

The Lactating Breast: An Overview from Down Under

International Society for Research in Human Milk
and Lactation: 2006 Macy-György Award

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ABSTRACT

My research into the physiology of lactation began at the University of Sydney in the early 1960s with funding from the Australian Dairy Industry. In 1972 I moved to The University of Western Australia to teach medical students and initiated a research program in human lactation. Coincidentally this was a very significant time for human lactation because 1972 was the nadir for breastfeeding in many Western countries, including Australia. In Western Australia the proportion of women choosing to breastfeed increased from <50% to the current rate of ~95%. Because the invasive conventional techniques used to study lactation in laboratory and domestic animals could not be applied to investigation of the regulation of milk synthesis in lactating women, it was necessary to develop new approaches to the study of mammary gland metabolism in women. These methods have included measuring breast volume using a computerized breast measurement system, measuring milk macro- and micro-components on small volumes of breast milk, bioluminescent metabolomic assays, and ultrasound analysis of breast function. Currently, my research is directed toward understanding the control of synthesis, secretion, and removal of milk in women with the aim of developing clinical protocols for the assessment of the normal and abnormal function of the lactating breast.

INTRODUCTION

I WAS BORN on May 20, 1941 in Glen Innes in Northern New South Wales, Australia, and raised on a rural property at nearby Dundee. At the age of 3 months I was hospitalized with whooping cough and breastfeeding was no longer possible. My education began in a one-teacher primary school at Dundee and progressed to a small secondary school at Glen

Innes. I then completed a Bachelor of Rural Science degree at the University of New England and won a postgraduate scholarship (1963–1966) to undertake a Ph.D. at the University of Sydney in W.G. (Wally) Whittlestone's research unit under the supervision of Alick Lascelles. I was Alick's first Ph.D. student; this had many advantages in that I learned much about the university environment and strategies for gaining research funding as well as receiving a

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strong grounding in the basic research method. My postgraduate research introduced me to the intrigues of the lactating mammary gland and focused on the synthesis of milk fat in the dairy cow.¹

Postdoctoral studies were then undertaken at the National Institute for Research in Dairying, Reading, UK in Professor John Folley's laboratory in collaboration with Alfred Cowie. Alfred became a close friend and mentor. At this time we investigated the hormonal requirements for the maintenance of lactation in rabbits, an interesting laboratory research mammal because rabbits suckled their young once each day (so it was easy to measure milk production by weigh-suckle-weigh) and it was possible to hand milk up to 80 mL milk at one session from a NZ White. Among other things we found that rabbits only required prolactin for the maintenance of lactation following hypophysectomy.² Before he died I was very honored when Alfred gave me his copy of Sir Astley Cooper's two volumes of, *The Anatomy of the Breast*, a remarkable presentation of studies by Cooper himself, published a year prior to his death.³ Just as Claudius Galen (129–216 AD) influenced anatomical drawing up to and beyond the time of Leonardo da Vinci (1452–1519 AD), the anatomy of the lactating breast in current anatomy textbooks is still based on Astley Cooper's 1840 dissections. Cooper's book provided the inspiration for Donna Geddes (then Ramsay) to revisit the anatomy of the lactating human mammary gland. Rather than use Cooper's "body snatchers" to obtain experimental material, Donna was able to use the latest ultrasound equipment to carefully construct a three-dimensional model of the lactating breast.⁴ Although the results from ultrasound imaging were largely in agreement with Cooper's description of his findings from the dissection of milk ducts that had been injected with hot colored wax, a number of his findings, including the presence of lactiferous sinuses, were not supported by the ultrasound images.

From 1967 to 1968, I held the position of research instructor in physiology at the University of Pennsylvania, working with David Kronfeld. David was originally from New Zealand, completed his undergraduate studies in Australia and his doctoral studies in the

United States. I gained much from his generosity, considered advice, and philosophy of life. Returning to the University of Sydney in 1968–1971 my research in dairy cows led to the conclusion that the initiation of lactation began in two stages: first the gland developed the capacity to synthesize the unique milk constituents such as lactose during pregnancy, and then at parturition there was a rapid initiation of copious milk secretion.⁵ These events were subsequently termed lactogenesis I and II, respectively. At this time Nick Kuhn proposed that progesterone withdrawal was the lactogenic trigger in rats.⁶ In an attempt to determine if his findings had a more general application, I investigated the lactogenesis II in species with varying mechanisms for the withdrawal of progesterone and demonstrated that Kuhn's hypothesis that progesterone withdrawal was the lactogenic trigger also was valid for sheep.⁷

With the entry of the United Kingdom into the European Common Market and the concomitant major loss of exports by the Australian Dairy Industry, my research fellowship on the physiology of lactation in dairy cows was terminated in 1971 and in 1972 I was successful in obtaining a lectureship to teach biochemistry to third-year medical students at The University of Western Australia in Perth. Coincidentally, this was a very significant time for human lactation because 1972 was the year that the gradual decline in the proportion of women choosing to breastfeed that had occurred during the 20th century was arrested in many Western countries, including Australia, Sweden, and the United States. For example, in 1972 in Western Australia only approximately 48% of women chose to breastfeed their babies, and I clearly remember a member of the National Health and Medical Research Council, Grants Committee asking me during an interview why I wanted to do research on these *unusual* women, i.e., breastfeeding mothers. It obviously was not viewed as a positive career move in the early 1970s. On the other hand, being the odd one out in one of the most isolated cities in the world has had its advantages. Perhaps the greatest advantage was that the Australian Breastfeeding Association had recently formed a branch in Perth and my request for subjects to participate in my research was met with

overwhelming enthusiasm. Furthermore, over the past 34 years at The University of Western Australia 5 M.Sc. students and 31 Ph.D. students have completed their degrees under my supervision and much of the credit for the research carried out in my laboratory can be attributed directly to the efforts and creativity of these students. From success in developing sensitive assays to measure the macro- and micro-components of breast milk, noninvasive methods to measure milk synthesis and milk production in women, to the current use of ultrasound to explore the anatomy of the lactating breast, milk ejection and infant sucking have all depended for their success on the abilities of these postgraduate students.

Arriving at The University of Western Australia to teach medical students, with a research interest in the role of progesterone withdrawal in triggering lactogenesis II and the knowledge that unlike other mammals studied, the major fall in progesterone occurred after birth in women, directed my research toward human lactation. My transformation from research into lactation in farm animals to women was also facilitated by both the encouragement and practical support of my wife, who was breastfeeding our 6-month-old daughter at the time. The delayed withdrawal of progesterone in women⁸ compared with the ewe⁷ and Kevin Nicholas's findings in the rat,⁹ were indeed consistent with a frame shift in lactogenesis II to about 1.5 days postpartum in women (Fig. 1). Although the rapid increase in the concentration of lactose in the mammary secretion

was a good marker of lactogenesis II because the increase in lactose synthesis provided an osmotic draw of fluid into the alveoli to maintain osmotic equilibrium, other markers, such as citrate, sodium, and protein also proved useful, particularly in investigating lactogenesis II in women who delivered preterm.¹⁰ Furthermore, Arthur et al.¹¹ developed sensitive bioluminometric assays capable of measuring the concentration of the metabolites of lactose synthesis in breast milk (a noninvasive alternative to biopsy of breast tissue) and found from these "metabolomic" studies that the concentration of these metabolites also changes in association with the changes in concentration of lactose in milk and that lactogenesis II was delayed in women with type 1 diabetes.¹² Although lactogenesis II precedes the traditional observation of "milk coming in," it still remains a puzzle why some women experience massive painful counterproductive breast engorgement as milk "comes in" on day 2 or 3 postpartum.

The delay in the initiation of copious milk secretion in women 1 to 2 days after birth¹³ seems contrary to the metabolic needs of the newborn baby, but may function to provide a window of opportunity for colostrum to provide effective innate immune protection to the lining of the baby's gastrointestinal and respiratory tracts. This suggestion is consistent with Vorbach et al.'s¹⁴ hypothesis that the mammary gland evolved from a protective inflammatory response on the epithelial surface to a protective mucus-secreting skin gland, and finally to a lactating mammary gland deriving its nutritional function from the protective proteins lysozyme (α -lactalbumin and lactose) and xanthine oxido-reductase (fat globule secretion).

Hyttén¹⁵ in 1953 was the first to attempt to determine breast growth during pregnancy by placing a glass dome over the breast and measuring the volume of water displaced from the dome by the breast. At my suggestion one of my Ph.D. students "road tested" this procedure and left me in no doubt about what she thought of it. There had to be a better way, and on making enquiries about volume measurement, I was constantly directed to Western Australia's extensive mining industry, in which stereophotography was used to construct three-dimensional models of mountains and mineral stock-

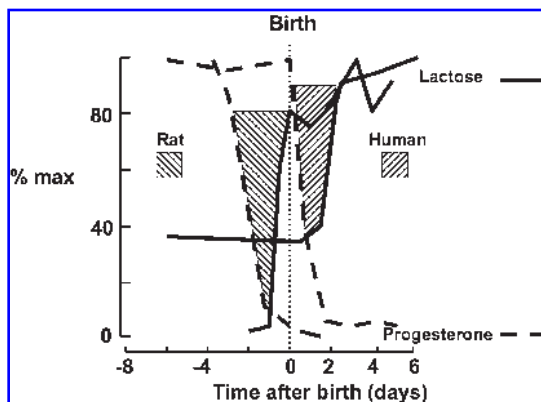


FIG. 1. Comparison of the changes in the concentration of blood progesterone and milk lactose at term in rats⁹ and humans.⁸

piles. In collaboration with a mining company we were able to provide proof of the concept that it was possible to measure breast volume with this technology, and this led to a successful grant application to develop a computerized breast measurement system. The changes in breast volume (breast growth) during pregnancy varied greatly among women, but was significantly related to the increase in concentration of human placental lactogen in the blood, whereas the metabolic development of the breast (measured by the 24-hour excretion of lactose in urine) was significantly related to the increase in the concentration of prolactin in blood.¹⁶ Following lactogenesis II, milk synthesis is maintained by the frequent removal of milk either by the baby or expressing the breast with a breast pump. In investigating the control of milk production during established lactation in healthy mothers, it was first necessary to refine methods used to measure milk production,^{17,18} as the conventional baby weigh-suckle-weigh was not applicable to mobile babies breastfed into their second year of life; furthermore, "prolonged breastfeeding" was not accepted culturally in Australia during the 1970s.

There is now considerable evidence that the milk intake of babies fed to need reflects the appetite of the baby rather than the volume of milk present in the breasts.^{19,20} Therefore, measurement of the milk intake by the baby does not necessarily provide information on the short-term rate of milk synthesis in the breast. However, we found that the decrease in breast volume from the beginning to the end of a breastfeed was closely correlated ($r = 0.93$) with the volume of milk consumed by the baby;²¹ therefore, it was reasonable to conclude that the increase in breast volume between breastfeeds was a measure of the short-term rate of milk synthesis (Fig. 2). Because the short-term rate of milk synthesis was low when the breast was at its maximum volume during the day and highest when the breast was at minimum volume, it was concluded that a local inhibitory control mechanism was regulating the short-term rates of milk synthesis in women.²² Consequently, the investigation of milk synthesis in women requires independent

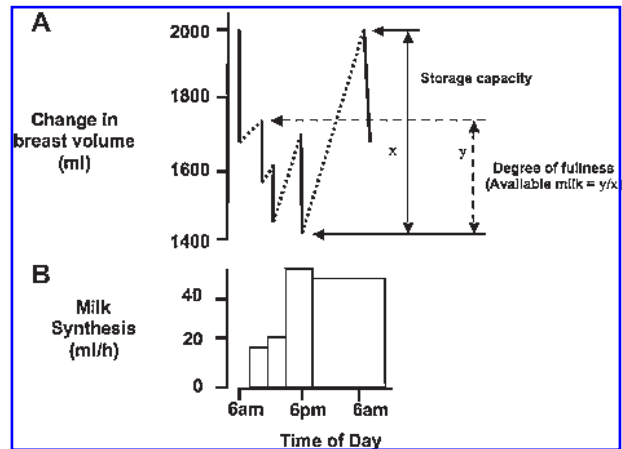


FIG. 2. (A) Changes in breast size over a 24-h period during (—) and between breastfeeds (·····). (B) Rate of milk synthesis between breastfeeds.²¹

measurements of milk production for each breast,²³ whereas for studies on infant nutrition, distinction between breasts is not important. In this connection it is of interest that in studies in collaboration with Tom Hale²⁴ we found that if a breast is expressed every hour, from the third breast expression milk production per hour became relatively constant and was not significantly different from the hourly production calculated from the 24-hour milk productions determined by test weighing in the mothers' homes. Furthermore, it was found that the degree of fullness of the breast (see Fig. 2) explained the changes in the fat content of milk from the beginning to the end of a breast feed,²⁵ and conversely changes in the fat content of milk before and after each breast feed over a 24-hour period can be used to calculate the storage capacity of a mother's breast.²³ These findings suggest that the idea of focusing on obtaining the high fat (energy) hind milk at each breastfeed could be achieved at one breastfeed, but would not provide a higher energy intake if continued over a 24-hour period.

About 7 years ago our research funding underwent a sea change when the Chairman of the Board of Medela AG, Switzerland, Michael Larsson, approached me with a request to undertake research into the function of electric breast pumps.²⁶ This has been and continues to be a very productive relationship, because improving the efficiency of electric breast

pumps requires a fundamental understanding of breast anatomy, breast milk synthesis, milk secretion, milk ejection, the physiology of breastfeeding, and infant appetite. Therefore, it is essential to carry out basic research on the physiology and biochemistry of human lactation in order to understand the requirements for effective breast expression. For example, it was necessary to develop a method of detecting milk ejection by observing the dilation of milk ducts by ultrasound imaging²⁷ so that the effectiveness of different pumping patterns on stimulating milk ejection could be assessed.

Although this year I reached what used to be the mandatory retirement age in Australia, my research group is currently the largest and most active that it has been in my career and I look forward to this situation continuing into the future. Certainly, the quest for knowledge about the physiology of the lactating breast continues and many problems remain to be solved. The nature and interaction between the local and systemic molecular mechanisms regulating breast milk synthesis are poorly understood. Why is there a contradiction between social attitudes toward breastfeeding in public and the basic *calm and connecting* biological role of oxytocin?²⁸ Why is there an unacceptably high incidence of nipple pain and mastitis in Western women?²⁹ The observation that the incidence of mastitis in rural Aboriginal women in Australia is very low and comparable to other mammals with the exception of an equally high incidence in dairy cows, suggests that mastitis is related to cultural practices. It is of interest that traditional Aboriginal women co-slept with their infants and breastfed them into their sixth year of life³⁰. Many factors, such as the function of lactose (a dimer of galactose and glucose with an unusual $\beta 1,4$ linkage) in milk, the very low proportion of the baby's total energy intake derived from protein during exclusive breastfeeding (about half that recommended for adults), and the physiology of the change in the fat content of milk during a breastfeed (perhaps a cultural artifact of less frequent breastfeeding) to name but a few areas that are poorly understood and await further investigation. Furthermore, the role of

mammary stem cells³¹ in human milk is also of interest, particularly as Kevin Nicholas's graduate student Kaylene Simpson and colleagues in Jane Visader's laboratory have recently transplanted a single mammary stem cell with LacZ reporter gene into a "de-epithelialized" fat pad of mice and this single stem cell reconstituted an entire mammary gland *in vivo* during a subsequent pregnancy.³² Although science is not a highly paid career option, it is a creative vocation that provides opportunities for travel to interesting places and opportunities to meet interesting people. As indicated, it is also rewarding to follow the careers of past postgraduate students and indeed their postgraduate students.

Since 1972 there has been a progressive increase in breastfeeding, so that now approximately 95% of Western Australian women choose to breastfeed (Fig. 3), but only about 60% of these mothers are still breastfeeding at 6 months postpartum. Therefore, almost all mothers in Australia know that breastfeeding is best for their babies; the challenge now is to support these mothers so that they can have a successful and fulfilling lactation for the first 6 months and beyond. Much of this increase in breastfeeding can be attributed to the experience-based advice provided first by the Australian Breastfeeding Association and more recently by assistance from Lactation Consultants. Since, for the mother, breastfeeding is a learned behavior normally acquired in childhood by osmosis learning—as observed in traditional Aboriginal communities by Thomson; "Little 'mothers,' each with a mud baby and clay breasts hung from the neck, at play after

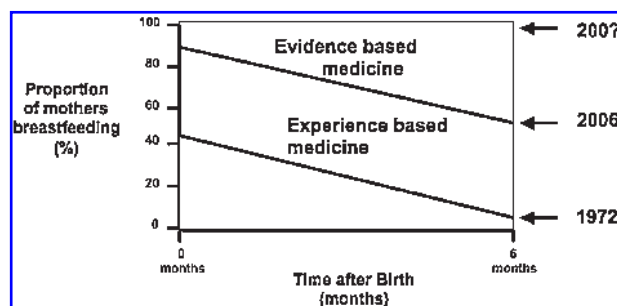


FIG. 3. The proportion of mothers breastfeeding in Perth in 1972 and 2006.

the arrival of a new baby in the camp." It is to be anticipated that in Western societies, in which breastfeeding in public is not readily accepted, most new mothers will not have acquired breastfeeding skills, such as "positioning and attachment." Therefore, experience-based advice on how to breastfeed has greatly improved breastfeeding rates. However, the same cannot be said for evidence-based advice. Wickes³³ in 1953 summarized the then current situation as follows:

In some ways this historical review might be regarded as a chronicle of man-made errors, for many of the ideas in the writings that have been quoted were intended to be an improvement upon nature though few have succeeded. Those who have heeded nature anyway, namely the nursing mothers, have seldom found it necessary to put pen to paper.

In reality, things have not improved much over the past 50 years.

Metabolically it is interesting to compare the lactating breast to the brain. Both organs have a very high metabolic activity, with the brain requiring about 23% of the body's daily energy production and the breast about 25%. Evolutionary considerations imply that any organ or activity that consumes one-fourth of the body's energy production must rank highly in ensuring the survival of the species. Although research into the function and dysfunction of the brain clearly is commensurate with its evolutionary importance for human survival, there is not a similar recognition by medical scientists of the importance of the lactating breast. For example, tens of thousands of scientists attend neuroscience conferences each year, whereas fewer than 200 scientists attend the International Society for Research in Human Milk and Lactation conferences every other year. Consequently, much less is known about the physiology and pathology of the lactating breast than other metabolically equivalent organs in the human body. Indeed, with the exception of the lactating breast, every significant organ in the human body has standard diagnostic tests to assess their normality. There is no agreed normal clinical value set for milk pro-

duction, the content of both the macronutrients and micronutrients in breast milk, or the concentration of metabolically active and innate immune factors in breast milk. As a result the effectiveness of breastfeeding is mostly judged on whether it is exclusive or partial breastfeeding. In this context it is surprising that so many positive outcomes for the infant are observed. There is a similar lack of normal clinical values for lactating mothers, at least in Australia. There are not even normal levels for blood prolactin during lactation, yet the administration of drugs to increase the concentration of prolactin is not uncommon practice. It is beyond belief that in the 21st century that an organ that requires 25% of daily energy production does not have a medical specialty. Fortunately, the formation of the Academy of Breastfeeding Medicine has provided the beginnings for the development of an evidence-based medical specialty in human lactation. The development of evidence-based diagnosis and treatment of lactation problems will do much to ensure that the 95% of mothers who commence breastfeeding in Australia will have a sustained and successful breastfeeding experience.

Nevertheless, the question still remains, "Why is it that we know so little about an organ that consumes 25% of daily energy production and contributes so much to intellectual and physical development as well as to a healthy life?" The lactating breast must be accorded the same scientific and medical status as other equivalent organs in the human body; then those of us researching this fascinating organ will no longer be viewed as studying *unusual* women.

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REFERENCES

- Hartmann PE, Lascelles AK. The effect of starvation on the uptake of the precursors of milk fat by the bovine mammary gland. *Austr J Biol Sci* 1965;18:1025–1034.
- Hartmann PE, Cowie AT, Hosking ZD. Changes in enzymic activity, chemical composition and histology of the mammary glands and metabolites in the blood of lactating rabbits after hypophysectomy and replacement therapy with sheep prolactin, human growth hormone or bovine growth hormone. *J Endocrinol* 1970;48:443–448.
- Cooper AP. *The Anatomy of the Breast*. Longmans, London, 1840.
- Ramsay DT, Kent JC, Hartmann RL, et al. Anatomy of the lactating human breast redefined with ultrasound imaging. *J Anatomy* 2005;206:525–534.
- Hartmann E. Changes in the composition and yield of the mammary secretion on cows during the initiation of lactation. *J Endocrinol* 1973;59:231–247.
- Kuhn NJ. Progesterone withdrawal as the lactogenic trigger in the rat. *J Endocrinol* 1969;44:39–54.
- Hartmann PE, Trevethan P, Shelton JN. Progesterone and oestrogen and the initiation of lactation in ewes. *J Endocrinol* 1973;59:249–259.
- Kulski JK, Smith M, Hartmann PE. Perinatal concentrations of progesterone, lactose and α -lactalbumin in the mammary secretion of women. *J Endocrinol* 1977;74:509–510.
- Nicholas KR, Hartmann PE. Progesterone control of the initiation of lactose synthesis in the rat. *Aust J Biol Sci* 1981;34:435–443.
- Cregan MD, de Mello TR, Kershaw D, et al. Initiation of lactation in women after preterm delivery. *Acta Obst Gynecol Scand* 2002;81:870–877.
- Arthur PG, Kent JC, Hartmann PE. Metabolites of lactose synthesis in milk from women during established lactation. *J Pediatr Gastroenterol Nutr* 1991;13:260–266.
- Arthur PG, Kent JC, Hartmann PE. Metabolites of lactose synthesis in milk from diabetic and non-diabetic women during lactogenesis II. *J Pediatr Gastroenterol Nutr* 1994;19:100–108.
- Saint L, Smith M, Hartmann PE. The yield and nutrient content of colostrum and milk of women from giving birth to 1 month post-partum. *Br J Nutr* 1984;52:87–95.
- Vorbach C, Capecchi MR, Penninger JM. Evolution of the mammary gland from the innate immune system? *BioEssays* 2006;28:606–616.
- Hyttén FE. Clinical and chemical studies in human lactation: VI The functional capacity of the breast. *BMJ* 1954 (April 17); 912–915.
- Cox DB, Kent JC, Casey TM, et al. Breast growth and the urinary excretion of lactose during human pregnancy and early lactation: Endocrine relationships. *Exp Physiol* 1999;84:421–434.
- Rattigan S, Ghisalberti AV, Hartmann PE. Breast-milk production in Australian women. *Br J Nutr* 1981;45:243–249.
- Prosser CG, Saint L, Hartmann PE. Mammary gland function during gradual weaning and early gestation in women. *Aust J Exp Biol Med Sci* 1984;62:215–228.
- Dewey KG, Lonnerdal B. Infant self-regulation of breast milk intake. *Acta Paediatr Scand* 1986;75:983–988.
- Hartmann PE, Sherriff J, Kent JC. Maternal nutrition and the regulation of milk synthesis. *Proc Nutr Soc* 1995;54:379–389.
- Daly SEJ, Kent JC, Huynh DQ, et al. The determination of short-term breast volume changes and the rate of synthesis of human milk using computerized breast measurement. *Exp Physiol* 1992;77:79–87.
- Daly SEJ, Kent JC, Owens RA, et al. Frequency and degree of milk removal and the short-term control of human milk synthesis. *Exp Physiol* 1996;81:861–875.
- Kent JC, Ramsay DT, Cregan MD, et al. Volume and frequency of breastfeeds and fat content of breast milk throughout the day. *Pediatrics* 2006;117:e387–e395.
- Lai CT, Hale T, Kent JC, et al. Hourly rate of milk synthesis in women. *12th International Conference of the International Society for Research in Human Milk and Lactation (ISRHMLQueens' College Cambridge, UK. September 10–14, 2004, p. 48.*
- Daly SEJ, Di Rosso A, Owens RA, et al. Degree of breast emptying explains changes in the fat content, but not fatty acid composition, of human milk. *Exp Physiol* 1993;78:741–755.
- Mitoulas LR, Lai CT, Gurrin LC, et al. Efficacy of breast milk expression using an electric breast pump. *J Hum Lact* 2002;18:344–352.
- Ramsay DT, Kent JC, Owens RA, et al. Ultrasound imaging of milk ejection in the breast of lactating women. *Pediatrics* 2004;113:361–367.
- Uvnäs Moberg K. *The Oxytocin Factor: Tapping the Hormone of Calm, Love and Healing*. Da Capo Press, Cambridge, MA, 2003.
- Fetherston CM, Lai CT, Hartmann PE. Relationships between symptoms and changes in breast physiology during lactation mastitis. *Breastfeeding Med* 2006;1:136–145.
- Thomson D. *Children of the Wilderness*. Gordon & Gotch, Melbourne, 1983.
- Cregan MD, Fan YP, Appelbee A, et al. Identification of nesting positive putative mammary stem cells in human breast milk. *Cell Tissue Res* 2007; in press.
- Visvader JE, Lindeman, GJ. Mammary stem cells and mammaryogenesis. *Cancer Res* 2006;66:9798–9801.
- Wickes IG. A history of infant feeding: Nineteenth century concluded and twentieth century. *Arch Dis Child* 1953;28:495–505.

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