Symposium: Human Lactogenesis II: Mechanisms, Determinants and Consequences

Lactogenesis and the Effects of Insulin-Dependent Diabetes Mellitus and Prematurity[1,2]

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ABSTRACT The initiation of lactation (lactogenesis II) by the mother must be synchronized to the delivery of the infant, permitting the transition of the newborn from continuous nourishment from the umbilical cord to comparable but intermittent life support from its mother's breasts. The onset of lactogenesis II can be adversely affected by a variety of factors. Over 80% of women who have delivered prematurely and are expressing milk for their infant had a compromised initiation of lactation, that is one or more lactogenesis II markers (lactose, citrate, sodium and total protein) in their milk $>$ 3 SD from the mean of the full-term women on d 5 postpartum. Similarly, the lactogenesis II markers (lactose, citrate and total nitrogen) in the milk of women with insulin-dependent diabetes mellitus take an additional 24 h to attain the concentrations of normal women. The mechanisms that lead to the development of delayed or compromised onset of lactogenesis II in women are poorly understood and require additional research. J. Nutr. 131: 3016S–3020S, 2001.

KEY WORDS: • *lactogenesis* • *prematurity* • *diabetes* • *progesterone*

The potential to secrete milk is a characteristic common to the females of all 4000^{+} species of mammal. However, the great variation that exists between species in maturity of the young at birth has resulted in the nutrient composition of milk being uniquely adapted to complement the young of each species. Furthermore, the maturity of the infants systems (such as digestive, hepatic, neural, renal, vascular, visual, skeletal and immune) varies between species. Thus, the composition of milk varies with stage of lactation specific to each species, so that the infant receives nutrition appropriate to its stage of development.

Lactation cycle

The marsupials represent one extreme of this adaptation because they have a short gestation and a relatively long lactation period with the body weight of the young increasing several thousand-fold from birth to weaning (1). Several distinct phases of mammary development have been defined in macropods (1). For example, in the Tammar Wallaby, the mammary gland increases several-fold in size during a 28-d pregnancy (phase 1) (2). After delivery, phase 2 of the lactation cycle is initiated by the single fetus-like infant attaching to one of four teats in the pouch. Phase 2 is associated with a high carbohydrate and low fat and protein milk (3) and is further subdivided into two 100-d phases. During phase 2A, the young Tammar Wallaby remains permanently attached to the teat, whereas in phase 2B the infant relinquishes the teat and suckles less frequently. The onset of phase 3 occurs at ~200 d of lactation and continues until 300–350 d postpartum, with the infant leaving the pouch at ~250 d. It is characterized by increased milk production (4) and a change in milk composition to low carbohydrate and high fat and protein (5). Furthermore, the Tammar is capable of asynchronous concurrent lactation, with adjacent mammary glands functioning independently. Thus, one small gland can be commencing phase 2, with the young Tammar Wallaby in the pouch attached permanently to the teat, while another larger gland is in phase 3 and suckled by an infant at foot.

Compared with marsupials, the lactation cycle of eutherian mammals is relatively simple consisting of mammogenesis, lactogenesis, galactopoiesis and involution. Lactogenesis refers to the expression of specific genes required for the synthesis of milk by the lactocytes (mammary secretory epithelial cells) and occurs in two stages (6) in those eutherian mammals that have been studied to date. Lactogenesis I occurs during pregnancy, whereas lactogenesis II occurs close to birth (7).

Lactogenesis I

Lactogenesis I represents the initiation of the synthesis of milk-specific components (e.g., lactose, casein and α-lactalbu-

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min), and its occurrence has been identified by the detection of these components in the mammary tissue, mammary secretion, blood or urine of the mother. There is considerable variation both between and within species on the timing of lactogenesis I, but it seems that mammals that produce a placental lactogen tend to initiate lactogenesis earlier in gestation (8). With the possible exception of the ewe (9), the timing of lactogenesis I does not seem to greatly influence lactogenesis II.

Lactogenesis II

Although, biochemically, lactogenesis I represents the initiation of the unique synthetic capacity of the mammary gland, lactogenesis II is the most critical stage of the lactation cycle. At this stage of the cycle, the initiation of milk synthesis by the mother must be synchronized to the time of birth to permit the transition of the newborn from continuous nourishment from the umbilical cord to comparable but intermittent life support from its mother's breasts.

Measurement. By definition, lactogenesis II is the initiation of copious milk secretion; therefore, the measurement of the rate of milk synthesis provides an obvious method of determining the onset of lactogenesis II (6). However, it is difficult to use this method to determine the synchronization of lactogenesis II with parturition because, in most species, lactogenesis II occurs either just before or at the time of parturition and the prepartum removal of secretion could in itself initiate lactogenesis II. In women, where lactogenesis II occurs after parturition, the measurement of the intake of breast milk by the infant over the first few days after birth is difficult and it is possible that the infant may not consume all of the available milk (colostrum). The exception being women who deliver prematurely, where (providing their breasts can be successfully expressed by either hand or breast pump) milk production can be measured.

Lactogenesis II is also associated with large changes in the composition of the mammary secretion as the transition from colostrum (high concentration of total protein, immunoglobulins, sodium and chloride; low concentrations of lactose, potassium, glucose and citrate) to mature milk with the reverse concentrations of these constituents. Consequently, changes in the concentration of one or a combination of these constituents can be used to identify the onset of lactogenesis II (10,11). However, there is some variation between species. Although the postnatal increase in the concentration of citrate in human milk is considered "the harbinger of lactogenesis" (12), in the sow, citrate decreases during lactogenesis II (13).

Hormonal control. Birth and lactogenesis II are closely coupled by their mutual relationship to the withdrawal of progesterone in eutherian mammals. The pioneering work of Kuhn (14) established that progesterone withdrawal was the trigger for lactogenesis II in rats, and this mechanism seems to control the timing of lactogenesis II in a number of other species, including humans (8). However, in contrast to other species, the major fall in progesterone occurs after birth in women (commencing with the delivery of the placenta); therefore, lactogenesis II is delayed until ~24 h after birth. Because this is contrary to the infant's perceived metabolic needs, there must be a strong selective advantage associated with this delay. The metabolic resilience of the human infant at birth is probably related to its significant fat reserves and its slow rate of postnatal growth. For example, the piglet is born with only 2% body fat (15) and doubles it birth weight in the first week of life, whereas the human infant merely regains it birth weight during the same period. Furthermore, each piglet consumes approximately three times more colostrum (50–100 mL) in the first 4 h after birth (16) than the human infant consumes (~20–30 mL) during the first 24 h of life (17). This difference in milk production clearly demonstrates the difference between these two species in the timing of lactogenesis II. In women, premature delivery, insulin-dependent diabetes mellitus (IDDM),[4] obesity, Cesarean section and endocrine disturbances can either delay or suppress lactogenesis II and affect the successful establishment of lactation (18).

Term mothers. Acute postpartum changes in the composition (e.g., lactose; **Fig. 1**) of the mammary secretion concurrent with an increase in milk production identify the onset of lactogenesis II at between 30 and 40 h after birth in women (10). Twenty-four hours after this time, women often experience a sudden feeling of breast fullness and their milk supply is said to be coming in. It has long been assumed that this was a marker of the initiation of lactation in women (19). Indeed, Cadogan stated: "When a child is first born, there seems to be no provision at all made for it; for the mother's milk seldom comes 'till the 3rd day; so that, according to nature, a child would be left a day and a half, or two days, without any food; to me, a very sufficient proof that it wants none." This conclusion was consistent with the widely held cultural belief at that time that colostrum was not good for the newborn infant.

The observations of Neifert et al. (20) that retention of placental fragments inhibited lactogenesis II supported the importance of progesterone withdrawal in determining the timing of lactogenesis II in women. Indeed, De Passille et al. (21) reported reduced growth rates and increased mortality in piglets from sows that had slightly higher concentrations of progesterone in their blood after farrowing, suggesting that a slow withdrawal of progesterone may compromise the establishment of lactation. If a similar response to progesterone withdrawal occurs in women, extreme caution should be exercised with progestagen administration in the perinatal period.

Preterm mothers. Women who have delivered preterm and express their milk seem more likely to produce less milk than women who deliver full-term (22), particularly in those women who do not express their milk for the first few days after birth (23). It is possible that the poor milk production at this time in some expressing women could be related to large differences among women in the effectiveness of breast pumps in removing the available milk from some women's breasts (24).

We recently investigated milk production and the concentration of four markers of lactogenesis II (milk citrate, lactose, sodium and total protein) at d 5 postpartum in 22 women who were expressing their milk for their preterm infant (11). The lactogenesis II markers for the preterm women had much greater variation about the mean women on d 5 postpartum than was observed in full-term breastfeeding women ($n = 16$). Furthermore, all the full-term women had all four markers within 3 SD of the mean for the full-term women and were classified as having successfully undergone lactogenesis II. However, only 18% of the preterm women had all four markers within 3 SD of the mean for full-term women (**Fig. 2**). The remaining preterm women had one or more of the markers > 3 SD from the mean concentration for full-term women. In

[4] Abbreviations used: GDM, gestational diabetes mellitus; IDDM, insulin-dependent diabetes mellitus.

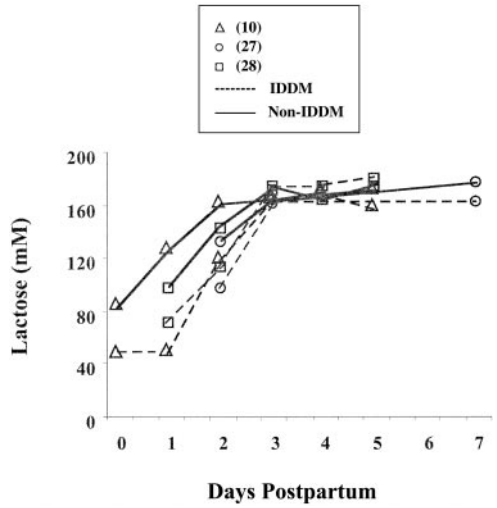

FIGURE 1 The change in concentration of lactose from birth to 7 d postpartum as observed by Arthur et al. (10), Δ, Cox (28), □ and Neubauer et al. (27), O in nondiabetic (solid line) and IDDM (dashed line) women.

addition, the preterm women who demonstrated one or more lactogenesis II marker outside the range for the full-term women had significantly lower milk productions than did preterm women with all four markers within the range for full-term women. Therefore, it was concluded that lactogenesis II had been compromised in 82% of the preterm women. Similar changes in the concentration of the markers of lactogenesis II occur in the mammary secretion in full-term women regardless of whether they choose to feed breast milk to their infants (25). Therefore, the efficiency of removal of available colostrum by breast pumping cannot explain the compromised lactogenesis II of the preterm women.

In Australia, 97% of women are prescribed antenatal corticosteroids (β-methasone) during uncomplicated preterm labor, and 85% are prescribed repeated doses when the risk of preterm delivery persists (26). Cowie et al. (7) hypothesized that during pregnancy, circulating progesterone and corticosteroids compete for the glucocorticoid receptors on the mammary epithelial cells. Therefore, any treatment with corticosteroids could override the progesterone inhibition of lactation during pregnancy. For example, we have found that the administration of therapeutic doses of β-methasone to the fetus or pregnant ewes at 104, 111 and 118 d of gestation significantly increased the concentration of lactose in the mammary secretion by d 125 of gestation (**Fig. 3**) and resulted in a poor lactation performance at term (Johnson, S., Newnham, J. P., Moss, T. & Hartmann, P. E., unpublished observations). Although such a link has not been established in women, the compromised lactogenesis II in preterm women may be related to the unintended stimulation of the breast by steroid hormone therapy for high risk pregnancies, and, thus, causing lactogenesis II to occur before delivery.

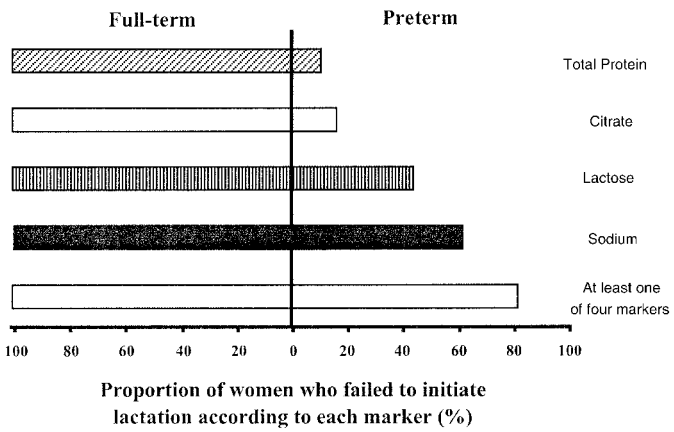

FIGURE 2 The proportion of full-term and preterm women who failed to initiate their lactation (11) according to the lactogenesis II markers (total protein, citrate, lactose and sodium).

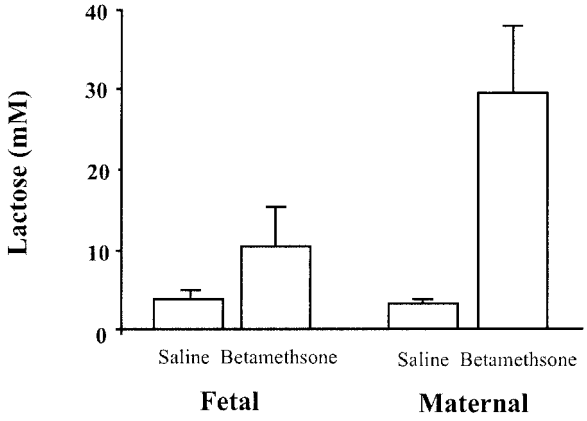

FIGURE 3 The concentration of lactose in the mammary secretion of pregnant ewes after administration of therapeutic doses of β-methasone to the fetus or ewe at 104, 111 and 118 d of gestation (Johnson, S., Newnham, J. P., Moss, T. & Hartmann, P. E., unpublished observations).

IDDM. Using lactose and citrate as markers of lactogenesis II, Arthur et al. (10) found that the onset of lactogenesis II was delayed by between 15 and 28 h in women with IDDM. Subsequently, Neubauer et al. (27) confirmed these findings and concluded that in women with IDDM the onset of lactogenesis II, as defined by the milk markers, lactose and total nitrogen (an approximation to total protein), was delayed by ~24 h (Fig. 1). However, no significant difference was found between IDDM and control women in the amount of milk produced over a 24-h period at d 7 postpartum. Because it had been claimed that better hospital management of the IDDM women had prevented the delay in the onset of lactogenesis II, Cox (28) repeated the study of Arthur et al. (10), comparing the initiation of lactation in 10 control women with 8 IDDM women. A significant delay was found in the timing of the increase in lactose in the colostrum from IDDM women (Fig. 1). Although no differences were observed in the change in concentration of either sodium or potassium in colostrum between control and IDDM women, significant differences were observed in the change in concentration of lactose and total protein; therefore, these women showed some of the characteristics of the compromised onset of lactogenesis II observed in preterm women.

Lactose, the most osmotically active component of human milk, draws water into the lactocyte (29). Thus, Arthur et al. (10) concluded that any delay in the increase in the concentration of lactose in IDDM women was associated with a decrease in milk volume in the first 3 d postpartum. Furthermore, in view of the importance of citrate in generating acetyl CoA from glucose (30), they also suggested that the delay in

the increase in the concentration of citrate may be associated with a delay in the de novo synthesis of medium-chain fatty acids. In this connection, Bitman et al. (31) reported that IDDM women who started pumping at 72 h postpartum initially produced milk with lower medium-chain fatty acid, total fat and cholesterol but higher oleic, linoleic and polyunsaturated long-chain fatty acid content than a reference population. Ferris et al. (32) observed that IDDM women commence breastfeeding, on average, 24 h later than nondiabetic women. Furthermore, it has subsequently been demonstrated that an increased number of early breastfeeding episodes (within the first 12 h postpartum) were critically important for stimulating the onset of lactogenesis II in women with IDDM (33).

The cause of the delay in the onset of lactogenesis II in IDDM women is unclear. Because human Placental Lactogen (hPL) is positively correlated with breast growth during pregnancy (34), there could be a possible involvement of hPL in the delay of the onset of lactogenesis II in some IDDM women. Previous studies have found that IDDM had no effect on circulating levels of progesterone, total estriol, prolactin (35) estradiol or human Chorionic Gonadotrophin (36) when compared with nondiabetic women. However, Botta et al. (35) described that circulating levels of hPL were significantly lower at all latter stages of pregnancy, compared with nondiabetic women, whereas Stewart et al. (36) found no difference between the two groups. There is no obvious reason for the discrepancy between these two studies. Furthermore, human hPL has been demonstrated to support lactogenesis II in the absence of maternal prolactin in rats (37).

Gestational diabetes mellitus (GDM). Normal human pregnancy is accompanied by hyperinsulinaemia and normal to slightly decreased glucose tolerance (38). However, GDM occurs in ~3% of Western women and is defined as diabetes diagnosed for the first time during pregnancy that then resolves postpartum (39). Obesity is a risk factor for delayed onset of lactation (18,40), and because GDM is most common in obese women (41), it is possible that the diabetes or the obesity could alter the onset of lactogenesis II. Our preliminary investigation of noninsulin-dependent gestational diabetic women suggested that there was not a marked delay in the onset of lactogenesis II because the concentration of lactose in the colostrum of GDM women was not significantly different from control women at 40–50 h postpartum (data not shown). However, earlier samples were not available for GDM women because they had difficulty expressing colostrum from their breasts during the first 2 d of lactation.

Current maternity practice rightly focuses on the importance of the appropriate positioning and attachment of the infant to the breast. However, the onset of lactogenesis II in women can be influenced by a variety of pathological factors (IDDM, premature delivery, obesity, prolactin deficiency and delayed progesterone withdrawal) as well as hormonal and anesthetic therapeutic agents administered during pregnancy and child birth. Taken together, these findings suggest that additional research is required to determine the causes of delayed onset of lactogenesis II in women and its influence on the establishment of successful breastfeeding. Clinically, delayed onset of lactogenesis II should be considered a potential factor in the perinatal management of human lactation.

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