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Calcium and Bone Metabolism During Pregnancy and Lactation

Christopher S. Kovacs[1]

Pregnancy and lactation both place significant demands on the mother to provide sufficient calcium (among other minerals and nutrients) to the fetus and neonate. Despite facing similar demands for calcium during pregnancy and lactation, the maternal adaptations differ significantly between these two reproductive periods. Women lose 300 to 400 mg of calcium daily through breast milk, and this calcium demand is met by a 5–10% loss of skeletal mineral content during 6 months of exclusive lactation. Most importantly, the lost mineral is fully restored within a few months of weaning, such that women who have breastfed do not have a long-term deficit in skeletal mineral content. This article will review our present understanding of the adaptations in mineral metabolism that occur during pregnancy and lactation, and will focus on recent evidence that the breast itself plays a central role in regulating the adaptations during lactation.

KEY WORDS: pregnancy; lactation; calcium metabolism; bone metabolism; osteoporosis.

Mineral metabolism in the mother must adapt to the demand created by the fetus and placenta, which together draw calcium and other minerals from the mother in order to mineralize the developing fetal skeleton. Similarly, mineral metabolism must adapt in the lactating woman in order to supply sufficient calcium to the milk. Potential adaptations include increased intake of mineral, increased efficiency of intestinal absorption of mineral, mobilization of mineral from the skeleton, and increased renal conservation of mineral. Despite a similar magnitude of calcium demand presented to pregnant and lactating women, the adjustments made in each of these reproductive periods differ significantly (Fig. 1). These hormone-mediated adjustments normally satisfy the needs of the fetus and infant with short-term depletions of maternal skeletal calcium content, but without long-term consequences to the maternal skeleton. However, in states of maternal malnutrition and vitamin D deficiency, the depletion of skeletal mineral content may be proportionately more severe, and may in turn be accompanied by increased skeletal fragility.

This article will review our present understanding of the adaptations that occur during pregnancy and lactation, making use of animal data to fill in the gaps where human data are unavailable. Due to restrictions on the length of the reference list, the reader is also referred to several other comprehensive reviews on the subject *(1–3)*.

PREGNANCY

The developing fetal skeleton accretes about 30 g of calcium by term, about 80% of it during the third trimester. This demand for calcium is largely met by a doubling of maternal intestinal calcium

[1] Faculty of Medicine - Endocrinology, Health Sciences Centre, Memorial University of Newfoundland, St. John's, Newfoundland, A1B 3V6, Canada; e-mail: ckovacs@mun.ca.

Abbreviations used: CT, calcitonin; DPA, single photon absorptiometry; DXA, dual X-ray absorptiometry; FSH, follicle stimulating hormone; GFR, glomerular filtration rate; GnRH, gonadotropin releasing hormone; LH, luteinizing hormone; PTH, parathyroid hormone; PTHrP, parathyroid hormone-related protein; SPA, single photon absorptiometry.

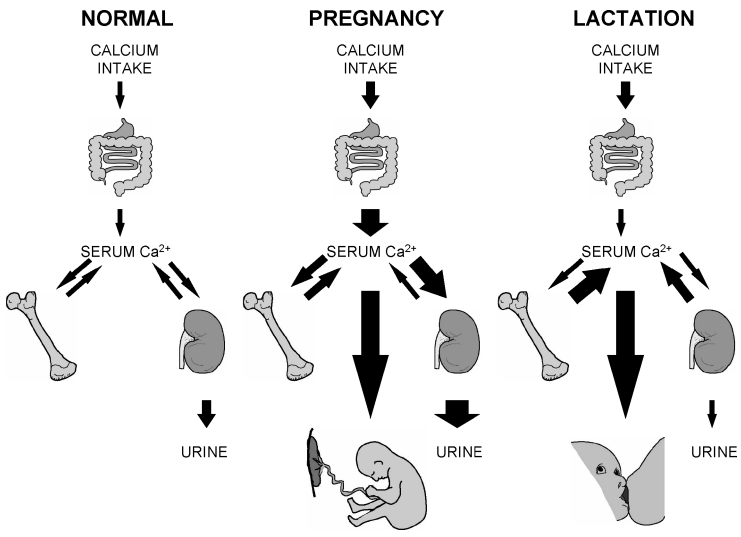

Fig. 1. Schematic illustration contrasting calcium homeostasis in human pregnancy and lactation, as compared to normal. The thickness of arrows indicates a relative increase or decrease with respect to the normal and non-pregnant state. Adapted with permission from ref. *(1)*, ©1997 The Endocrine Society.

absorption, mediated by 1,25-dihydroxyvitamin D and possibly other factors.

Mineral Ions and Calcitropic Hormones

Normal pregnancy results in altered levels of calcium and the calcitropic hormones, as schematically depicted in Fig. 2 *(1)*. The ionized calcium (the physiologically important fraction of calcium) remains constant throughout pregnancy. In contrast, the total serum calcium (which is the sum of the ionized, complexed and albumin-bound fractions of calcium in the circulation) falls in pregnancy due to a fall in the serum albumin. In clinical practice, the total serum calcium is more commonly measured than the ionized calcium. The commonly observed decrease in total serum calcium should not be mistaken for evidence of "physiological hyperparathyroidism of pregnancy," an erroneous concept that has persisted in some modern texts *(4,5)*. The fall in total serum calcium is an unimportant artifact of an unphysiological measurement; the ionized calcium is the relevant measurement and should always be assayed if there is any doubt about the true value of the serum calcium during pregnancy (or at any time). Serum phosphorus levels are also normal during pregnancy.

As observed by longitudinal measurements during pregnancy with modern 2-site immunoradiometric assays, serum parathyroid hormone (PTH) falls to the low-normal range (i.e., 10–30% of the mean non-pregnant value) during the first trimester, and then increases steadily to the mid-normal range by term *(6–10)*. Therefore, as judged by the serum PTH level, the parathyroids are modestly suppressed beginning early in the first trimester, and return to apparently normal function by the end of pregnancy. First-generation PTH assays in the 1970s and 80s were insensitive and measured multiple biologically inactive fragments of PTH; a few studies with these assays had detected higher levels of PTH during pregnancy in humans. Those early studies of PTH in pregnancy, combined with the observation that total serum calcium falls during pregnancy, reinforced the erroneous concept that secondary hyperparathyroidism occurs during pregnancy. Modern assays have made it very clear that in well-nourished women, the ionized calcium is normal throughout pregnancy, and that PTH is suppressed during early pregnancy. In contrast, there is evidence that PTH may increase above normal in late pregnancy in women from Malay who have very low intakes of calcium *(11)*.

Total 1,25-dihydroxyvitamin D levels double early in pregnancy and maintain this increase until term; free 1,25-dihydroxyvitamin D levels are increased from the third trimester and possibly earlier. The rise in 1,25-dihydroxyvitamin D may be largely independent of changes in PTH, since PTH

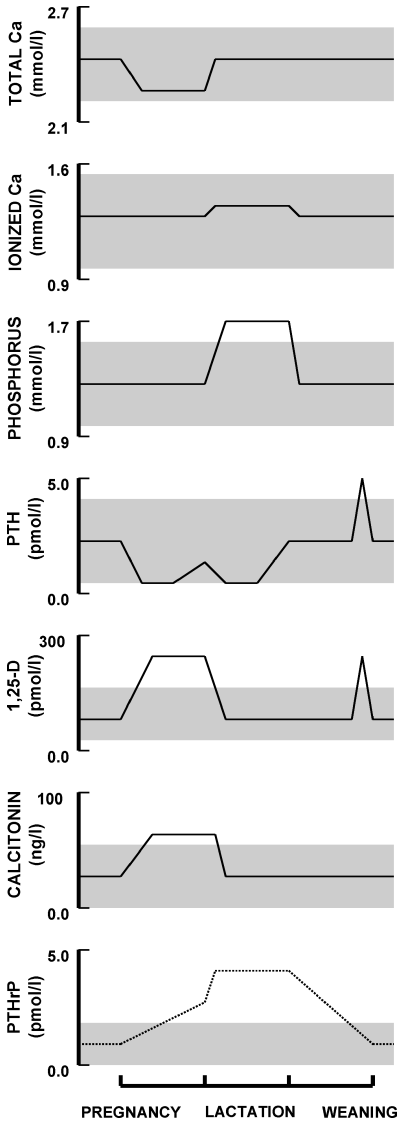

Fig. 2. Schematic illustration of the longitudinal changes in calcium, phosphorus, and calcitropic hormone levels in the circulation during pregnancy and lactation. Normal adult ranges are indicated by the shaded areas. The progression in PTHrP levels has been depicted by a dashed line to reflect that the data is less complete; the implied comparison of PTHrP levels in late pregnancy and lactation are uncertain extrapolations because no reports followed patients serially. In both situations PTHrP levels are elevated. Adapted with permission from ref. *(1)*, ©1997 The Endocrine Society.

levels are typically decreasing at the time of the increase in 1,25-dihydroxyvitamin D. The maternal kidneys likely account for most, if not all, of the rise in 1,25-dihydroxyvitamin D during pregnancy, although the decidua, placenta and fetal kidneys may contribute a small amount. The relative contribution of the maternal kidneys is based on several lines of evidence [reviewed in *(1)*], including the report of an anephric woman on hemodialysis who had low 1,25-dihydroxyvitamin D levels before and during a pregnancy *(12)*. The renal 1α−hydroxylase may be up-regulated in response to factors such as PTH-related protein (PTHrP), estradiol, prolactin, and placental lactogen.

Serum calcitonin levels are also increased during pregnancy, with the C-cells of the thyroid, breast and placenta possibly contributing to the circulating level of calcitonin. It has been postulated that calcitonin protects the maternal skeleton from excessive resorption of calcium, but this hypothesis remains unproved. No human studies have addressed the question, while recent studies in genetically engineered mice demonstrated that absence of calcitonin does not impair the ability of mice to increase skeletal mineral content during pregnancy *(13)*.

PTH-related protein (PTHrP) levels are increased during pregnancy, as determined by assays that detect PTHrP fragments encompassing amino acids 1–86. Since PTHrP is produced by many tissues in the fetus and mother (including the placenta, amnion, decidua, umbilical cord, fetal parathyroids, and breast), it is not clear which source(s) contribute to the rise detected in the maternal circulation. PTHrP may contribute to the elevations in 1,25-dihydroxyvitamin D and suppression of PTH that are noted during pregnancy. PTHrP may have other roles during pregnancy, such as regulating placental calcium transport in the fetus *(1,14)*. Also, PTHrP may have a role in protecting the maternal skeleton during pregnancy, since the carboxyl-terminal portion of PTHrP ("osteostatin") has been shown to inhibit osteoclastic bone resorption *(15)*.

Pregnancy induces significant changes in the levels of other hormones, including the sex steroids, prolactin, placental lactogen, and IGF-1. Each of these may have direct or indirect effects on calcium and bone metabolism during pregnancy, but these issues have been largely unexplored.

Intestinal Absorption of Calcium

Several clinical studies have demonstrated that intestinal absorption of calcium is doubled during pregnancy from as early as 12 weeks of gestation (the earliest time-point studied); this change appears to be a major maternal adaptation to meet the fetal need for calcium. This increase may be largely

the result of a 1,25-dihydroxyvitamin D-mediated increase in intestinal calbindin$_{9K}$-D and other proteins; based on evidence from limited animal studies, prolactin and placental lactogen (and possibly other factors) may also mediate part of the increase in intestinal calcium absorption. The increased absorption of calcium early in pregnancy may allow the maternal skeleton to store calcium in advance of the peak fetal demands that occur later in pregnancy.

Renal Handling of Calcium

The 24-h urine calcium excretion is increased as early as the 12th week of gestation (the earliest timepoint studied), and the amounted excreted may exceed the normal range. Since fasting urine calcium values are normal or low, the increase in 24-h urine calcium likely reflects the increased intestinal absorption of calcium (absorptive hypercalciuria). The elevated calcitonin levels of pregnancy might also promote renal calcium excretion.

Skeletal Calcium Metabolism

Animal models indicate that histomorphometric parameters of bone turnover are increased during pregnancy, which could be interpreted to mean that mineral is mobilized from the maternal skeleton to contribute to the fetal skeleton *(16)*. However, serial measurements of bone mineral density by dual X-ray absorptiometry (DXA) in several strains of normal mice have demonstrated that the bone mineral content increases by 5–10% during pregnancy *(13,17)*, and thus the increased bone turnover of pregnancy might (at least in rodents) reflect an anabolic or bone formative state, as opposed to a net bone resorptive state. As noted below in the lactation section, a net loss of bone mineral content occurs during lactation in humans and rodents. An increase in bone mineral content during pregnancy might serve to protect the maternal skeleton against excessive demineralization and fragility during lactation.

Comparable histomorphometric data are not available for human pregnancy. In one study *(18)*, 15 women who electively terminated a pregnancy in the 1st trimester (8–10 weeks) had bone biopsy evidence of increased bone resorption, including increased resorption surface, increased numbers of resorption cavities, and decreased osteoid. These findings were not present in biopsies obtained from non-pregnant controls, or in biopsies obtained at term from 13 women who had elective C-sections.

Most human studies of skeletal calcium metabolism in pregnancy have examined changes in "bone markers," that is, serum indices that reflect bone formation, and serum or urine indices that reflect bone resorption. These studies have been fraught with a number of confounding variables that cloud the interpretation of the results, including lack of pre-pregnancy baseline values; effects of hemodilution in pregnancy on serum markers; increased glomerular filtration rate (GFR) and renal clearance; altered creatinine excretion; placental, uterine and fetal contribution to the markers; degradation and clearance by the placenta; and lack of diurnally timed or fasted specimens. Given these limitations, many studies have reported that urinary markers of bone resorption (24-h collection) are increased from early to mid-pregnancy (including deoxypyridinoline, pyridinoline, and hydroxyproline). Conversely, serum markers of bone formation (generally not corrected for hemodilution or increased GFR) are often decreased from pre-pregnancy or non-pregnant values in early or mid-pregnancy, rising to normal or above before term (including osteocalcin, procollagen I carboxypeptides and bone specific alkaline phosphatase). It is conceivable that the bone formation markers are artifactually lowered by normal hemodilution and increased renal clearance of pregnancy, obscuring any real increase in the level of the markers. One study adjusted for the confounding effects of hemodilution and altered GFR, and demonstrated that osteocalcin production was not reduced in pregnancy *(19)*. Total alkaline phosphatase rises early in pregnancy due largely to contributions from the placental fraction; it is not a useful marker of bone formation in pregnancy.

Based on the scant bone biopsy data, and the measurements of bone markers (with aforementioned confounding factors), one may cautiously conclude that bone turnover is increased in human pregnancy, from as early as the 10 th week of gestation. There is comparatively little maternal-fetal calcium transfer occurring at this stage of pregnancy, as compared to the peak rate of calcium transfer in the third trimester. One might have anticipated that markers of bone turnover would increase particularly in the third trimester; however, no further increase is seen at that time.

Changes in skeletal calcium content during pregnancy would ideally be assessed by sequential bone density measurements in each woman; however, due to concerns about fetal radiation exposure, few such studies have been done. Those studies used the older techniques of single photon absorptiometry (SPA) of the wrist and dual-photon absorptiometry (DPA) of the spine and hip, with abdominal shielding in place to protect the fetus. Such studies were confounded by changes in body composition and weight during normal pregnancy that can lead to artifactual changes in bone density. Using these outdated techniques (SPA/DPA), no significant change was observed in cortical or trabecular bone density during pregnancy *(1)*. No studies have used the modern technique of DXA sequentially during pregnancy. Instead, numerous recent studies have utilized DXA before conception (range 1–8 months prior, but not always stated) and after delivery (range 1–6 weeks postpartum) *(19–25)*. Most studies involved 16 or fewer subjects. One study found no change in lumbar spine bone density measurements obtained preconception and within 1–2 weeks post-delivery *(21)*, whereas the other studies reported decreases of 4–5% in lumbar spine bone density with the postpartum measurement taken between 1–6 weeks postdelivery. The puerperium is associated with bone density losses of 1–3% per month in women who lactate (see lactation section, below), and thus it is important that the postpartum measurement be done as soon as possible after delivery. Finally, ultrasound measurements of the os calcis have been performed in longitudinal studies during pregnancy, and such studies have shown a progressive decrease in indices thought to correlate with bone mineral density *(26)*. Thus, although the longitudinal studies with SPA/DPA suggested no change in trabecular or cortical bone density during pregnancy, the subsequent evidence from pre-conception and postdelivery DXA measurements and sequential peripheral ultrasound measurements suggest that there may be a small net loss of maternal bone mineral content during normal human pregnancy. None of the aforementioned studies can address the question whether skeletal calcium content is increased early in pregnancy in advance of the third trimester, as has been observed in normal mice. Further studies, with larger numbers of patients, will be needed to clarify the extent of bone loss during pregnancy.

It seems certain that any acute changes in bone metabolism during pregnancy do not normally cause long-term changes in skeletal calcium content or strength. Numerous studies of osteoporotic or osteopenic women have failed to find a significant association of parity with bone density or fracture risk *(1,27)*. Although many of these studies could not separate out the effects of parity from those of lactation, it may be reasonable to conclude that if parity has any effect on bone density or fracture risk, it must be only a very modest effect. A more recent study of twins indicated that there may be a small protective effect of parity and lactation on maintaining bone mineral content *(28)*.

Osteoporosis in Pregnancy

Occasionally, women experience fragility fractures and are noted to have low bone mineral density during or shortly after pregnancy. Most will not have had a prior bone mineral density measurement, and thus the possibility that low bone density was present prior to pregnancy cannot be excluded. It is conceivable that the weight of the gravid uterus may precipitate vertebral compression fractures in women with low bone density. Some women may experience excessive resorption of calcium from the skeleton due to changes in mineral metabolism induced by pregnancy, or due to other factors such as low dietary calcium intake and vitamin D insufficiency. Furthermore, the apparently increased rate of bone turnover in pregnancy may contribute to fracture risk, because a high rate of bone turnover is an independent risk factor for fragility fractures outside of pregnancy. Therefore, fragility fractures in pregnancy or the puerperium may be a consequence of preexisting low bone density, loss of bone mineral content during pregnancy, and increased bone turnover, among other possible factors. Additional changes in mineral metabolism occur during lactation which may further increase fracture risk in some women (see below).

Focal, transient osteoporosis of the hip is a rare, self-limited form of pregnancy-associated osteoporosis. It is not a manifestation of altered calcitropic hormone levels or mineral balance during pregnancy, but rather might be a consequence of local factors. It is included herein for the sake of completeness and to emphasize that it is not, as its name implies, a disorder of mineral metabolism. The theories proposed to explain the condition include femoral venous stasis due to the gravid uterus, reflex sympathetic dystrophy, ischemia, trauma, viral infections, marrow hypertrophy, immobilization, and fetal pressure on the obturator nerve. These patients present with

unilateral or bilateral hip pain, limp and/or hip fracture in the third trimester. There is objective evidence of reduced bone density of the symptomatic femoral head and neck which has been shown by MRI to be the consequence of increased water content of the femoral head and the marrow cavity; a joint effusion may also be present. The symptoms and the radiological appearance usually resolve within 2–6 months post-partum.

Primary Hyperparathyroidism

Although probably a rare condition (there are no data available on its prevalence), primary hyperparathyroidism in pregnancy has been associated in the literature with an alarming rate of adverse outcomes in the fetus and neonate, including a 30% rate of spontaneous abortion or stillbirth, a 50% rate of tetany, and a 25% rate of neonatal death *(1)*. The adverse postnatal outcomes are thought to result from suppression of the fetal and neonatal parathyroid glands; this suppression may occasionally be prolonged after birth for months. Maternal PTH does not cross the placenta; the suppression of fetal parathyroids may be a consequence of increased flux of calcium across the placenta to the fetus, aided by the maternal hypercalcemia. To prevent these adverse outcomes, surgical correction of primary hyperparathyroidism during the second trimester has been almost universally recommended *(29)*. Several case series have found elective surgery to be well tolerated, and to dramatically reduce the rate of adverse events when compared to the earlier cases reported in the literature. However, many of the women in those early cases had a relatively severe form of primary hyperparathyroidism that is not often seen today (symptomatic, with nephrocalcinosis and renal insufficiency). Recent case reports suggest that the milder, asymptomatic form of primary hyperparathyroidism commonly seen today may have similar risks of adverse maternal, fetal or neonatal outcomes; consequently, parathyroidectomy continues to be mandated during pregnancy whenever the condition is identified *(30–33)*.

Familial Hypocalciuric Hypercalcemia

Although familial hypocalciuric hypercalcemia (FHH) has not been reported to adversely affect the mother during pregnancy, the maternal hypercalcemia can cause fetal and neonatal parathyroid suppression with subsequent tetany *(34)*.

Hypoparathyroidism and Pseudohypoparathyroidism

During pregnancy, hypoparathyroid women have been found to have a rise in serum calcium, fewer hypocalcemic symptoms, and to require less supplemental calcium *(1)*. This is consistent with a limited role for PTH in the pregnant woman, and suggests that an increase in 1,25-dihydroxyvitamin D and/or increased intestinal calcium absorption will occur in the absence of PTH. However, it is clear from other case reports *(1)* that some pregnant hypoparathyroid women required increased calcitriol replacement in order to avoid worsening hypocalcemia. In some reports, it was not clear whether the artifact of low total serum calcium was being treated, as opposed to a physiologically relevant (and symptomatic) low ionized calcium. It is clearly important to maintain a normal ionized calcium level in pregnant women, since maternal hypocalcemia has been associated with the development of intrauterine fetal hyperparathyroidism and fetal death. Late in pregnancy, hypercalcemia may occur in hypoparathyroid women unless the calcitriol dosage is substantially reduced or discontinued. This effect appears to be mediated by increasing levels of PTHrP in the maternal circulation in late pregnancy, an effect that becomes more pronounced during lactation (see below).

In limited case reports of pseudohypoparathyroidism (an inherited condition manifest by resistance to PTH), pregnancy has been noted to normalize the serum calcium level, reduce the PTH level by half, and increase the 1,25-dihydroxyvitamin D level 2- to 3-fold *(35)*. The presence of PTHrP cannot explain the improvements because such patients are resistant to the actions of both PTH and PTHrP. The mechanism by which pseudohypoparathyroidism is improved in pregnancy remains unclear, but it may be a post-receptor effect.

Vitamin D Deficiency

Vitamin D insufficiency and deficiency have not been sufficiently studied in humans to understand what levels of supplementation are necessary or ideal *(36)*, but adequate vitamin D supplementation of the mother should be maintained throughout pregnancy. The evidence cited above has established how intestinal calcium absorption is normally doubled during pregnancy to meet the fetal demand for mineral and maintain maternal calcium homeostasis; in the absence of that vitamin D-dependent effect on

intestinal calcium transfer, one can anticipate that the mother would have difficulty maintaining the mineral supply without compromising her skeleton and perhaps the fetus as well. However, there is a paucity of clinical data on the effects of pregnancy on mineral metabolism in women who are vitamin D deficient. Clearly chronic maternal hypocalcemia of any cause is potentially detrimental to human fetuses. Evidence from animal models of vitamin D deficiency and absence of the vitamin D receptor indicates that the mothers have impaired fertility and smaller gestational litter sizes; the fetuses are also smaller but have normal serum calcium and fully mineralized skeletons *(37–40)*. The animal models indicate that the mother may suffer the brunt of the effects of severe hypocalcemia in late pregnancy, including tetany, seizures, cardiac arrhythmias, and death *(37,41)*. In both humans and in animals, vitamin D deficiency and absence of the vitamin D receptor can lead to very profound effects in the neonate, including rickets and craniotabes (see below on Lactation).

Low Calcium Intake

Intuitively, limited maternal intake of calcium and other minerals may adversely affect fetal skeletal development, or perhaps lead to severe losses of maternal bone mineral content during pregnancy. Most of the studies cited in this review have involved women who were well nourished, and who may also have been taking a calcium supplement during pregnancy. Limited information is available from women who customarily had low (<300 mg/day) dietary intakes of calcium, with discrepant results as to whether or not calcium supplementation altered the maternal bone density response during pregnancy, or the neonatal bone density at birth *(3)*. More recently, a short crossover trial of 1.2 g calcium supplementation daily (10 days on supplement, 10 days on placebo) in Mexican women during pregnancy demonstrated that a bone resorption marker was suppressed in the third trimester by the addition of a calcium supplement. Thus, there is at least short-term evidence that bone turnover in the mother can be suppressed by supplemental calcium intake. Also, a double-blind study of 2 g calcium supplementation during pregnancy in 256 women demonstrated that the neonatal bone mineral content was improved in the infants of calcium-supplemented mothers who were in the lowest quintile of calcium intake versus the infants of mothers in the same quintile that took a placebo *(42)*. There was no benefit of calcium supplementation to infants whose mothers were in the higher quintiles of dietary calcium intake. Thus, there is limited evidence that low calcium intake might adversely affect fetal development, and it remains especially prudent to recommend calcium supplementation during pregnancy to women known to have low dietary intake of calcium.

LACTATION

About 280 to 400 mg of calcium is lost through breast milk daily, with losses as great as 1000 mg calcium or more in women who are nursing twins. A temporary demineralization of the skeleton appears to be the main mechanism by which lactating humans meet these calcium requirements. This demineralization does not appear to be mediated by PTH or 1,25-dihydroxyvitamin D. Instead, it is mediated by PTHrP released from the breast tissue, combined with the effects of low estrogen levels on bone turnover.

Mineral Ions and Calcitropic Hormones

The normal lactational changes in maternal calcium, phosphorus, and calcitropic hormone levels are schematically depicted in Fig. 2 *(1)*. The mean ionized calcium level of exclusively lactating women is increased, although it remains within the normal range. Serum phosphorus levels are also higher during lactation, and the level may exceed the normal range. The serum phosphorus is likely increased due to accelerated skeletal resorption combined with reduced renal phosphorus excretion.

Intact PTH, as determined by a 2-site IRMA assay, has been found to be reduced 50% or more in lactating women during the first several months. It rises to normal at weaning, but may rise above normal post-weaning. In contrast to the high 1,25-dihydroxyvitamin D levels of pregnancy, maternal free and bound 1,25-dihydroxyvitamin D levels fall to normal within days of parturition and remain there throughout lactation.

PTHrP levels are significantly higher in lactating women and mice than in non-lactating controls, as measured by 2-site IRMA assays. The source of PTHrP appears be the breast or mammary tissue, since PTHrP has been detected in milk at concentrations exceeding 10,000 times the level found in the

blood of patients with hypercalcemia of malignancy or normal human controls. A small rise in the maternal level of PTHrP can be demonstrated after suckling *(43)*. Furthermore, blood levels of PTHrP are reduced in lactating mice that had the PTHrP gene ablated only from breast tissue, as compared to normal lactating mice *(44)*. PTHrP appears to play several roles within the breast, as indicated by studies in animals that suggest that PTHrP may regulate mammary development and blood flow. The calcium receptor is expressed in mammary tissue during lactation, where it regulates PTHrP production as well as the calcium and water content of the milk (Fig. 3) *(45)*. It is also expressed in the developing and virgin mammary tissue, but not in mammary tissue during pregnancy.

It has become clear that PTHrP plays a key role during lactation in regulating the demineralization of the skeleton. In response to suckling *(43)* and signaling from the calcium sensing receptor expressed by lactating mammary tissue (Fig. 3) *(45)*, PTHrP reaches the maternal circulation from mammary tissue and stimulates resorption of calcium from the maternal skeleton, renal tubular reabsorption of calcium, and (indirectly) suppression of PTH (Fig. 4). In a sense, the breast becomes an accessory parathyroid gland during lactation, but the "hyperparathyroidism" of lactation is increased secretion of PTHrP and not PTH. The strongest evidence in support of this model comes from the study of mice in which the PTHrP gene was ablated at the onset of lactation but only within mammary tissue *(44)*. The lactational decrease in bone mineral content was significantly blunted in the absence of mammary gland production of PTHrP. Other evidence for the central role of PTHrP in lactation comes from humans, in that PTHrP levels correlate negatively with PTH levels and positively with the ionized calcium levels of lactating women *(43,46)*, and that higher PTHrP levels correlate with greater losses of bone mineral density during lactation in humans *(47)*. Furthermore, observations in aparathyroid women provide evidence of the impact of PTHrP in calcium homeostasis during lactation (see below).

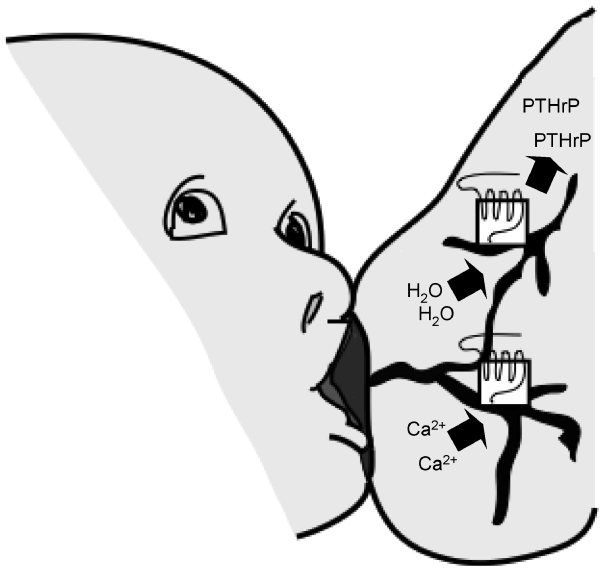

Fig. 3. The role of the calcium receptor in the breast. The calcium receptor (represented schematically) is expressed in the breast during lactation, wherein it has several key functions as elucidated from recent studies in mice *(45)*. The calcium receptor monitors the systemic concentration of calcium to control PTHrP synthesis and, thereby, the supply of calcium to the breast. An increase in systemic calcium or administration of a calcimimetic inhibited PTHrP expression, whereas a decrease in systemic calcium stimulated PTHrP expression. The calcium receptor also directly regulates the calcium and fluid composition of milk. Administration of calcium or a calcimimetic stimulated the transport of calcium into the breast, and the administration of a calcimimetic also enhanced the entry of water into the milk, thereby making it less viscous.

Calcitonin levels are elevated in the first 6 weeks of lactation. Recent studies in mice lacking the gene that encodes calcitonin indicate that calcitonin may modulate the rate of skeletal resorption during lactation. Calcitonin-null mice lost more than 50% of skeletal mineral content during 3 weeks of lactation, approximately twice that of normal littermate sisters *(13)*. The calcitonin-depleted mice still regained all of the lost mineral content after weaning, which indicates that although calcitonin is needed in the short-term to prevent severe losses of mineral content and potential skeletal fragility, calcitonin is not required in the long-term because the skeletal losses of mineral are restored anyway.

Intestinal Absorption of Calcium

Intestinal calcium absorption decreases to the non-pregnant rate from the increased rate of pregnancy. This corresponds to the fall in 1,25-dihydroxyvitamin D levels to normal.

Renal Handling of Calcium

In humans, the GFR falls during lactation, and the renal excretion of calcium is typically reduced to

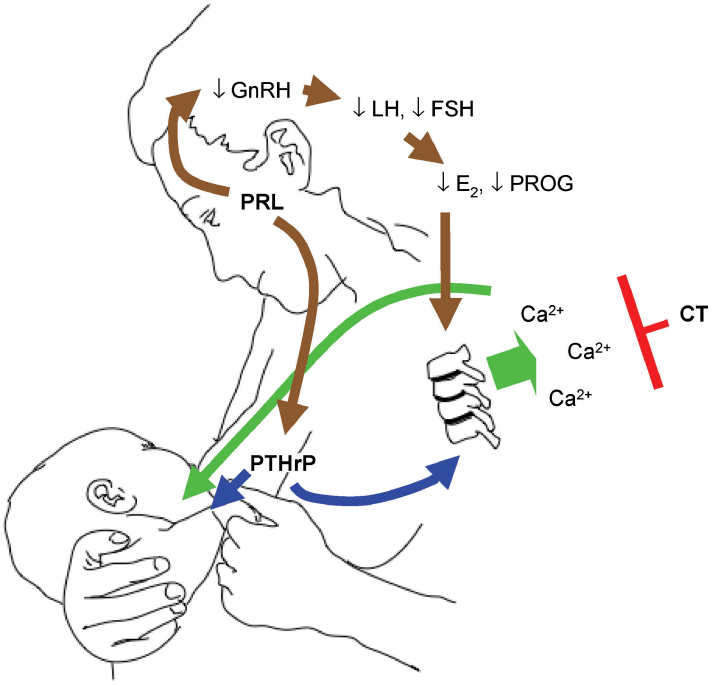

Fig. 4. The role of the breast in controlling skeletal demineralization during lactation. Suckling induces release of prolactin. Suckling and prolactin both inhibit the hypothalamic GnRH pulse center, which in turn suppresses the gonadotropins (LH, FSH), leading to low levels of the ovarian sex steroids (estradiol and progesterone). PTHrP production and release from the breast is controlled by several factors, including suckling, prolactin, and the calcium receptor. PTHrP enters the bloodstream and combines with systemically low estradiol levels to markedly upregulate bone resorption. Increased bone resorption releases calcium and phosphate into the blood stream, which then reaches the breast ducts and is actively pumped into the breast milk. PTHrP also passes into the milk at high concentrations, but whether PTHrP plays a role in regulating calcium physiology of the neonate is unknown. Calcitonin (CT) modulates the skeletal responsiveness, as demonstrated by a doubling of mineral losses from the skeleton in mice that lack the gene that encodes calcitonin.

very low levels. This observation suggests that the tubular reabsorption of calcium must be increased in order to account for reduced calcium excretion in the setting of increased serum calcium.

Skeletal Calcium Metabolism

Histomorphometric data from animals consistently show increased bone turnover during lactation, and losses of 30% of bone mineral are achieved during 2–3 weeks of normal lactation in the rat [reviewed in *(1)*] while a similar amount is lost in the lactating mouse within 21 days *(17)*. The loss is greatest in trabecular bone of rats and mice. Comparative histomorphometric data are lacking for humans, and in place of that, serum markers of bone formation, and urinary markers of bone resorption have been assessed in numerous cross-sectional and prospective studies of lactation. Some of the confounding factors discussed with respect to pregnancy apply to the use of these markers in lactating women. During lactation, GFR is reduced and the intravascular volume is more contracted. Urinary markers of bone resorption (24-h collection) have been reported to be elevated 2–3 fold during lactation and to be higher than the levels attained in the third trimester. Serum markers of bone formation (not adjusted for hemoconcentration or reduced GFR) are generally high during lactation, and increased

over the levels attained during the third trimester. Total alkaline phosphatase falls immediately postpartum due to loss of the placental fraction, but may still remain above normal due to the elevation in the bone-specific fraction. Despite the confounding variables, these findings suggest that bone turnover is significantly increased during lactation.

In women, serial measurements of bone density during lactation (by SPA, DPA or DXA) have shown a fall of 3 to 10.0% in bone mineral content after two to 6 months of lactation at trabecular sites (lumbar spine, hip, femur and distal radius), with smaller losses at cortical sites *(1,27)*. The peak rate of loss is 1–3% per month, far exceeding the rate of 1–3% per year that can occur in women with postmenopausal osteoporosis who are considered to be losing bone rapidly. Loss of bone mineral from the maternal skeleton appears to be a normal consequence of lactation and may not be preventable by raising the calcium intake above the recommended dietary allowance. Several recent studies have demonstrated that calcium supplementation does not significantly reduce the amount of bone lost during lactation *(48–51)*. The lactational decrease in bone mineral density correlates with the amount of calcium lost in the breast milk *(52)*.

The mechanisms controlling the rapid loss of skeletal calcium content are not well understood. The reduced estrogen levels of lactation are clearly important, but are unlikely to be the sole explanation. To estimate the effects of estrogen deficiency during lactation, it is worth noting the alterations in calcium and bone metabolism that occur in reproductive-age women who have estrogen deficiency induced by GnRH agonist therapy for endometriosis and other conditions. Six months of acute estrogen deficiency induced by GnRH agonist therapy leads to 1–4% losses in trabecular (but not cortical) bone density, increased urinary calcium excretion and suppression of 1,25-dihydroxyvitamin D and PTH levels *(1)*. During lactation, women are not as estrogen deficient but lose more bone mineral density (at both trabecular and cortical sites), have normal (as opposed to low) 1,25-dihydroxyvitamin D levels, and have reduced (as opposed to increased) urinary calcium excretion. The difference between isolated estrogen deficiency and lactation may be due to the effects of other factors (such as PTHrP) that add to the effects of estrogen withdrawal in lactation (Fig. 5). The relative influences of estrogen deficiency and PTHrP have been partly determined in normal mice, in which it has been demonstrated that treatment with pharmacological doses of estrogen blunted but did not abolish the normal demineralization that occurs during lactation *(53)*.

The bone density losses of lactation are substantially reversed during weaning at a rate of 0.5 to 2% per month *(1,27,50)*. The mechanism for this restoration of bone density is uncertain and largely unexplored; there is preliminary evidence from animal

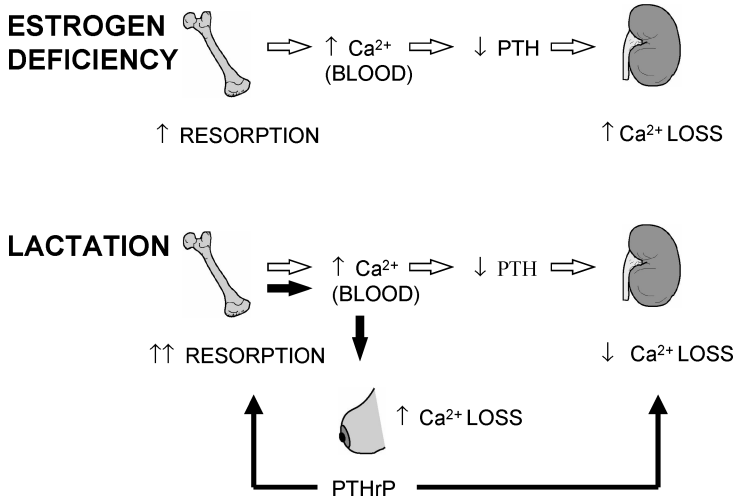

Fig. 5. The roles of estrogen deficiency and PTHrP. Acute estrogen deficiency (e.g., GnRH analog therapy) increases skeletal resorption and raises the blood calcium; in turn, PTH is suppressed and renal calcium losses are increased. During lactation, the combined effects of PTHrP (secreted by the breast) and estrogen deficiency increase skeletal resorption, reduce renal calcium losses, and raise the blood calcium, but calcium is directed into breast milk. Reprinted with permission from ref. *(1)*, ©1997 The Endocrine Society.

models to suggest that PTH, 1,25-dihydroxyvitamin D, calcitonin, and estrogen may not be required for the regain to be achieved. In the long-term, the consequences of lactation-induced depletion of bone mineral appear clinically unimportant. The vast majority of epidemiologic studies of pre- and postmenopausal women have found no adverse effect of a history of lactation on peak bone mass, bone density, or hip fracture risk.

Osteoporosis of Lactation

Rarely, a woman will suffer a fragility fracture during lactation, and osteoporotic readings will be confirmed by DXA. Like osteoporosis in pregnancy, this may represent a coincidental, unrelated disease; the woman may have had low bone density prior to conception. Alternatively, some cases might represent an exacerbation of the normal degree of skeletal demineralization that occurs during lactation. For example, excessive PTHrP release from the lactating breast into the maternal circulation might cause excessive bone resorption, osteoporosis, and fractures. PTHrP levels were high in one case of lactational osteoporosis, and were found to remain elevated for months after weaning *(54)*. Some cases of lactational osteoporosis might represent the human equivalent of calcitonin deficiency, based on the evidence from the murine model that absence of calcitonin exacerbates the normal losses of mineral during lactation. Future cases of lactational osteoporosis should be more extensively assessed to determine whether PTHrP excess or calcitonin deficiency are to blame.

Hypoparathyroidism and Pseudohypoparathyroidism

Calcitriol requirements of hypoparathyroid women fall early in the postpartum period, especially if a woman breastfeeds, and hypercalcemia may occur if the calcitriol dosage is not substantially reduced *(55)*. As observed in one seminal case, this is consistent with PTHrP reaching the maternal circulation in amounts sufficient to allow stimulation of 1,25-dihydroxyvitamin D synthesis, and maintenance of normal (or slightly increased) maternal serum calcium *(56)*.

The effect of lactation on pseudohypoparathyroidism has been less well documented. Since these patients are likely resistant to the renal actions of PTHrP, and the placental sources of 1,25-dihydroxyvitamin D are lost at parturition, the calcitriol requirements might well increase and may require further adjustments during lactation.

Vitamin D Deficiency

Skeletal demineralization occurs during lactation due to PTHrP-stimulated bone resorption and the low estradiol levels, and it appears that vitamin D sufficiency may not be required to maintain lactation. Even in women with very low calcium intakes, the same amount of mineral was lost from the skeleton as compared to women who had supplemented calcium intakes, and the breast milk calcium content was unaffected by calcium intake or vitamin D status *(57–59)*. Conversely, since high calcium intakes do not affect the degree of skeletal demineralization that occurs during lactation *(48–51)*, it is unlikely that increasing vitamin D supplementation above normal would affect skeletal demineralization either. Whether vitamin D deficiency affects the ability to restore the skeleton after weaning has not been examined.

It remains prudent to advise vitamin D supplementation for the mother during lactation for her own long-term skeletal health. Supplementation of the mother will indirectly lead to higher levels of vitamin D in the nursing infant, although higher doses than normal are required due to the low penetrance of vitamin D into breast milk *(36)*. Consequently, breast fed babies of vitamin D deficient mothers should directly receive vitamin D supplements to avoid nutritional rickets. Evidence from animal models suggests that lactation can provoke tetany and sudden death in mothers that are vitamin D deficient, perhaps because the lactational drain of mineral overwhelms the mother's ability to maintain a normal ionized calcium level. However, such extreme outcomes have not been reported for vitamin D deficient women who lactate.

IMPLICATIONS

The studies of pregnant women suggest that the fetal calcium demand is met in large part by intestinal calcium absorption, which more than doubles from early in pregnancy. The studies of biochemical markers of bone turnover, DXA and ultrasound are not conclusive, but are compatible with the possibility that the maternal skeleton contributes calcium to the developing fetus. In comparison,

the studies in lactating women suggest that skeletal calcium resorption is a dominant mechanism by which calcium is supplied to the breast milk, while renal calcium conservation is also apparent. These observations indicate that the maternal adaptations to pregnancy and lactation have evolved differently over time, such that dietary calcium absorption dominates in pregnancy, whereas the temporary borrowing of calcium from the skeleton dominates during lactation. Lactation programs an obligatory skeletal calcium loss irrespective of maternal calcium intake, but the calcium is completely restored to the skeleton after weaning. The rapidity of calcium loss and regain by the skeleton of the lactating woman are through mechanisms that are at best, only partly understood. A full elucidation of the mechanisms of bone loss and restoration in the lactating woman might lead to the development of novel approaches to the treatment of osteoporosis and other metabolic bone diseases. Finally, while it is apparent that some women will experience fragility fractures as a consequence of pregnancy or lactation, the vast majority of women can be reassured that the changes in calcium and bone metabolism during pregnancy and lactation are normal, healthy, and without adverse consequences in the long-term.

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