmilk stem cells to explain some of the short and long term benefits of breastmilk ingestion in early life promises new trajectories of research that may expand the future use of breastmilk. To date, research has focused on further characterizing the stem cells in breastmilk and has yet to further delve into the exact mechanism of action that occurs when ingested by an infant. In the future, research will likely begin to focus on the therapeutic potential of these stem cells both when consumed naturally, and after being manipulated in the laboratory.

Figure Captions:

Figure 1. Study Selection Process

Figure 2. Breastmilk Stem Cell Research over Time

Figure 3. Breastmilk stem cells spread throughout the infant's body after ingestion where they differentiate into multiple cell lineages and are believed to be involved in growth and development

References

1. Hassiotou F, Geddes DT, Hartmann PE. Cells in Human Milk: State of the Science. *Journal of Human Lactation.* 2013;29(2):171-182.
2. Twigger A-J, Hodgetts S, Filgueira L, Hartmann PE, Hassiotou F. From Breast Milk to Brains: The Potential of Stem Cells in Human Milk. *Journal of Human Lactation.* 2013;29(2):136-139.
3. Bode L, McGuire M, Rodriguez JM, et al. It's alive: microbes and cells in human milk and their potential benefits to mother and infant. *Advances in nutrition (Bethesda, Md.).* 2014;5(5):571-573.
4. American Academy of Pediatrics. Breastfeeding and the use of human milk. *Pediatrics.* 2012;129(3):e827-e841.
5. Hinde K, Carpenter AJ, Clay JS, Bradford BJ. Holsteins favor heifers, not bulls: Biased milk production programmed during pregnancy as a function of fetal sex. *PLoS ONE.* 2014;9(2):e86169.
6. Riskin A, Almog M, Peri R, Halasz K, Srugo I, Kessel A. Changes in immunomodulatory constituents of human milk in response to active infection in the nursing infant. *Pediatric research.* 2012;71(2):220-225.
7. Hassiotou F, Hepworth AR, Metzger P, et al. Maternal and infant infections stimulate a rapid leukocyte response in breastmilk. *Clinical & Translational Immunology.* 2013;2(e3):1-10.
8. Genzel-Boroviczeny O, Wahle J, Koletzko B. Fatty acid composition of human milk during the 1st month after term and preterm delivery. *European journal of pediatrics.* 1997;156(2):142-147.
9. Montagne P, Cuilliere ML, Mole C, Bene MC, Faure G. Immunological and nutritional composition of human milk in relation to prematurity and mother's parity during the first 2 weeks of lactation. *Journal of Pediatric Gastroenterology & Nutrition.* 1999;29(1):75-80.
10. Dvorak B, Fituch CC, Williams CS, Hurst NM, Schanler RJ. Increased epidermal growth factor levels in human milk of mothers with extremely premature infants. *Pediatric research.* 2003;54(1):15-19.
11. Kovacs A, Funke S, Marosvolgyi T, Burus I, Decsi T. Fatty acids in early human milk after preterm and full-term delivery. *Journal of pediatric gastroenterology and nutrition.* 2005;41:454-459.
12. National Institutes of Health. Stem Cell Basics: Introduction. *Stem Cell Information* 2002; http://stemcells.nih.gov/info/basics/pages/basics1.aspx. Accessed February 20, 2014.
13. Cregan MD, Fan Y, Appelbee A, et al. Identification of nestin-positive putative mammary stem cells in human breastmilk. *Cell Tissue Research.* 2007;329:129-136.
14. Kabos P, Ehtesham M, Kabosova A, Black KL, Yu JS. Generation of neural progenitor cells from whole adult bone marrow. *Experimental neurology.* 2002;178(2):288-293.
15. Lendahl U, Zimmerman LB, McKay RD. CNS stem cells express a new class of intermediate filament protein. *Cell.* 1990;60(4):585-595.
16. Dahlstrand J, Zimmerman LB, McKay RD, Lendahl U. Characterization of the human nestin gene reveals a close evolutionary relationship to neurofilaments. *Journal of cell science.* 1992;103 (Pt 2):589-597.
17. Zulewski H, Abraham EJ, Gerlach MJ, et al. Multipotential nestin-positive stem cells isolated from adult pancreatic islets differentiate ex vivo into pancreatic endocrine, exocrine, and hepatic phenotypes. *Diabetes.* 2001;50(3):521-533.
18. Toma JG, Akhavan M, Fernandes KJ, et al. Isolation of multipotent adult stem cells from the dermis of mammalian skin. *Nature cell biology.* 2001;3(9):778-784.
19. Hassiotou F, Beltran A, Chetwynd E, et al. Breastmilk is a novel source of stem cells with multilineage differentiation potential. *Stem Cells.* 2012;30(10):2164-2174.

20. International Society for Stem Cell Research. The value of stem cells. 2015; http://www.closerlookatstemcells.org/from-lab-to-you/stem-cells-and-research. Accessed January 1, 2016.
21. Fan Y, Chong YS, Choolani MA, Cregan MD, Chan JK. Unravelling the mystery of stem/progenitor cells in human breast milk. *PLoS One.* 2010;5(12):e14421.
22. Thomas E, Zeps N, Cregan M, Hartmann P, Martin T. 14-3-3sigma (sigma) regulates proliferation and differentiation of multipotent p63-positive cells isolated from human breastmilk. *Cell cycle (Georgetown, Tex.).* 2011;10(2):278-284.
23. Indumathi S, Dhanasekaran M, Rajkumar JS, Sudarsanam D. Exploring the stem cell and non-stem cell constituents of human breast milk. *Cytotechnology.* 2013;65(3):385-393.
24. Sani M, Hosseini SM, Salmannejad M, et al. Origins of the breast milk-derived cells; an endeavor to find the cell sources. *Cell Biol International.* 2015;39(5):611-618.
25. Hassiotou F, Hartmann PE. At the dawn of a new discovery: the potential of breast milk stem cells. *Advances in nutrition (Bethesda, Md.).* 2014;5(6):770-778.
26. Zhou L, Yoshimura Y, Huang YY, et al. Two independent pathways of maternal cell transmission to offspring: through placenta during pregnancy and by breast-feeding after birth. *Immunology.* 2000;101(4):570-580.
27. Arvola M, Gustafsson E, Svensson L, et al. Immunoglobulin-secreting cells of maternal origin can be detected in B cell-deficient mice. *Biology of reproduction.* 2000;63(6):1817-1824.
28. Hassiotou F, Heath B, Ocal O, et al. Breastmilk stem cell transfer from mother to neonatal organs. Experimental Biology; 2014; San Diego, USA.
29. Dutta P, Burlingham WJ. Stem cell microchimerism and tolerance to non-inherited maternal antigens. *Chimerism.* 2010;1(1):2-10.
30. Patki S, Kadam S, Chandra V, Bhonde R. Human Breast Milk is a Rich Source of Multipotent Mesenchymal Stem Cells. *Human Cell.* 2010;23:35-40.
31. Hosseini SM, Talaei-Khozani T, Sani M, Owrangi B. Differentiation of human breast-milk stem cells to neural stem cells and neurons. *Neurology research international.* 2014;2014:807896.
32. Hassiotou F, Hepworth AR, Beltran AS, et al. Expression of the pluripotency transcription factor OCT4 in the normal and aberrant mammary gland. *Frontiers in Oncology.* 2013;3:1-15.
33. Twigger AJ, Hepworth AR, Tat Lai C, et al. Gene expression in breastmilk cells is associated with maternal and infant characteristics. *Scientific reports.* 2015;5:12933.
34. Briere CE, Young E, McGrath JM, Jensen T, Finck C. Stem Cells in Breastmilk for Preterm Infants. Paper presented at: Pediatric Academic Society2016; Baltimore, MD.
35. Briere CE, Jensen T, McGrath JM, Young E, Finck C. Identification of stem cell phenotypes in preterm and full-term breast milk. *(under review).* 2016.