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Could use of Selective Serotonin Reuptake Inhibitors During Lactation Cause Persistent Effects on Maternal Bone?

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Abstract
The lactating mammary gland elegantly coordinates maternal homeostasis to provide calcium for milk. During lactation, the monoamine serotonin regulates the synthesis and release of various mammary gland-derived factors, such as parathyroid hormone-related protein (PTHrP), to stimulate bone resorption. Recent evidence suggests that bone mineral lost during prolonged lactation is not fully recovered following weaning, possibly putting women at increased risk of fracture or osteoporosis. Selective Serotonin Reuptake Inhibitor (SSRI) antidepressants have also been associated with reduced bone mineral density and increased fracture risk. Therefore, SSRI exposure while breastfeeding may exacerbate lactational bone loss, compromising long-term bone health. Through an examination of serotonin and calcium homeostasis during lactation, lactational bone turnover and post-weaning recovery of bone mineral, and the effect of peripartum depression and SSRI on the mammary gland and bone, this review will discuss the hypothesis that peripartum SSRI exposure causes persistent reductions in bone mineral density through mammary-derived PTHrP signaling with bone.

Keywords serotonin · lactation · bone · Selective Serotonin Reuptake Inhibitor (SSRI)

Introduction

Lactation is an evolutionary strategy that pre-dates the origin of mammals. The mammary gland produces milk for the offspring in response to systemic and local hormonal cues. It is theorized that the mammary gland evolved from a modified hair follicle into a secreting apocrine gland to provide moisture and antimicrobial properties to shelled eggs [1]. Fossil records from more than 200 million years ago in the Triassic period suggest the presence of both lactose and the casein family of milk proteins, which are crucial for the provision of calcium, phosphate, and complex proteins to hatchlings. With the evolution of placental reproduction, this milk-like fluid diverged from solely providing nutrients to the egg to become the complex milk that characterizes eutherian lactation [2].

Over 200 years ago, the mammary gland was a defining feature in the taxonomic grouping of species into the class of Mammalia [1]. Throughout evolution, the mammary gland has adapted to provide species- and environment-specific nutrition for the offspring, thereby ensuring reproductive success. Nursing promotes maternal-offspring bonding to support neonate survival [3, 4]. Not only does milk contain all of the nutrients necessary to nourish the neonate, but it also contains bioactive factors that confer immunological and endocrine competence to both dam and infant [5]. The mammary gland coordinates a series of elegant signaling cascades to provide calcium for milk, a process crucial for infant skeletalization. The monoamine serotonin regulates maternal calcium homeostasis by stimulating mammary-derived endocrine signals, including the synthesis of parathyroid hormone-related protein (**PTHrP**), a potent stimulator of bone resorption [6]. Selective serotonin reuptake inhibitor (**SSRI**) antidepressants elevate cellular exposure to serotonin and have been independently associated with reduced bone mineral density (**BMD**) [7]. Lactational SSRI exposure may stimulate epigenetically-regulated, serotonin-driven signals from the mammary gland, including PTHrP, thereby causing excessive bone resorption during lactation from which women are unable to recover post-weaning. This review will examine evidence both supporting and refuting the hypothesis that SSRIs and lactation cause persistent, post-weaning effects on BMD (Fig. 1).

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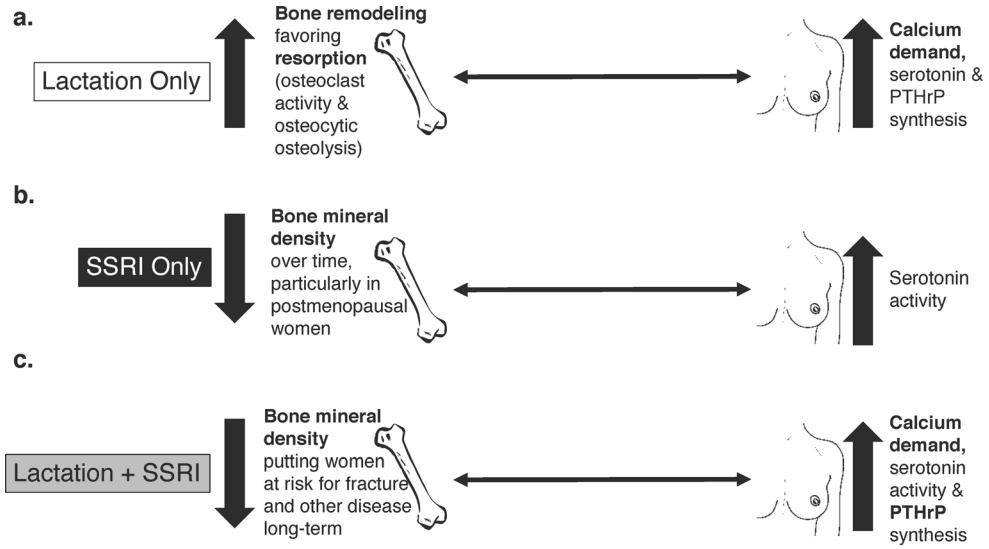

Fig. 1 Breast to bone communication during lactation, with SSRI exposure, and while breastfeeding with concurrent SSRI exposure. a. During lactation, elevated calcium demand from the mammary gland coordinates serotonin and PTHrP signaling. Bone tissue during lactation undergoes significant remodeling through both osteoclastic bone resorption and osteocytic osteolysis. Due to PTHrP binding to its receptor on bone, resorption is favored over formation. **b**. Women taking an SSRI have elevated serotonin activity at both the breast and the bone, due to inhibition of the serotonin transporter. SSRIs have been associated with decreased BMD and high fracture risk, particularly in postmenopausal women. **c**. Women who are breastfeeding while taking an SSRI may be at risk for reduced BMD long-term, due to elevated bone resorption associated with lactation and SSRI exposure.

Serotonin

Serotonin Synthesis and Physiological Distribution

To understand how lactational SSRI exposure could predispose women to long-term bone loss, mammary-derived signals known to regulate bone must be discussed. Serotonin was originally characterized in 1948 in the blood as a vasoconstrictor of large vessels [8], but its role as an intracellular signaling molecule has been preserved throughout the last two billion years of evolution [9]. Along with catecholamines, indoleamines such as serotonin and melatonin are derived from tryptophan. L-tryptophan is converted into 5-hydroxy-L-tryptophan (**5-HTP**) by the rate-limiting enzyme TPH. Amino acid decarboxylase (**AADC**) then converts 5-HTP to 5-hydroxy- tryptamine (**5-HT**; serotonin) in a non-specific enzymatic reaction. There are two major isoforms of TPH: one that acts predominately in the central nervous system (**TPH2**), and a second isoform that is ubiquitous in the periphery (**TPH1**). TPH2 is expressed in the dorsal raphe nuclei and myenteric plexus neurons in the intestine, while TPH1 is abundant in the pineal gland and enterochromaffin cells, along with a variety of other peripheral tissues including the liver, pancreas, adipose tissue, and heart [10–14]. The TPH1 enzyme has been identified in the mammary gland of mice, rats, humans, bovines, and bats [15–18]. Although 5-HTP can cross the blood-brain-barrier, serotonin cannot. L-tryptophan can also cross the blood-brain barrier, but it must compete with other branched-chain amino acids for a common amino acid transporter [19]. L-tryptophan is also the precursor for the kynurenine pathway, which ultimately leads to the biosynthesis of nicotinamide adenine dinucleotide (**NAD^+**). Kynurenines play a role in various central nervous system disorders, as well as immunoregulation [20]. Of the tryptophan that remains after requirements are met for protein synthesis, approximately 90% is directed towards kynurenines, while only 3% of dietary tryptophan is used for serotonin synthesis [21, 22]. The peripheral and neuronal pools of serotonin are largely considered independent of one another [23, 24].

Serotonin can bind to any one of its at least 14 cell-surface receptors (**5HTR**), which are grouped into seven families and expressed ubiquitously throughout the mammalian body [25]. Aside from the type 3 receptors, which are ligand-gated ion channels, all serotonin receptors are G-protein coupled (**GPCR**). 5HTR1 receptors (1a, 1b, 1d, 1e, 1f) are coupled to $G_{i/o}$ and negatively regulate cyclic AMP (**cAMP**) formation. The 5HTR2a, 2b, and 2c receptors preferentially couple to $G_{q/11}$ to increase inositol phosphates and cytosolic calcium concentrations. The final receptor subtypes, including 5HTR4, 5HTR6, and 5HTR7, couple to G_s GPCRs to promote cAMP formation via activation of adenylate cyclases. Various 5HTRs, such as 2c and 7, can splice to yield different isoforms, adding to the complex diversity of serotonin receptors [25]. All of the serotonin receptor families except for 5HTR6 have been identified in the mammary gland [26–28]. In particular, 5HTR7 has been implicated in the regulation of mammary epithelial cell shape and secretory activity [29–31], while 5HTR2b has a role in regulating calcium and mammary-to-bone signaling during lactation [28, 32].

In the pineal gland and the retina, serotonin can be converted to melatonin by serotonin N-acetyltransferase (**SNAT**) and hydroxyindole-O-methyltransferase [33]. SNAT is not expressed in the mammary gland [15]. Instead, serotonin is catabolized into its inactive form (5-hydroxyindole acetylaldehyde) by the enzyme monoamine oxidase (**MAO**). 5-hydroxyindole acetylaldehyde is then converted to 5-hydroxyindole acetic acid (**5HIAA**) by aldehyde dehydrogenase. 5-HIAA can be detected in plasma and its excretion in urine is used as a marker of whole body serotonin turnover [34].

In circulating blood, serotonin is stored in platelet granules [35]. Platelets do not contain TPH enzymes, but instead take up serotonin from the periphery using the ubiquitous serotonin reuptake transporter (**SERT**). During platelet activation, serotonin is secreted from the platelet granules, prompting further platelet aggregation and vasoconstriction of surrounding blood vessels [36, 37]. In tissues, SERT removes serotonin from the extracellular space, effectively terminating cell surface receptor-mediated signaling. Approximately 95% of peripheral serotonin is synthesized in the enterochromaffin cells of the gut in a non-lactating animal [38, 39]. However, during lactation, the mammary gland contributes more significantly to circulating serotonin stores than the gut [40]. As such, the mammary gland coordinates serotonin homeostasis during lactation.

Serotonin and Epigenetics

Mammary gland serotonin may coordinate breast-to-bone signaling through epigenetic mechanisms. Epigenetics is defined as perturbations in the genome that regulate gene transcription without modifying the underlying DNA sequence [41]. DNA is packaged in the form of chromatin, with nucleosomes as the basic structural repeating unit. Each nucleosome contains 147 base pairs of DNA wrapped twice around an octamer consisting of two copies of each core histone protein (H2A, H2B, H3, H4). Biochemical modifications of the histone's globular domain or amino-acid tail cause the histone to be "open" where the subunits are spaced apart and the DNA is accessible to transcriptional machinery. Conversely, histones can be "closed" with DNA tightly packed together and inaccessible to transcriptional machinery. The epigenetic modifications that regulate the state of chromatin fall into two main categories: DNA methylation and histone modifications [42, 43]. While the longstanding paradigm has been that epigenetic modifications are stable throughout a lifespan, there is increasing evidence that epigenetic regulation is more plastic than formerly believed [44–46].

DNA methylation is defined as the addition of a methyl group to the C5 position of cytosine at CpG islands, which are regions consisting of a high (>55%) CG content [42]. DNA methyltransferases (**DNMT**) catalyze methylation events. DNMT3a and DNMT3b establish new patterns of *de novo* methylation during embryonic development. The methylation pattern is then maintained by DNMT1 during cell division, transferring methyl groups to hemi-methylated DNA strands following replication [47]. Methylation of cytosine to generate 5-methylcytosine is not the only covalent modification of DNA. CpG dinucleotides can be oxidized and converted to a variety of co-modifications to include: 5-hydroxymethylcytosine [48, 49], 5-formylcytosine, and 5-carboxylcytosine [50] by the ten-eleven translocase (**TET**) family of DNA dioxygenases, a family which can also demethylate DNA [51]. DNA methylation is a powerful means by which gene expression can be post-transcriptionally modified to regulate molecular signaling.

Components of the serotonin pathway are subject to epigenetic regulation involving DNA methylation. Methylation of the *Sert* promoter in the brain has been shown to predict amygdala reactivity [52] and greater *Sert* promoter methylation in peripheral blood mononuclear cells was associated with exacerbated effects of early-life stress in infant Rhesus macaques [53]. In the sea slug *Aplysia* central nervous system, serotonin induced methylation of the promoter region of the *Creb2* gene, which is implicated in the persistence of memory [54]. DNA methylation of the *Sert* promoter in human placental cells isolated from the fetal side was reduced in mothers with gestational diabetes mellitus compared to control mothers, suggesting that DNA methylation of the fetus is sensitive to the metabolic status of the mother [55]. Finally, the *Mao* promoter is hypermethylated in human cholangiocarcinoma [56]. Recently, it has been shown that serotonin itself can be transamidated to glutamine residues by the enzyme transglutaminase. Termed "serotonylation", this process has been implicated in platelet activation [57], insulin release from pancreatic β-cells [58], smooth muscle contraction [59], and glucose metabolism in skeletal muscle [60]. Though still a relatively new field, the potential that serotonin itself can act as an epigenetic modifier reveals promising research opportunities [61].

Serotonin induces epigenetic modifications in the lactating mammary gland. The presence or absence of serotonin in the mammary gland is correlated with hypo- or hyper-methylation of the sonic hedgehog (***Shh***) promoter during lactation. SHH is expressed in mammary tissue from puberty to lactation [62, 63] and has been implicated in breast cancer pathogenesis [64, 65]. When a hedgehog (**HH**) ligand binds to patched-1 (**PTCH1**), its 12-pass transmembrane protein receptor, smoothened (**SMO**) can accumulate intracellularly and trigger downstream gene transcription of glioblastoma (**GLI**) transcription factors 1, 2, and 3, which have both shared and divergent regulatory roles [66]. Serotonin alters methylation patterns of the *Shh* promoter in lactating mouse mammary glands. SHH is upstream of PTHrP, a potent regulator of bone resorption and therefore maternal calcium homeostasis [67]. Accordingly, serotonin-driven reductions in *Shh* promoter methylation are associated with increased SHH signaling, elevated PTHrP synthesis and secretion from the mammary

gland, and increased markers of bone resorption and serum calcium [68]. During lactation, serotonin epigenetically regulates signaling in the lactating mammary gland that drives whole-body perturbations in calcium homeostasis. This information is important groundwork for understanding the mechanism by which SSRIs, which increase serotonin signaling, may impact maternal BMD.

Serotonin in Pregnancy: Effects on the Dam

Women are at great risk for being diagnosed with depression during their childbearing years [69]. SSRIs are considered the first choice for pregnant and breastfeeding women [70, 71] and elevate serotonin activity throughout the body. To appreciate the potential effects of SSRIs on mammary gland and bone health, it is imperative to understand serotonergic biology during pregnancy and lactation. Serotonin biosynthesis in the mammary gland was discovered while performing genetic screens of the prolactin-knockout mouse [15]. Shortly prior to this discovery, the TPH1 knockout (**TPH1 KO**) mouse was developed by Walther and colleagues [72]. The TPH1 KO mouse had lobuloalveoli that were hypersensitive to prolactin, and TPH1 KO mammary glands failed to undergo involution even after three days of milk stasis [15]. Development of the TPH1 KO mouse and characterization of mammary serotonin receptors, have inspired research into serotonergic regulation of pregnancy and lactation.

Serotonin is associated with development and various disease states during pregnancy. *Tph1* expression increases in pancreatic β-cells during pregnancy concurrent with an increase in serotonin synthesis and storage in pancreatic islets [73–75]. Acting through the 5HTR3 family, serotonin decreases the β-cell threshold for glucose, increasing glucose-stimulated insulin secretion during pregnancy [76]. Both serotonin content and *Tph1* mRNA expression are increased in the hearts of pregnant versus non-pregnant mice and 5HTR3A knockout dams experience sudden cardiac death during late pregnancy [77]. Serotonin also regulates uterine contractility [78], with various 5HTR subtypes identified in the bovine uterus [79]. Guinea pig uterine and carotid arteries were less responsive to serotonin as a contractile agent when the tissues were isolated from pregnant versus non-pregnant animals [80]. In women, high serotonin concentrations are associated with both preeclampsia [81, 82] and hyperemesis gravidarum (severe nausea) [83]. During the transition from pregnancy to lactation, 5HTR expression either increases or decreases in the liver of dairy cows and mice [84, 85], depending on the subtype of serotonin receptor. Finally, cows milked one day prepartum had greater serum and colostrum serotonin concentrations compared to cows milked four hours postpartum, implicating that mammary gland serotonin is sensitive to parturition [86]. Due to the wide variety of serotonin receptors distributed throughout the body, serotonin has essential, but widely varied, roles in regulating normal pregnancy, as well as pathologies associated with pregnancy. Although SSRIs are considered the safest antidepressant during pregnancy, dysregulated serotonin signaling from SSRIs may produce a wide range of effects on peripartum maternal homeostasis.

Serotonin in Pregnancy: Effects on the Offspring

Serotonin is a key regulatory signaling molecule in development. Pups born to TPH1 KO mice have previously been shown to be smaller than those of heterozygous or wild type (**WT**) mice [87], with fewer pups per litter [88]. Various groups have characterized a complete serotonergic network in the placenta and trophoblast cell lines [89–91], suggesting that serotonin may regulate the maternal-fetal interface. Serotonin is best known for its role in the brain. Both SERT and various 5HTRs are expressed early in brain development [92], and their dysregulation is associated with neurodevelopmental disorders such as autism spectrum disorder and schizophrenia [93, 94]. Throughout pregnancy, there is a progressive switch from placental to fetal production of serotonin, highlighting serotonin's crucial role in development [95]. The birth of mouse pups regulates the initiation of cell cluster (barrel) formation in the somatosensory cortex by reducing serotonin content, informing sensory map formation [96]. Additionally, work in Rhesus macaques has shown that offspring with the s allele of SERT that were exposed to alcohol during gestation had lower 5HIAA concentrations in their cerebrospinal fluid as adults compared to exposed offspring with the l/l SERT allele [97].

Obesity and dysregulated fat metabolism during pregnancy may adversely affect serotonin homeostasis in the dam and infant. Japanese macaques that consumed a high fat diet (**HFD**) throughout pregnancy developed fetuses with perturbed serotonin signaling in the brain [98]. Additionally, TPH1 KO mice fed a HFD throughout pregnancy were able to lactate at the onset of lactation, while control mice on the HFD could not. In obese mothers, serotonin may inhibit lactogenesis and reduced mammary serotonergic signaling could be beneficial [99]. Given the complexity of serotonin receptor subtypes in terms of tissue spatial and temporal distribution [100], serotonin can have a wide and varied impact during pregnancy that should be carefully considered when administering SSRIs that manipulate serotonergic biology.

Serotonin in Lactation

Serotonin is a homeostatic regulator of lactation [101]. Mammary gland serotonin was first discovered as part of an autocrine-paracrine feedback loop that inhibits mammary gland development and milk synthesis [15]. Intramammary infusion of serotonin decreased milk yield and intramammary infusion of 5-HTP at dry-off in dairy cattle accelerated involution in dairy cows [16, 102]. Additionally, the 5HTR1b and

2a receptors have been associated with milk production in dairy cattle [103, 104]. When serotonin was administered to the basolateral side of normal mammary breast line MCF10A cells, transepithelial electrical resistance (**TEER**) was negatively affected, suggesting a breakdown of tight junctions [28, 105]. Tight junctions between mammary epithelial cells (**MECs**) must remain tightly sealed in order to prevent milk constituents from leaking from the lumen between bouts of nursing [105]. It was later shown that serotonergic regulation of MEC tight junctions is biphasic in mouse and human mammary epithelium. At low concentrations, serotonin potentiates the sealing of tight junctions, increasing TEER and preventing milk leakage from the lumen. By contrast, with longer exposure and at higher concentrations, serotonin decreases TEER and disrupts tight junctions between MECs [27], specifically by signaling through the 5HTR7 class of receptors [27, 28, 30]. At high concentrations, serotonin also directly reduces milk protein gene expression in mouse and bovine MECs [15, 16].

Serotonin regulates maternal calcium homeostasis during lactation. Serotonin and calcium concentrations are elevated in the serum of lactating versus nulliparous mice [106, 107]. Work from our lab has shown that TPH1 KO dams have decreased calcium concentrations on days 1 and 10 of lactation compared to WT dams. TPH1 KO mice also have less mammary gland 5HTR2b mRNA and protein as well as reduced expression of key calcium sensors and transporters in the mammary gland, including calcium release-activated calcium channel 1 (**ORAI1**), and plasma membrane Ca2+-ATPase 2 (**PMCA2**) [31]. Basolateral ORAI1 expression in the MEC is required for optimal calcium transport into milk as well as milk ejection [108]. On the apical side of the MEC, PMCA2 is responsible for pumping 60 to 70% of calcium into milk [109] and is stimulated by basolateral calcium sensing receptor (**CaSR**) activation [110]. TPH1 KO dams also have reduced CaSR mRNA and protein expression during lactation [31]. Perturbed calcium signaling in TPH1 KO dams was later correlated to serotonin's epigenetic regulation of SHH expression and downstream activation of PTHrP [68].

Serotonin induces the synthesis and secretion of PTHrP in the lactating mammary gland [6]. During lactation, the mammary gland becomes an accessory parathyroid gland. In nulliparous animals, the CaSR on parathyroid cells senses blood calcium concentrations and responds by secreting parathyroid hormone (**PTH**). PTH binds to its receptor (**PTH1R**) on the intestines, bones, and kidneys to modulate systemic calcium concentrations. During lactation, the mammary gland acts as an endocrine, parathyroid-like organ to coordinate calcium homeostasis through the secretion of PTHrP, which binds to the same PTH1R receptor on bone [111]. Feeding 5-HTP to rats during the transition from pregnancy to lactation increased PTHrP content in the mammary gland and the circulation, as well as milk and serum calcium concentrations [17]. In dairy cows, circulating serotonin concentrations were positively correlated with PTHrP concentrations and negatively correlated with the incidence of hypocalcemia [112]. Serum calcium and serotonin, as well as milk calcium and serotonin, were also positively correlated in early lactation dairy cows [113]. Intravenous infusion of 5-HTP to late-lactation, non-pregnant dairy cows decreased circulating calcium concentrations and urine calcium excretion, while increasing milk calcium concentrations [114]. Finally, infusion of 5-HTP to Wisconsin and Swiss dairy cows during the transition period modulated calcium homeostasis [115, 116], without alterations in PTH concentrations [117]. Serotonin has been repeatedly and definitively correlated with PTHrP and calcium homeostasis, particularly in early lactation. Yet the mechanisms underlying mammary gland coordination of the serotonin-PTHrP-calcium axis during lactation are not yet fully characterized. Defining the relationship between these key endocrine factors during lactation could be crucial to determining the risk, if any, to the health of breastfeeding mothers taking SSRIs.

Calcium and Bone Metabolism During Lactation

Calcium Requirements During Lactation

Breastfeeding women taking SSRIs may be at risk of compromised bone health because the mammary gland is a potent regulator of maternal calcium homeostasis. Calcium and phosphorous excreted in breast milk are crucial for infant skeletalization. Breast milk provides all of the calcium and phosphorous necessary to support a growing neonate. Coordinated signaling cascades between the breast, bone, intestine, and kidney maintain maternal calcium homeostasis during lactation [118]. Suckling-induced prolactin stimulates the absorption of calcium across the maternal intestine [119], and urine calcium excretion declines to very low levels in lactating mothers [120]. Additionally, lactation is associated with significant bone turnover and reductions in maternal BMD [118]. Normally, maternal calcium homeostasis is efficiently maintained during lactation. Dairy cows, however, are vulnerable to calcium dysregulation at the onset of lactation and are therefore a valuable model to understand mammary gland control of calcium.

During the transition from pregnancy to lactation, the dairy cow mammary gland puts significant demand on maternal calcium reserves due to copious milk production. Two to three days pre- and post-partum in dairy cows, circulating calcium concentrations decrease as calcium is shuttled to the mammary gland for milk synthesis [113, 121]. The resulting hypocalcemia can be classified as clinical (**CH**; <1.4 mM circulating calcium) or subclinical (**SCH**; <2.0 mM) [122]. There has

been recent debate about whether normocalcemia should be more stringently defined due to symptoms of SCH evident at calcium concentrations higher than the current threshold of 2.0 mM [123, 124]. Both clinical and subclinical hypocalcemia are associated with an increased risk of other early lactation disorders, such as displaced abomasum, ketosis, retained placenta, metritis, depressed immunity, and longer interval to pregnancy [125–130]. Although both SCH and CH are costly in terms of economic impact on the farmer and animal health, SCH is more insidious. While the physical signs of CH are obvious (hypothermia, tetany, recumbency, or even death), SCH often does not manifest in outward signs, so it is difficult to detect and treat. SCH is also more prevalent, affecting between 40 to 60% of multiparous cows, compared with CH which is estimated to affect only 7.5% of cows in U.S. dairy herds [131–133]. Currently-practiced prevention strategies such as feeding a negative dietary cation-anion difference (**DCAD**) diet have reduced the incidence of hypocalcemia [134–136]. Yet there are still a large number of dairy cows that suffer from a negative calcium balance at parturition which could detrimentally affect their health in current and subsequent lactations. DCAD diets work primarily through re-sensitizing PTH1R on various tissues to induce calcium mobilization [137, 138]. It is possible that manipulation of PTHrP through the serotonergic axis could affect the timing or magnitude of calcium mobilization to prevent hypocalcemia in dairy cows. Understanding calcium homeostasis during lactation in other species may provide a window into preventing calcium-related disorders, and their long-term consequences, in breastfeeding women.

The negative energy balance during pregnancy and early lactation in women and rodents is not as severe as that of dairy cows. It is estimated that lactating women lose, on average, 200 to 210 mg of calcium per day, from which the nursing infant is expected to accrete 100 mg per day [118]. To maintain total calcium concentrations between 2.1 to 2.7 mM, women experience increased intestinal absorption, renal conservation, and skeletal resorption of calcium [118]. There is a milieu of calcium-regulating hormones associated with lactation, including PTH, calcitriol, calcitonin, and PTHrP. PTH is typically suppressed during lactation in women who consume adequate calcium [139, 140]. Calcitriol (1,25-dihydroxyvitamin D_3 or **1,25(OH)$_2$D$_3$**) is the hormonally active metabolite of vitamin D and coordinates intestinal absorption of calcium and phosphate [141]. Lactating rodents have high serum calcitriol, indicating an increased rate of intestinal calcium absorption [142, 143]. By contrast, while intestinal absorption of calcium is doubled during pregnancy in women, it decreases to pre-pregnant rates during lactation [144, 145]. Additionally, urine calcium excretion decreases in both rodents and humans during lactation [118, 139, 145, 146]. Calcitonin, which is stimulated by high calcium concentrations and opposes the effects of PTH, is typically unchanged during lactation [145, 147] and is negatively regulated by calcitriol [148]. Given that neither PTH nor calcitriol make a significant contribution to the coordination of calcium accrual during lactation, research has focused on the role of PTHrP in regulating skeletal resorption to provide adequate supplies of calcium for the lactating mother. We hypothesize that increased serotonin-driven PTHrP signaling during lactation may cause irreversible damage to maternal bone.

Bone Turnover During Lactation

Bone turnover is rapid and significant during lactation and lactational bone loss is remarkably conserved across species. Lactating rodents typically resorb 25 to 35% of their bone mineral over a 21-day lactation [146, 149–151]. The African Green Monkey loses 20% BMD at the lumbar spine over a 20-week lactation [152] while whole-body bone mineral content (**BMC**) decreases by 3.3% in young Rhesus macaques in the first three months of lactation [153]. Foundational work showed that beagle lactation is characterized by cancellous bone remodeling [154]. Farm animals also undergo lactational bone loss: bone weight and bone strength decrease in sows during lactation and are restored during the subsequent gestation [155]. The onset of lactation is associated with decreased BMC and BMD in lactating sheep and goats [156]. Dairy cows have increased biochemical markers of bone resorption in the blood and urine around parturition, likely positively associated with milk yield [157, 158]. Finally, women are consistently reported to have an on average decline in BMD and BMC of three to ten percent after three to six months of exclusive breastfeeding [118]. Lactational declines in BMD correlate to the amount of milk produced, as women nursing twins and triplets lose more bone mass than women nursing only one child [159]. The duration of lactation may also affect the amount of bone resorbed, with greater losses the longer a woman breastfeeds [160–163].

Lactational bone loss is a normal and hormonally programmed process, not to be associated with a pathology in and of itself. A series of endocrine signals, including prolactin [164], fibroblast growth factor 21 [106], estrogen [144, 165], PTH [139, 140], and PTHrP [67] coordinate to regulate bone mass during lactation. In lactating rodents and women, the greatest reductions in BMD are at the lumbar spine, with more modest bone loss from the hip, femur, tibia, and distal radius where there is less trabecular bone and more cortical bone [118]. Studies suggest that bone loss during lactation cannot be attenuated by a woman's dietary calcium intake. Women consuming high concentrations of calcium [166] and women accustomed to a very low calcium intake [167] both experience bone resorption during lactation. Interestingly, recent work in mice demonstrated that preventing bone loss during lactation had no effect on maternal calcium metabolism (PTH, PTHrP, vitamin D), milk production, or milk calcium content

when dietary calcium levels were sufficient. However, when dietary calcium intake was insufficient and bone resorption was inhibited, 41% of dams died of hypocalcemia [168]. This work suggests that maternal bone loss is not absolutely required to meet milk calcium demands if dietary calcium is sufficient in rodents. However, it is difficult for pregnant and lactating women to obtain sufficient calcium through the diet to meet increased reproductive demands [118].

Bone turnover during lactation occurs through at least two mechanisms. The first is osteoclast-mediated bone resorption. Bone is continuously renewed in the adult skeleton through the process of bone remodeling. During lactation, both bone formation and resorption are increased (as indicated by increased osteoblast and osteoclast activity, respectively) [67, 146, 149, 154]. During rapid lactational bone turnover, resorption outpaces formation, so that net bone loss is achieved. Osteoclastogenesis is regulated by the receptor activator of nuclear factor-κB (**RANK**) / receptor activator of nuclear factor-κB ligand (**RANKL**) / osteoprotegerin (**OPG**) signaling axis. In response to PTHrP binding to PTH1R during lactation, RANKL is secreted by cells of the osteoblast lineage. When RANKL binds to RANK on osteoclasts, a signaling cascade results in translocation of nuclear factor-κB (**NF-κB**) to the nucleus, leading to downstream differentiation and activation of mature osteoclasts [165]. Osteoclasts are large multinucleated cells that adhere to the bone surface using actin-rich podosomes. The tightly-sealed podosomes create an extracellular bone resorbing compartment with a ruffled basolateral plasma membrane. The osteoclast then acidifies the extracellular compartment through a proton pump in the ruffled border and the resulting low pH dissolves the mineralized bone matrix and releases calcium and phosphorous contained in the hydroxyapatite crystals. Concurrently, degradation of bone collagen is carried out by specific matrix metalloproteases (**MMP**) and lysosomal cysteine proteases such as cathepsin K [169]. Osteoclast activity is counterbalanced through secretion of osteoblast-derived OPG, a decoy receptor that binds RANKL in the extracellular space to prevent its binding to RANK [170]. During lactation, there is a greater mRNA abundance of RANKL and greater RANKL/OPG ratio in the bone compared to nulliparous animals [149]. Following forced weaning, RANKL expression decreases, while OPG expression remains elevated, resulting in decreased bone resorption. There is also significant osteoclast apoptosis during involution [149, 171]. Interestingly, mice missing a distal enhancer of the RANKL gene had reduced RANKL mRNA expression in the bone compared to controls, yet lost bone at a similar rate during lactation [172]. These data suggest that there are redundant mechanisms that have evolved to ensure sufficient calcium can be mobilized from the skeleton during lactation to support maternal homeostasis.

Osteocytic osteolysis is an additional mechanism by which calcium is mobilized from bone during lactation. Within the endosteum, the membranous lining of the medullary cavity that contains bone marrow, a network of cells including osteoblasts, osteoclasts, and osteocytes coordinate bone remodeling. Osteoblasts synthesize and secrete the collagen matrix and inorganic salt crystals that combine to give bone its hardness. As the matrix surrounding the osteoblast calcifies, some osteoblasts embed in unmineralized, organic matrix called osteoid and begin to extend dendritic processes connecting the periosteal and endosteal lining as well as cells in the bone marrow. In this way, osteoblasts differentiate into osteocytes. Osteocytes are the most abundant cell type in bones and reside within a complex network of interconnected lacunae and canaliculi channels within the bone matrix. The lacunar-canalicular network represents an area from which calcium can be mobilized that is larger than the combination of endosteum, periosteum, and trabecular surfaces [173–175].

During murine lactation, the osteocyte lacunae and canaliculi are enlarged in both cortical and trabecular bone. Concurrently, osteocytes express genes typically associated with osteoclasts, such as tartrate-resistant acid phosphatase and cathepsin K. Fascinatingly, lactational osteocytic osteolysis is mediated by the PTH1R receptor, as lactating animals with a disruption for PTH1R do not undergo perilacunar remodeling [176]. Given its relatively recent discovery, osteocytic osteolysis is still being characterized. In cortical bone, MMP13 contributes to osteocyte perilacunar remodeling [177, 178] and lacunar-canalicular expansion causes a 13% reduction in the elastic modulus of bone during lactation [179]. This most recent finding has important implications for the characterization of bone stiffness during lactation. Previous reports attributed the lactational reduction in bone stiffness to endocortical bone loss or thinning [180], despite computational analyses suggesting that diaphyseal geometry alone could not explain lactation-induced reductions in bone stiffness [150]. Osteolytic osteolysis, controlled at least in part by PTHrP signaling, represents an important mechanism by which the osteocyte may have a dramatic overall impact in maintaining mineral homeostasis during lactation. As of now, there is no direct evidence that SSRIs impact osteoclastic resorption or osteocytic osteolysis during lactation. However, given the relationship between mammary gland serotonin, PTHrP, and bone mobilization, it may be time to consider the effect of SSRIs on maternal bone.

The Role of PTHrP in Lactational Bone Turnover

A key component of the lactational crosstalk between breast and bone is PTHrP. As serotonin induces mammary PTHrP synthesis, we hypothesize that elevated serotonin signaling associated with SSRIs could cause inappropriate induction of PTHrP during lactation. During mammary gland development, PTHrP is a crucial signaling molecule in mammary epithelial and mesenchymal cells. Loss of PTHrP signaling

arrests mammary gland development in the bud stage without ductal growth [181]. In a healthy non-pregnant, non-lactating woman, PTHrP is mainly an autocrine-paracrine signaling factor [182]. As such, PTHrP is normally undetectable in the adult circulation [183], except during humoral hypercalcemia of malignancy [184–186], hyperprolactinemia [187], and breast-to-bone metastasis [188]. PTHrP is also detectable in the circulation during fetal development [189], pregnancy [190], and lactation [191, 192]. In breast cancer, PTHrP has been associated with osteolysis at the site of bone metastases [193], a process that is potentially mediated by SHH signaling [68]. In fact, PTHrP inhibition may suppress breast cancer metastasis [194]. Even in non-cancerous models, elevated circulating concentrations of PTHrP are correlated with bone loss in humans and mice [67, 192].

PTHrP is strikingly similar to PTH in terms of molecular structure, causing them to bind to the same PTH1R receptor [195]. PTH binds more tightly to PTH1R than PTHrP [196] and PTH infusions cause more potent elevations in circulating calcium and $1,25(OH)_2$ vitamin D concentrations compared to PTHrP infusions [197]. Short pulses of both PTH and PTHrP (as opposed to sustained high levels) have been shown to stimulate bone formation [198]. Sustained PTH/PTHrP activity increases expression of RANKL and decreases expression of OPG in the osteoblast to stimulate osteoclast differentiation and subsequent bone resorption [199]. PTHrP signaling in bone may be mediated by upstream SHH signaling, as SHH promotes osteoblast and osteoclast differentiation and activity through modulation of PTHrP [200–202]. Given that serotonin stimulates both SHH and PTHrP, sustained serotonin signaling associated with SSRI use may put women who are breastfeeding while taking SSRI at risk of elevated PTHrP activity.

During lactation, the mammary gland is the main source of circulating PTHrP. Genetic disruption of PTHrP in mouse MECs reduces circulating PTHrP levels and subsequently preserves bone mass [67]. PTHrP is also found in extremely high concentrations in milk, although researchers are still unclear as to what role PTHrP may play in neonatal development [203, 204]. Both systemic hypocalcemia [205] and disruption of CaSR at the onset of lactation [206] increase mammary PTHrP production. During lactation, the mammary gland acts as a "calcium-sensing" organ that is sensitive to the drain of maternal calcium into milk. As calcium moves into milk, low circulating calcium reduces CaSR activation on the basolateral surface of the MEC. As a result, PTHrP is secreted into the circulation, where it binds to its receptor on osteoblasts to stimulate maternal skeletal mobilization. As calcium is liberated from bone, calcium binding to CaSR suppresses PTHrP secretion, attenuating bone resorption [111].

When finely-tuned mammary gland calcium sensing mechanisms are disrupted, severe pathology can result. Postpartum osteoporosis has been reported in several breastfeeding mothers [207–209]. The fractures associated with postpartum osteoporosis can be debilitating, with women presenting with as many as six to ten concurrent vertebral fractures. In several of these patients, PTHrP concentrations are significantly elevated, with one patient maintaining elevated PTHrP concentrations for months post-weaning [207]. A recent case report suggests that elevated PTHrP may promote osteoclast activity in women with postpartum osteoporosis [210]. These case reports support the hypothesis that elevated PTHrP signaling during lactation can cause persistent, potentially long-term damage to maternal bone.

Elevated plasma PTHrP has been shown to predict lactational BMD loss even after controlling for various other factors, including estrogen, PTH, and breastfeeding status [192]. However, PTHrP is most effective in promoting bone turnover when estrogen levels are low, as they are during lactation [146]. Treating lactating mice with estrogen and the bisphosphonate pamidronate decreased rates of bone resorption, but did not entirely prevent bone loss [67]. Postmenopausal women also experience bone loss as a result of reduced systemic estrogen, but not nearly to the extent of a lactating woman. In a startling comparison, while a rate of bone loss greater than 1% per year is considered rapid in postmenopausal women, lactating women experience declines in BMD of 1 to 3% per month [211]. Ovariectomy experiments in rodents also show that lactation-associated bone loss is greater than bone loss as a result of estrogen depletion [146]. In addition to decreased estrogen concentrations, PTHrP has also been correlated with elevated levels of serotonin, as discussed earlier in this review (see *Serotonin in lactation*). Serotonin has been shown to induce PTHrP expression in vascular smooth muscle cells [212], MECs, and breast cancer cell lines [6]. Treatment of the breast cancer cell line MDA-MB-231 with serotonin increased expression of runt-related transcription factor 2 (**RUNX2**). RUNX2 expression inhibited osteoblast differentiation and stimulated osteoclast differentiation by the PTHrP/RANKL pathway, ultimately promoting osteolytic bone lesions [213]. Across species and in vitro models, there is strong evidence for serotonergic regulation of bone metabolism via PTHrP signaling. Women with increased serotonin signaling as a result of SSRI exposure may be at risk of PTHrP-induced bone resorption, causing persistent damage to bone.

Is Bone Recovered Following Lactation?: Epidemiological Studies

In order to evaluate if women using SSRIs during lactation are at risk for reduced BMD later in life, it is necessary to separately examine the long-term effects of lactation and SSRIs on bone health. Immediately following the cessation of lactation in rodents, there is widespread osteoclast apoptosis, a decrease in RANK/RANKL expression, and an increase in osteoblast

number and activity, all of which represent anabolic bone formation [149, 171]. Urinary markers of bone resorption decrease and serum markers of bone formation increase following weaning [149, 214]. Mice with a conditional knockout of PTHrP in osteoblasts recover from lactation normally, even though PTHrP is required to maintain adult bone mass and strength [215]. Osteocytic osteolysis ends with the cessation of lactation, and osteocytes take on an "osteoblast-like" phenotype to aid in bone formation [176]. Rodents restore whole-body BMC within two to four weeks after weaning [149, 151, 214]. A significant limitation of using rodents to estimate lactation-associated bone loss, however, is their short lifespan. As such, it is necessary to turn to human epidemiological data to evaluate bone recovery following lactation. The duration of total lactation, site of the skeleton, and time of life in which the follow-up analysis is performed (pre- or post-menopausal) all affect the evaluation of bone recovery post-weaning.

Postmenopausal women have a greater incidence of osteoporosis due to declining estrogen levels. Osteoporosis is characterized by skeletal fragility and microarchitectural deterioration, leading to a consequent increase in fracture risk. An estimated 50% of women over the age of 50 experience an osteoporosis-related fracture of the femoral neck, based on data from the National Health and Examination Survey (**NHANES**) III (1988-1994) [216]. Due to the aging United States population, the cost of osteoporosis care is expected to rise to $25.3 billion by 2025 [217]. Greater BMD acquired during adolescence protects against postmenopausal osteoporosis [218], with peak BMD achieved during the average United Staets woman's key reproductive years around the age of 26 ± 4 years old [219]. As such, insults to bone during the years where a woman reaches peak bone mass could have a significant effect on her long-term bone health. If lactation irreversibly reduces BMD, women who breastfeed their children could have reduced bone strength compared to nulliparous women. Known risk factors for bone loss, such as certain medications or poor diet/exercise, may only elevate a breastfeeding woman's risk of osteoporosis and/or fracture later in life.

Evidence that Mothers who Breastfeed Fully Recover Bone Mass

Many studies suggest that, by 12 months after weaning, bone mass lost during lactation is fully recovered [118, 145, 160, 161]. Bone undergoes rapid remineralization post-weaning, such that women who breastfeed for six months or more and become subsequently pregnant within the following 18 months do not have reduced BMD at the spine or the hip at the end of the second pregnancy [215]. Kovacs summarized the findings of over five dozen papers that showed lactation had a neutral or protective effect on BMD and fracture risk [118]. Among these was a study of Finnish women evaluated 16 to 20 years following the birth of their last child. Women who breastfed for longer than 33 total months had greater hip and tibia bone strength index estimates compared to women who breastfed less than 12 months [220]. In another study, adolescent mothers were evaluated between the ages of 20 to 25 years old, which is significant because BMD was measured relatively soon after the cessation of lactation. Mothers who breastfed during adolescence had higher bone mass than mothers who had not breastfed, with as much as 7% higher BMD in the femoral neck [221]. In a study of twin pairs who had experienced a different number of pregnancies and duration of lactation, there was no effect of lactation on BMD. But in a cross-sectional analysis looking at the twins and their female relatives, parous women who had breastfed had higher BMD than parous women who had never breastfed [222]. Obviously, a study among twins intrinsically adjusts for age, genetic variation, and many environmental factors, so this population is particularly informative. In many population studies, there was no association between breastfeeding and bone mass, to include women in Germany [223], Norway [224], Sri Lanka [225], Turkey [226] and the United States [227–229]. Due to the large number of epidemiological studies with similar conclusions, the longstanding paradigm held by experts in the field is that lactation does not adversely affect long-term bone health [118, 230, 231].

Evidence that Mothers who Breastfeed do not Fully Recover Bone Mass

Despite conventional wisdom surrounding lactational bone loss and recovery, there is evidence to suggest that women, particularly postmenopausal women, do not fully restore skeletal integrity after weaning. Women who breastfeed for a long duration seem to be at particular risk of incomplete bone restoration following weaning. Several studies examined the Korean NHANES data from 2010 to 2011 for associations between breastfeeding and BMD. In postmenopausal women, lumbar spine BMD was negatively correlated with duration of breastfeeding independent of age, body mass index (**BMI**), smoking or alcohol consumption, physical activity, 25-hydroxyvitamin D, or daily intake of calcium / calories [232, 233]. The prevalence of osteoporosis was also higher in Korean women who breastfed longer than 18 months [233]. The more months a Korean woman spent breastfeeding across her entire lifetime, the greater her risk for low BMD [234]. Cumulative breastfeeding longer than 24 months in postmenopausal Mexican mestizo women was correlated with osteoporosis and osteopenia [235]. Similarly, postmenopausal women who breastfed more than one year per child had the highest risk for osteoporosis in a Turkish population [236].

Importantly, in these and other studies, the most robust effects of lactation duration on BMD were seen at the lumbar spine. Breastfeeding for longer than 18 months was associated

with a two-fold increase in the risk of vertebral fracture in postmenopausal Italian women [237]. In a study of postmenopausal Israeli women, each additional month of breastfeeding increased the risk of low BMD by 6.6%, most robustly at the lumbar spine [238]. Premenopausal African women from Zimbabwe and Uganda who had an extended duration of lactation greater than five years had lower lumbar spine BMD than women who had breastfed for a shorter duration [239]. Similarly, premenopausal women from seven areas in the developing world had lower distal radius and midshaft ulna BMD if they had breastfed for more than five years [240]. The frequency of osteoporosis at the lumbar spine was negatively affected by breastfeeding duration in postmenopausal Turkish women [241], and lactation duration was negatively correlated with BMD at the spine, forearm, and femur in postmenopausal Indian women [242]. Arguably the most recent findings in support of the negative implications of breastfeeding on bone used HR-pQCT to evaluate 58 Australian women who exclusively breastfed for five months compared with 48 control women. Exclusive lactation was associated with increased cortical porosity, reduced matrix mineralization, reduced trabecular number, and increased trabecular spacing at the distal tibia and distal radius. When these same women were followed up a median of 2.6 years later following an on average eight-month lactation, the structural changes in the tibia and radius had not improved from when women were scanned during lactation [243].

Given the milieu of supporting data, most of which is relatively recent, it may be pertinent to reevaluate the effects of lactation on the skeleton. Specifically, extended periods of lactation appear to have detrimental impacts on the postmenopausal lumbar spine. There is power in this data being collected in varying cohorts around the world, all with different nutritional standard intakes and environmental factors. As technology advances and clinicians are able to examine microarchitectural features of the skeleton more readily, as shown in Bjørnerem and colleagues' work [243], the impacts of lactation on the skeleton will assuredly become more clearly defined. The combination of lactation with pharmacological agents known to affect the skeleton, such as SSRIs, may predispose women to significant risk of low BMD, osteoporosis, and fracture.

Depression, Antidepressants, and Bone

Depression and Breastfeeding

To examine the relationship between lactation, SSRIs, and bone, it is necessary to understand peripartum depression and SSRI use. Data from NHANES 2005 to 2008 indicates that 22% of adults in the United States had depressive symptoms. Women were overrepresented in this study, with 26% of women and 18% of men reporting depressive symptoms [244]. It is estimated that 10 to 15% of women experience perinatal depression [245–247]. Postpartum depression (**PPD**) is characterized by at least one major depressive disorder occurring during pregnancy or in the six months following parturition [248]. Characteristic symptoms of PPD include depressed mood, inability to care for the infant due to sleep deprivation and loss of energy, and diminished concentration or indecisiveness [249]. Women who have depressive symptoms are more likely to stop breastfeeding earlier than non-depressed women [250–252]. In the United States, black women are the least likely to initiate and maintain breastfeeding compared to Hispanic and white mothers [253]. Other factors that might limit breastfeeding include work-related issues, embarrassment, concern about pain, low income and education, and insufficient breastfeeding support from hospital staff and family [254].

Breastfeeding confers significant benefits to both mother and infant [255] which include maternal protection against breast and ovarian carcinoma [256, 257] and infant protection against diarrhea, asthma, and obesity [258]. Interestingly, breastfeeding without formula supplementation for at least 6 months is associated with higher BMD of the breastfed child during adolescence [259, 260]. Both the American Academy of Pediatrics and the World Health Organization recommend 6 months of exclusive breastfeeding [261, 262]. The United States does not come close to meeting these recommended standards. Healthy People 2020 objectives seek to increase the proportion of infants who are breastfed exclusively through six months from only 14.1% in 2009 to 25.5% in 2020. An additional Healthy People 2020 objective is to "decrease the proportion of women delivering a live birth who experience postpartum depressive symptoms" [263]. Concurrent depression and breastfeeding can have confounding effects on bone. For example, one case study documented how a woman with PPD and lactational osteoporosis did not report her back pain to her physician because she had overriding concerns about suicidal ideation and feelings of hopelessness that outweighed her back pain [264]. While breastfeeding confers significant benefits to both mother and infant, there must be additional care taken in monitoring, prescribing medications, and assuring maternal and infant safety for breastfeeding mothers with PPD.

Use of SSRIs During Lactation

Serotonergic biology is intimately associated with postpartum depression, lactation, and bone health. Depressed patients have decreased tryptophan, serotonin, and 5HIAA concentrations in the serum and cerebrospinal fluid, as well as altered 5HTR expression and binding in the brain and reduced SERT density in platelets [265]. The most popular class of antidepressants, SSRIs, modulate depressive symptoms through

increased serotonin signaling in the brain. In the brain, serotonin is packaged in vesicles in the presynaptic neuron and released into the synapse following an action potential. In the synapse, serotonin interacts with various 5HTRs before reuptake by SERT back into the presynaptic neuron. SSRIs block SERT reuptake of serotonin, maintaining elevated serotonin concentrations in the synaptic cleft. Initially, 5HTR1a somatodendritic autoreceptors sense elevated serotonin concentrations and slow the rate of serotonergic neuron firing [266]. Long-term exposure to SSRIs causes downregulation of 5HTR1a and disinhibition of serotonin release at axon terminals, increasing the concentrations of serotonin available to bind to postsynaptic receptors. The initial downregulation of postsynaptic serotonin neurotransmission by 5HTR1a is often cited as the reason for the four- to eight-week delay before SSRIs become efficacious in alleviating anxiety- or depression-like symptoms [267].

A study of NHANES data in the United States from the 1999-2000 survey to the 2011-2012 survey demonstrated that the use of SSRIs has doubled from 4.3% to 8.5% in 13 years [268]. SSRIs were introduced in the late 1980s as an alternative to tricyclic antidepressants (**TCA**). In fact, between 1993 and 1995, the increase in prescription rates of TCA was 12.4%, while there was a 133.8% increase in SSRI prescriptions. SSRIs have a more favorable side effect profile than TCAs [269]. For this reason, and because of the low risk to the infant, SSRIs are the preferred antidepressants to use while breastfeeding [270, 271]. In a study of 228,876 singelton pregnancies in Tennessee between 1997 and 2005, antidepressant users in the peripartum period were 2.7 times more likely to fill a prescription for SSRIs than non-SSRI antidepressants [270]. Lactating mice are more responsive to SSRIs than nulliparous animals [107]. SSRIs administered to the mother are commonly found in breastmilk, but not necessarily in the infant's serum [271]. Platelet serotonin is depleted by SSRI administration. Platelets lack TPH1 and therefore cannot synthesize serotonin. As SSRIs inhibit platelet SERT activity, the platelets cannot take up serotonin. Therefore, platelet serotonin concentrations decrease following SSRI administration, despite increased SSRI activity the cellular and tissue level [272, 273]. Women who exclusively breastfeed have lower platelet serotonin compared to mixed-feeding mothers and more breastfeedings per day cause a proportionate decline in platelet serotonin [274, 275]. Given serotonin's role as a homeostatic regulator of lactation, SSRI treatment could cause perturbations in mammary gland and lactation physiology.

Despite their advantages over other antidepressants, SSRIs are not without their own negative effects during lactation. It can be difficult to isolate the effects of SSRI exposure on the mammary gland directly versus behavior that indicates the need for an SSRI, as depressed women also have a shorter duration of breastfeeding [276]. One study found that women taking SSRIs experienced delayed secretory activation at the onset of lactation compared to controls, although SSRI users were represented by only eight women out of a cohort of 431 women [277]. In a larger cohort, among 284 women using SSRIs before or at delivery, 80% initiated breastfeeding compared with 90% of the 183 control women. Additionally, 50% of SSRI users performed exclusive breastfeeding for two weeks compared to 65% of control women [278]. Intramammary SSRI administration to dairy cows accelerated mammary gland involution and repressed milk yield [16]. Given that high doses of serotonin negatively regulate both tight junctions and MEC homeostasis, SSRIs may delay the onset of lactation in particular.

Although within the same class, different SSRIs are prescribed for different mental health conditions and have variable side effect profiles. Among SSRI antidepressants, fluoxetine is a popular choice for both bench researchers and prescribers. In the United States, fluoxetine was the first SSRI available for clinical use, and as such there is a wide body of clinical and basic research using fluoxetine [279]. In breastfeeding mothers, fluoxetine has a relatively long half-life of two to three days, with an oral bioavailability close to 100% [70]. Fluoxetine concentrations peak in milk eight to nine hours after oral administration, so milk can be "pumped and dumped" to prevent exposure to the infant [280]. The developmental outcomes of *in utero* and lactational fluoxetine exposure have been reviewed elsewhere [281, 282]. Lactational use of SSRIs, and fluoxetine specifically, is common. As such, a large population of women may be vulnerable to understudied side effects of peripartum SSRI exposure.

SSRIs and Bone in the Peripartum Period

Fluoxetine exposure during pregnancy and lactation may affect long-term maternal bone health. Importantly, fluoxetine reduces BMD and bone strength in animal studies. Mice treated with a high dose of fluoxetine (20 mg/kg) have less bone formation and whole-body BMD than control mice, although there was no effect in mice dosed with 5 mg/kg fluoxetine [283]. The length of exposure to fluoxetine also appears to have an effect. Short-term (three weeks) fluoxetine exposure increased bone mass by acting directly on the osteoclasts to prevent bone resorption, while longer use (six weeks) was associated with net bone loss via a decrease in bone formation [284]. Another study showed that fluoxetine can inhibit both bone resorption and bone formation *in vitro* in human osteoclasts and osteoblasts depending on the dose [285]. During fracture healing, fluoxetine inhibits osteoblast proliferation and differentiation, as well as bone mineralization [286]. Fluoxetine administration resulted in low BMD in both C57BL/6J [283, 284, 287] and Swiss Webster [288] mice. Fluoxetine is not only useful in animal studies – it is also

commonly prescribed to pregnant and lactating women. Therefore, it is necessary to understand the potential effects of fluoxetine on maternal bone health.

It is difficult to isolate the negative effects of depression versus antidepressants on bone health. Are depressed people taking an SSRI more prone to low BMD or does the SSRI that a depressed person takes reduce BMD [289]? Depressed individuals have lower BMD than non-depressed patients, with the most pronounced effects at the spine, hip, and distal radius [290–292]. Both depression and osteoporosis are three-fold more common in women than in men, and women are more vulnerable to depression-associated low BMD [293]. Indeed, women over the age of 65 accounted for 74% of all fractures in the United States in 2005 [217]. Antidepressant use is also correlated with bone loss. A large cohort study of postmenopausal women in Finland showed that SSRI use was associated with bone loss in a dose-dependent manner even after the cofounder of depression was taken into account [294]. Bone loss is more severe in SSRI users: compared to TCAs, SSRIs were associated with increased fracture risk [295] and lower total hip BMD [296], especially after prolonged use [297]. Importantly, the association between SSRIs and low bone mass / increased fracture risk is evident across all ages and sexes, from adolescents [298], to young adults [299], to older men [300] and women [294, 301]. Infants exposed to SSRI *in utero* are also shorter and have a smaller head circumference [302]. While there are studies that find no association between BMD and antidepressants [303–306], overwhelming evidence suggests that SSRIs negatively affect bone mineral density and increase fracture risk. For pregnant and breastfeeding mothers, SSRI use may carry additional long-term consequences on bone.

Although animal studies often examine only short-term consequences of drug exposure, the negative effects on bone may last long beyond SSRI administration. After cessation of therapy, fluorinated SSRIs such as fluoxetine and fluvoxamine are detectable in the bone and bone marrow compartment at an order of magnitude higher than in plasma. In one study, while there was still fluoxetine present in the bone at 84 days post-withdrawal, brain sequestration of fluoxetine was only evident up to 40 days post-withdrawal [307]. Importantly, drugs and toxins sequestered in bone can be released in breast milk. In the 1940s and early 1950s, there was a significant release of radioactive material into the Russian Techa River. Exposed mothers had detectable levels of Strontium-90 (^{90}**Sr**) in their breast milk. In the years 1950 and 1951, the maternal diet accounted for much of the ^{90}Sr in breast milk. But in 1952 and 1953, when there was very little dietary intake of ^{90}Sr, the maternal skeleton instead contributed as much as 35% of the ^{90}Sr found in breast milk [308]. The implications of these studies cannot be overstated. Given the demonstrated negative effects of SSRIs on both maternal and infant bone, as well as the epigenetically-regulated lactational insult to bone, breastfeeding mothers taking antidepressants may represent a crucially vulnerable population in terms of long-term bone health.

This review has focused on the hypothesis that SSRIs cause persistent reductions in maternal BMD through elevated mammary gland serotonin-PTHrP activity during lactation (Fig. 1). There are aspects of this hypothesis that need further examination. For example, direct, mechanistic relationships must be established between SSRIs and serotonin activity in the mammary gland, as well as SSRIs and lactational hormones such as PTHrP and PTH. Additionally, time course studies must be undertaken to examine post-weaning bone recovery in detail, in animals with and without SSRI exposure. Serotonin signaling is active in bone cells [309–311] and serotonin has direct effects on bone turnover [283, 288, 312]. As such, SSRIs may have direct effects on bone during lactation, not mediated by the mammary gland. Recent work has shown that fluoxetine triggers a brain-serotonin-dependent rise in sympathetic output that impairs bone formation while increasing bone resorption, leading to net bone loss [284]. This represents just one example by which SSRIs could have an effect on bone during lactation independent of the mammary gland. There is very little work on the effect of SSRIs on the lactating mammary gland and MEC, including but not limited to intramammary calcium signaling. Finally, while SSRIs are considered safe for the infant, there remain questions as to the potential epigenetic effects of antidepressants on the neonate, both during pregnancy and lactation.

Conclusions

Excessive serotonin signaling associated with SSRIs during pregnancy and lactation may mediate mammary gland-derived cascades that compromise long-term maternal bone health. Non-neuronal serotonin is known to coordinate several aspects of calcium homeostasis during lactation, including calcium flux across the MEC, *Shh* promoter methylation, and PTHrP synthesis and secretion. Exposure to SSRI antidepressants during the peripartum period may exacerbate the mammary serotonin-PTHrP axis, resulting in persistent reductions in BMD and increasing the risk of osteoporosis and fracture. Yet there are still significant gaps in the serotonin / SSRI-calcium-PTHrP axis that warrant careful examination. While much research has focused on the effects of peripartum SSRIs on the gestational and breastfed neonate, the long-term maternal consequences require more thorough examination. While SSRIs are vitally important to many pregnant and breastfeeding mothers' mental health, there is evidence that SSRI exposure during pregnancy and lactation may have persistent effects on maternal bone mineral density.

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References

1. Oftedal OT. The mammary gland and its origin during synapsid evolution. J Mammary Gland Biol Neoplasia. 2002;7:225–52.
2. Capuco AV, Akers RM. The origin and evolution of lactation. J Biol. 2009;8:37.
3. Liu J, Leung P, Breastfeeding YA. Active Bonding Protects against Children's Internalizing Behavior Problems. Nutrients. 2014;6:76–89.
4. Feldman R. The neurobiology of mammalian parenting and the biosocial context of human caregiving. Horm Behav. 2016;77:3–17.
5. Goldman AS. Evolution of the mammary gland defense system and the ontogeny of the immune system. J Mammary Gland Biol Neoplasia. 2002;7:277–89.
6. Hernandez LL, Gregerson KA, Horseman ND. Mammary gland serotonin regulates parathyroid hormone-related protein and other bone-related signals. Am J Physiol Endocrinol Metab. 2012;302: E1009–15.
7. Tsapakis EM, Gamie Z, Tran GT, Adshead S, Lampard A, Mantalaris A, et al. The adverse skeletal effects of selective serotonin reuptake inhibitors. Eur Psychiatry. 2012;27:156–69.
8. Rapport MM, Green AA, Page IH. Crystalline serotonin. Science. 1948;108:329–30.
9. Turlejski K. Evolutionary ancient roles of serotonin: long-lasting regulation of activity and development. Acta Neurobiol Exp. 1996;56:619–36.
10. Côté F, Thévenot E, Fligny C, Fromes Y, Darmon M, Ripoche MA, et al. Disruption of the nonneuronal tph1 gene demonstrates the importance of peripheral serotonin in cardiac function. Proc Natl Acad U S A. 2003;100:13525–30.
11. Lesurtel M, Soll C, Humar B, Clavien PA. Serotonin: a double-edged sword for the liver? Surgeon. 2012;10:107–13.
12. El-Merahbi R, Löffler M, Mayer A, Sumara G. The roles of peripheral serotonin in metabolic homeostasis. FEBS Lett. 2015;589:1728–34.
13. Namkung J, Kim H, Park S. Peripheral serotonin: a new player in systemic energy homeostasis. Mol Cells. 2015;38:1023–8.
14. Watts SW. Oh the places you'll go! My many colored serotonin (apologies to Dr. Suess). Am J Physiol Heart Circ Physiol. 2016;311:H1225–33.
15. Matsuda M, Imaoka T, Vomachka AJ, Gudelsky GA, Hou Z, Mistry M, et al. Serotonin regulates mammary gland development via an autocrine-paracrine loop. Dev Cell. 2004;6:193–203.
16. Hernandez LL, Stiening CM, Wheelock JB, Baumgard LH, Parkhurst AM, Collier RJ. Evaluation of serotonin as a feedback inhibitor of lactation in the bovine. J Dairy Sci. 2008;91:1834–44.
17. Laporta J, Peters TL, Weaver SR, Merriman KE, Hernandez LL. Feeding 5-hydroxy-l-tryptophan during the transition from pregnancy to lactation increase calcium mobilization from bone in rats. Domest Anim Endocrinol. 2013;44:176–84.
18. Vela Hinojosa C, León Galván MA, Tapia Rodríguez M, López Ortega G, Cerbón Cervantes MA, Rodríguez CA, et al. Differential expression of serotonin tryptophan hydroylase and monoamine oxidase A in the mammary gland of the Myotis velifer bat. PLoS One. 2013;8:e75062.
19. Olendorf WH. Brain uptake of radiolabeled amino acids, amines, and hexoses after arterial injection. Am J Physiol. 1971;221:1629–39.
20. Wang Q, Liu D, Song P, Zou MH. Tryptophan-kynurenine pathway is dysregulated by inflammation, and immune activation. Front Biosci (Landmark Ed). 2015;20:1116–43.
21. van Praag HM, Lemus C. Monoamine precursors in the treatment of psychiatric disorders. In: Wurtman RJ, Wurtman JJ, editors. Nutrition and the brain. New York: Raven Press; 1986. p. 89–139.
22. Richard DM, Dawes MA, Mathias CW, Acheson A, Hill-Kapturczak N, Dougherty DM. L-tryptophan: Basic metabolic functions, behavioral research, and therapeutic indications. Int J Tryptophan Res. 2009;2:45–60.
23. Rahman MK, Nagatsu T, Sakurai T, Hori S, Abe M, Matsuda M. Effect of pyridoxal phosphate deficiency on aromatic L-amino acid decarboxylase activity with L-DOPA and L-5-hydroxytryptophan as substrates in rats. Jpn J Pharmacol. 1982;32:803–11.
24. Mann JJ, McBride PA, Brown RP, Linnoila M, Leon AC, DeMeo M, et al. Relationship between central and peripheral serotonin indexes in depressed and suicidal psychiatric inpatients. Arch Gen Psychiatry. 1992;49:442–6.
25. Hannon J, Hoyer D. Molecular biology of 5-HT receptors. Behav Brain Res. 2008;195:198–213.
26. Hernandez LL, Limesand SW, Collier JL, Horseman ND, Collier RJ. The bovine mammary gland expresses multiple functional isoforms of serotonin receptors. J Endocrinol. 2009;203:123–31.
27. Pai VP, Marshall AM, Hernandez LL, Buckley AR, Horseman ND. Altered serotonin physiology in human breast cancers favors paradoxical growth and cell survival. Breast Cancer Res. 2009;11: R81.
28. Stull MA, Pai V, Vomachka AJ, Marshall AM, Jacob GA, Horseman ND. Mammary gland homeostasis employs serotonergic regulation of epithelial tight junctions. Proc Natl Acad Sci U S A. 2007;104:16708–13.
29. Pai VP, Horseman ND. Biphasic regulation of mammary epithelial resistance by serotonin through activation of multiple pathways. J Biol Chem. 2008;283:30901–10.
30. Pai VP, Hernandez LL, Stull MA, Horseman ND. The type 7 serotonin receptor, 5-HT 7, is essential in the mammary gland for regulation of mammary epithelial structure and function. Biomed Res Int. 2015;2015:364736.
31. Laporta J, Keil KP, Vezina CM, Hernandez LL. Peripheral serotonin regulates maternal calcium trafficking in mammary epithelial cells during lactation in mice. PLoS One. 2014;9:e110190.
32. Reiter RJ. Pineal melatonin: cell biology of its synthesis and of its physiological interactions. Endocr Rev. 1991;12:151–80.
33. Yamaguchi Y, Hayashi C. Simple determination of high urinary excretion of 5-hydroxyindole-3-acetic acid with ferric chloride. Clin Chem. 1978;24:149–50.
34. Holmsen H, Weiss HJ. Secretable storage pools in platelets. Annu Rev Med. 1979;30:119–34.
35. McNicol A, Isreals SJ. Platelet dense granules: Structure, function and implications for haemostasis. Thrombosis Research. 1999;95: 1–18.
36. Berger M, Gray JA, Roth BL. The expanded biology of serotonin. Ann Rev Med. 2009;60:355–66.
37. Erspamer V. Occurrence of indolealkylamines in nature. In: Erspamer V, editor. Handbook of experimental pharmacology: 5-hydroxytryptamine and related indolealkylamines. New York: Springer Verlag; 1966. p. 132–81.
38. Biochemistry GMD, Physiology of serotonergic transmission. In: Brookhart JM, Mountcastle VB, editors. Handbook of

physiology: the nervous system. New Jersey: Wiley-Blackwell for the American Physiological Society; 1977. p. 573–623.
39. Weaver SR, Jury NJ, Gregerson KA, Horseman ND, Hernandez LL. Characterization of mammary-specific disruptions for Tph1 and Lrp5 during murine lactation. Sci Rep. 2017;7:15155.
40. Dupont C, Armant DR, Brenner CA. Epigenetics: Definition, mechanism, and clinical perspective. Semin Reprod Med. 2009;27:351–7.
41. Berger SL. The complex language of chromatin regulation during transcription. Nature. 2007;447:407–12.
42. Borrelli E, Nestler EJ, Allis CD, Sassone-Corsi P. Decoding the epigenetic language of neuronal plasticity. Neuron. 2008;60:961–74.
43. Antequera F. Structure, function and evolution of CpG island promoters. Cell Mol Life Sci. 2003;60:1647–58.
44. Métivier R, Gallais R, Tiffoche C, Le Péron C, Jurkowska RZ, Carmouche RP, et al. Cyclical DNA methylation of a transcriptionally active promoter. Nature. 2008;452:45–50.
45. Schwarz JM, Nugent BM, McCarthy MM. Developmental and hormone-induced epigenetic changes to estrogen and progesterone receptor genes in brain are dynamic across the life span. Endocrinology. 2010;151:4871–81.
46. Crider KS, Yang TP, Berry RJ, Bailey LB. Folate and DNA methylation: A review of molecular mechanisms and the evidence for folate's role. Adv Nutr. 2012;3:21–38.
47. Uysal F, Akkoyunlu G, Ozturk S. Dynamic expression of DNA methyltransferases (DNMTs) in oocytes and early embryos. Biochimie. 2015;116:103–13.
48. Kriaucionis S, Heintz N. The nuclear DNA base 5-hydroxymethylcytosine is present in Purkinje neurons and the brain. Science. 2009;324:929–30.
49. Tahiliani M, Koh KP, Shen Y, Pastor WA, Bandukwala H, Brudno Y, et al. Conversion of 5-methylcytosine to 5-hydroxymethylcytosine in mammalian DNA by MLL partner TET1. Science. 2009;324:930–5.
50. Ito S, Shen L, Dai Q, Wu SC, Collins LB, Swenberg JA, et al. Tet proteins can convert 5-methylcytosine to 5-formylcytosine and 5-carboxylcytosine. Science. 2011;333:1300–3.
51. Holloway T, González-Maeso J. Epigenetic mechanisms of serotonin signaling. ACS Chem Neurosci. 2015;6:1099–109.
52. Nikolova YS, Koenen KC, Galea S, Wang CM, Seney ML, Sibille E, et al. Beyond genotype: serotonin transporter epigenetic modification predicts human brain function. Nat Neurosci. 2014;17:1153–5.
53. Kinnally EL, Capitanio JP, Leibel R, Deng L, LeDuc C, Haghighi F, et al. Epigenetic regulation of serotonin transporter expression and behavior in infant rhesus macaques. Genes Brain Behav. 2010;9:575–82.
54. Rajasethupathy P, Antonov I, Sheridan R, Frey S, Sander C, Tuschl T, et al. A role for neuronal piRNAs in the epigenetic control of memory-related synaptic plasticity. Cell. 2012;149:693–707.
55. Blazevic S, Horvaticek M, Kesic M, Zill P, Hranilovic D, Ivanisevic M, et al. Epigentic adaptation of the placental serotonin transporter gene (SLC6A4) to gestational diabetes mellitus. PLoS One. 2017;12:e0179934.
56. Huang L, Frampton G, Rao A, Zhang KS, Chen W, Lai JM, et al. Monoamine oxidase A expression is suppressed in human cholangiocarcinoma via coordinated epigenetic and IL-6-driven events. Lab Invest. 2010;92:1451–60.
57. Walther DJ, Peter JU, Winter S, Holtje M, Paulmann N, Grohmann M, et al. Serotonylation of small GTPases is a signal transduction pathway that triggers platelet alpha-granule release. Cell. 2003;115:851–62.
58. Paulmann N, Grohmann M, Voigt JP, Bert B, Vowinckel J, Bader M, et al. Intracellular serotonin modulates insulin secretion from pancreatic beta-cells by protein serotonylation. PLoS Biol. 2009;7:e1000229.
59. Watts SW, Priestley JR, Thompson JM. Serotonylation of vascular proteins important to contraction. PLoS One. 2009;4:e5682.
60. Al-Zoairy R, Pedrini MT, Khan MI, Engl J, Tschoner A, Ebenbichler C, et al. Serotonin improves glucose metabolism by serotonylation of the small GTPase Rab4 in L6 skeletal muscles. Diabetol Metab Syndr. 2017;9:1.
61. Muma NA, Mi Z. Serotonylation and transamidation of other monoamines. ACS Chem Neurosci. 2015;6:961–9.
62. Lewis MT, Ross S, Strickland PA, Sugnet CW, Jimenez E, Scott MP, et al. Defects in mouse mammary gland development caused by conditional haploinsufficiency of Patched-1. Development. 1999;126:5181–93.
63. Lewis MT, Ross S, Strickland PA, Sugnet CW, Jimenez E, Hui C, et al. The Gli2 transcription factor is required for normal mouse mammary gland development. Dev Biol. 2001;238:133–44.
64. Sterling JA, Oyajobi BO, Grubbs B, Padalecki SS, Munoz SA, Gupta A, et al. The hedgehog signaling molecule Gli2 induces parathyroid hormone-related peptide expression and osteolysis in metastatic human breast cancer cells. Cancer Res. 2006;66:7548–53.
65. Nwabo Kamdje AH, Seke Etet PF, Vecchio L, Muller JM, Krampera M, Lukong KE. Signaling pathways in breast cancer: therapeutic targeting of the microenvironment. Cell Signal. 2014;26:2843–56.
66. Lipinski RJ, Gipp JJ, Zhang J, Doles JD, Bushman W. Unique and complimentary activities of the Gli transcription factors in Hedgehog signaling. Exp Cell Res. 2006;312:1925–38.
67. VanHouten JN, Wysolmerski JJ. Low estrogen and high parathyroid hormone-related peptide levels contribute to accelerated bone resorption and bone loss in lactating mice. Endocrinology. 2003;144:5521–9.
68. Laporta J, Keil KP, Weaver SR, Cronick CM, Prichard AP, Crenshaw TD, et al. Serotonin regulates calcium homeostasis in lactation by epigenetic activation of hedgehog signaling. Mol Endocrinol. 2014;28:1866–74.
69. Marcus SM, Flynn HA, Blow FC, Barry KL. Depressive symptoms among pregnant women screened in obstetrics screenings. J Womens Health (Larchmt). 2003;12:373–80.
70. Davanzo R, Copertino M, De Cunto A, Minen F, Amaddeo A. Antidepressant drugs and breastfeeding: a review of the literature. Breastfeed Med. 2011;6:89–98.
71. Tran H, Robb AS. SSRI use during pregnancy. Semin Perinatol. 2015;39:545–7.
72. Walther DJ, Peter JU, Bashammakh S, Hortnagl H, Voits M, Fink H, et al. Synthesis of serotonin by a second tryptophan hydroxylase isoform. Science. 2003;299:76.
73. Schraenen A, Lemaire K, de Faudeur G, Hendrickx N, Granvik M, Van Lommel L, et al. Placental lactogens induce serotonin biosynthesis in a subset of mouse beta cells. Diabetologia. 2010;53:2589–99.
74. Kim H, Toyofuku Y, Lynn FC, Chak E, Uchida T, Mizukami H, et al. Serotonin regulates pancreatic beta cell mass during pregnancy. Nat Med. 2010;16:804–8.
75. Iida H, Ogihara T, Min MK, Hara A, Kim YG, Fujimaki K, et al. Expression mechanism of tryptophan hydroxylase 1 in mouse islets during pregnancy. J Mol Endocrinol. 2015;55:41–53.
76. Ohara-Imaizumi M, Kim H, Yoshida M, Fujiwara T, Aoyagi K, Toyofuku Y, et al. Serotonin regulates glucose-stimulated insulin secretion from pancreatic beta cells during pregnancy. Am Proc Natl Acad Sci U S A. 2013;110:19420–5.
77. Park H, Oh CM, Park J, Park H, Cui S, Kim HS, et al. Deletion of the serotonin receptor type 3A in mice leads to sudden cardiac death during pregnancy. Circ J. 2015;79:1807–15.

78. Jeffrey JJ, Ehlich LS, Roswit WT. Serotonin – an inducer of collagenase in myometrial smooth-muscle cells. J Cell Physiol. 1991;146:399–406.
79. Ontsouka EC, Reist M, Graber H, Blum JW, Steiner A, Hirsbrunner G. Expression of messenger RNA coding for 5-HT receptor, alpha and beta adrenoreceptor (subtypes) during oestrus and dioestrus in the bovine uterus. J Vet Med A Physiol Pathol Clin Med. 2004;51:385–93.
80. Weiner CP, Thompson LP, Liu KZ, Herrig JE. Pregnancy reduces serotonin-induced contraction of guinea pig uterine and carotid arteries. Am J Physiol. 1992;263:H1764–9.
81. Middelkoop CM, Dekker GA, Kraayenbrink AA, Popp-Snijders C. Platelet-poor plasma serotonin in normal and preeclamptic pregnancy. Clin Chem. 1993;39:1675–8.
82. Bolte AC, van Geijn JP, Dekker GA. Pathophysiology of preeclampsia and the role of serotonin. Eur J Obstet Gynecol Reprod Biol. 2001;95:12–21.
83. Cengiz H, Dagdeviren H, Caypinar SS, Kanawati A, Yildiz S, Ekin M. Plasma serotonin levels are elevated in pregnant women with hyperemesis gravidarum. Arch Gynecol Obstet. 2015;291:1271–6.
84. Laporta J, Peters TL, Merriman KE, Vezina CM, Hernandez LL. Serotonin (5-HT) affects expression of liver metabolic enzymes and mammary gland glucose transporters during the transition from pregnancy to lactation. PLoS One. 2013;8:e57847.
85. Laporta J, Hernandez LL. Serotonin receptor expression is dynamic in the liver during the transition period in Holstein dairy cows. Domest Anim Endocrinol. 2015;51:65–73.
86. Laporta J, Gross JJ, Crenshaw TD, Bruckmaier RM, Hernandez LL. Short communication: Timing of first milking affects serotonin (5-HT) concentrations. J Dairy Sci. 2014;97:2944–8.
87. Côté F, Fligny C, Bayard E, Launay JM, Gershon MD, Mallet J, et al. Maternal serotonin is crucial for murine embryonic development. Proc Natl Acad Sci U S A. 2007;104:329–34.
88. Amireault P, Sibon D, Côté F. Life without peripheral serotonin: Insights from tryptophan hydroxylase 1 knockout mice reveal the existence of paracrine/autocrine serotonergic networks. ACS Chem Neurosci. 2013;4:64–71.
89. Yavarone MS, Shuey DL, Sadler TW, Lauder JM. Serotonin uptake in the ectoplacental cone and placenta of the mouse. Placenta. 1993;14:149–61.
90. Huang WQ, Zhang CL, Di XY, Zhang RQ. Studies on the localization of 5-hydroxytryptamine and its receptors in human placenta. Placenta. 1998;19:655–61.
91. Viau M, Lafond J, Vaillancourt C. Expression of placental serotonin transporter and 5-HT 2A receptor in normal and gestational diabetes mellitus pregnancies. Reprod Biomed Online. 2009;19:207–15.
92. Bonnin A, Peng W, Hewlett W, Levitt P. Expression mapping of 5-HT1 receptor subtypes during fetal and early postnatal mouse forebrain development. Neuroscience. 2006;141:781–94.
93. Anderson GM, Gutknecht L, Cohen DJ, Brailly-Tabard S, Cohen JH, Ferrari P, et al. Serotonin transporter promoter variants in autism: functional effects and relationship to platelet hyperserotonemia. Mol Psychiatry. 2002;7:831–6.
94. Gaspar P, Cases O, Maroteaux L. The developmental role of serotonin: news from mouse molecular genetics. Nat Rev Neurosci. 2003;4:1002–12.
95. Bonnin A, Goeden N, Chen K, Wilson ML, King J, Shih JC, et al. A transient placental source of serotonin for the fetal forebrain. Nature. 2011;472:347–50.
96. Toda T, Homma D, Tokuoka H, Hayakawa I, Sugimoto Y, Ichinose H, et al. Birth regulates the initiation of sensory map formation through serotonin signaling. Dev Cell. 2013;27:32–46.
97. Schneider ML, Moore CF, Barr CS, Larson JA, Kraemer GW. Moderate prenatal alcohol exposure and serotonin genotype interact to alter CNS serotonin function in rhesus monkey offspring. Alcohol Clin Exp Res. 2011;35:912–20.
98. Sullivan EL, Grayson B, Takahashi D, Robertson N, Maier A, Bethea CL, et al. J Neurosci. 2010;30:3826–30.
99. Weaver SR, Bohrer JC, Prichard AS, Perez PK, Streckenbach LJ, Olson JM, et al. Serotonin deficiency rescues lactation on day 1 in mice fed a high fat diet. PLoS One. 2016;11:e0162432.
100. Raymond JR, Mukhin YV, Gelasco A, Turner J, Collinsworth G, Gettys TW, et al. Multiplicity of mechanisms of serotonin receptor signal transduction. Pharmacol Ther. 2001;92:179–212.
101. Collier RJ, Hernandez LL, Horseman ND. Serotonin as a homeostatic regulator of lactation. Domest Anim Endocrinol. 2012;43:161–70.
102. Hernandez LL, Collier JL, Vomachka AJ, Collier RJ, Horseman ND. Suppression of lactation and acceleration of involution in the bovine mammary gland by a selective serotonin reuptake inhibitor. J Endocrinol. 2011;209:45–54.
103. Zhang CL, Chen H, Wang YH, Zhang RF, Lan XY, Lei CZ, et al. Serotonin receptor 1B (HTR1B) genotype associated with milk production traits in cattle. Res Vet Sci. 2008;85:265–8.
104. Qanbari S, Pimentel EC, Tetens J, Thaller G, Lichtner P, Sharifi AR, et al. A genome-wide scan for signatures of recent selection in Holstein cattle. Anim Genet. 2010;41:377–89.
105. Nguyen DA, Neville MC. Tight junction regulation in the mammary gland. J Mammary Gland Biol Neoplasia. 1998;3:233–46.
106. Bornstein S, Brown SA, Le PT, Wang X, DeMambro V, Horowitz MC, et al. FGF-21 and skeletal remodeling during and after lactation in C57BL/6J mice. Endocrinology. 2014;155:3516–26.
107. Jury NJ, McCormick BA, Horseman ND, Benoit SC, Gregerson KA. Enhanced responsiveness to selective serotonin reuptake inhibitors during lactation. PLoS One. 2015;10:e0117339.
108. Davis FM, Janoshazi A, Janardhan KS, Steinckwich N, D'Agostin DM, Petranka JG, Desai PN, Roberts-Thomson SJ, Bird GS, Tucker DK, Fenton SE, Feske S, Monteith GR, Putney JW Jr. Essential role of Orai1 store-operated calcium channels in lactation. Proc Natl Acad Sci U S A. 2015;112:5827-5832.
109. Reinhardt TA, Lippolis JD, Shull GE, Horst RL. Null mutation in the gene encoding plasma membrane Ca2+-ATPase isoform 2 impairs calcium transport into milk. J Biol Chem. 2004;279:42369–73.
110. VanHouten JN, Neville MC, Wysolmerski JJ. The calcium-sensing receptor regulates plasma membrane calcium adenosine triphosphatase isoform 2 activity in mammary epithelial cells: a mechanism for calcium-regulated calcium transport into milk. Endocrinology. 2007;148:5943–54.
111. VanHouten JN. Calcium sensing by the mammary gland. J Mammary Gland Biol Neoplasia. 2005;10:129–39.
112. Laporta J, Moore SA, Peters MW, Hernandez LL. Short communication: circulating serotonin (5-HT) concentrations on day 1 of lactation as a potential predictor of transition-related disorders. J Dairy Sci. 2013;96:5146–50.
113. Moore SA, Laporta J, Crenshaw TD, Hernandez LL. Patterns of circulating serotonin and related metabolites in multiparous dairy cows in the peripartum period. J Dairy Sci. 2015;98:3754–65.
114. Laporta J, Moore SA, Weaver SR, Cronick CM, Olsen M, Prichard AP, et al. Increasing serotonin concentrations alter calcium and energy metabolism in dairy cows. J Endocrinol. 2015;226:43–55.
115. Hernández-Castellano LE, Hernandez LL, Weaver SR, Bruckmaier RM. Increased serum serotonin improves parturient calcium homeostasis in dairy cows. J Dairy Sci. 2017;100:1580–7.
116. Weaver SR, Prichard AP, Endres EL, Newhouse SA, Peters TL, Crump PM, et al. Elevation of circulating seorotnin improves calcium dynamics in the peripartum dairy cow. J Endocrinol. 2016;230:105–23.

117. Hernández-Castellano LE, Hernandez LL, Sauerwein H, Bruckmaier RM. Endocrine and metabolic changes in transition dairy cows are affected by pre-partum infusions of a serotonin precursor. J Dairy Sci. 2017;100:5050–7.
118. Kovacs CS. Maternal mineral and bone metabolism during pregnancy, lactation, and post-weaning recovery. Physiol Rev. 2016;96:449–547.
119. Charoenphandhu N, Krishnamra N. Prolactin is an important regulator of intestinal calcium transport. Can J Physiol Pharmacol. 2007;85:569–81.
120. Klein CJ, Moser-Veillon PB, Douglass LW, Ruben KA, Trocki O. A longitudinal study of urinary calcium, magnesium, and zinc excretion in lactating and nonlactating postpartum women. Am J Clin Nutr. 1995;61:779–86.
121. Ramberg CF Jr, Mayer GP, Kronfeld DS, Phang JM, Berman M. Calcium kinetics in cows during late pregnancy, parturition, and early lactation. Am J Physiol. 1970;219:1166–77.
122. Goff JP. The monitoring, prevention, and treatment of milk fever and subclinical hypocalcemia in dairy cows. Vet J. 2008;176:50–7.
123. Chapinal N, Carson ME, LeBlanc SJ, Leslie KE, Godden S, Capel M, et al. The association of serum metabolites in the transition period with milk production and early-lactation reproductive performance. J Dairy Sci. 2012;95:1301–9.
124. Martinez N, Risco CA, Lima FS, Bisinotto RS, Greco LF, Ribeiro ES, et al. Evaluation of peripartal calcium status, energetic profile, and neutrophil function in dairy cows at low or high risk of developing uterine disease. J Dairy Sci. 2012;95:7158–72.
125. Massey CD, Wang C, Donovan GA, Beede DK. Hypocalcemia at parturition as a risk factor for left displacement of the abomasum in dairy cows. J Am Vet Med Assoc. 1993;203:852–3.
126. Hammon DS, Evjen IM, Dhiman TR, Goff JP, Walters JL. Neutrophil function and energy status in Holstein cows with uterine health disorders. Vet Immunol Immunopathol. 2006;113:21–9.
127. Kimura K, Reinhardt TA, Goff JP. Parturition and hypocalcemia blunts calcium signals in immune cells of dairy cattle. J Dairy Sci. 2006;89:2588–95.
128. Martinez N, Sinedino LD, Bisinotto RS, Ribeiro ES, Gomes GC, Lima FS, et al. Effect of induced subclinical hypocalcemia on physiological responses and neutrophil function in dairy cows. J Dairy Sci. 2014;97:874–87.
129. Sepúlveda-Varas P, Weary DM, Noro M, von Keyserlingk MA. Transition diseases in grazing dairy cows are related to serum cholesterol and other analytes. PLoS One. 2015;10:e0122317.
130. Rodriguez EM, Aris A, Bach A. Associations between subclinical hypocalcemia and postparturient diseases in dairy cows. J Dairy Sci. 2017;100:7427–34.
131. Reinhardt TL, Lippolis JD, McCluskey BJ, Goff JP, Horst RL. Prevalence of subclinical hypocalcemia in dairy herds. Vet J. 2011;188:122–4.
132. Martín-Tesoro J, Martens H. Calcium and magnesium physiology and nutrition in relation to the prevention of milk fever and tetany (dietary management of macrominerals in preventing disease). Vet Clin North Am Food Anim Pract. 2014;30:643–70.
133. Caixeta LS, Ospina PA, Capel MB, Nydam DV. The association of subclinical hypocalcemia, negative energy balance and disease with bodyweight change during the first 30 days post-partum in dairy cows milked with automatic milking systems. Vet J. 2015;204:150–6.
134. Charbonneau E, Pellerin D, Oetzel GR. Impact of lowering dietary cation-anion difference in nonlactating dairy cows: A meta-analysis. J Dairy Sci. 2006;89:537–48.
135. Weich W, Block E, Litherland NB. Extended negative dietary cation-anion difference feeding does not negatively affect postpartum performance of multiparous dairy cows. J Dairy Sci. 2013;96:5780–92.
136. Leno BM, Ryan CM, Stokol T, Kirk D, Zanzalari KP, Chapman JD, et al. Effects of prepartum dietary cation-anion difference on aspects of peripartum mineral and energy metabolism and performance of multiparous Holstein cows. J Dairy Sci. 2017;100:4604–22.
137. Goff JP, Horst RL. Role of acid-base physiology on the pathogenesis of parturient hypocalcaemia (milk fever) – the DCAD theory in principal and practice. Acta Vet Scand Suppl. 2003;97:51–6.
138. Goff JP, Liesegang A, Horst RL. 2014. Diet-induced pseudohypoparathyroidism: A hypocalcemia and milk fever risk factor. J Dairy Sci. 2014;97:1520–8.
139. Cross NA, Hillman LS, Allen SH, Krause GF, Vieira NE. Calcium homeostasis and bone metabolism during pregnancy, lactation, and postweaning: a longitudinal study. Am J Clin Nutr. 1995;61:514–23.
140. Affinito P, Tommaselli GA, di Carlo C, Guida F, Nappi C. Changes in bone mineral density and calcium metabolism in breastfeeding women: a one-year follow up study. J Clin Endocrinol Metab. 1996;81:2314–8.
141. Diaz de Barboza G, Guizzardi S, Tolosa de Talamoni N. Molecular aspects of intestinal calcium absorption. World J Gastroenterol. 2015;21:7142–54.
142. Halloran BP, DeLuca HF. Calcium transport in small intestine during pregnancy and lactation. Am J Physiol. 1980;239:E64–8.
143. Boass A, Toverud SU, Pike JW, Haussler MR. Calcium metabolism during lactation: enhanced intestinal calcium absorption in vitamin D-deprived, hypocalcemic rats. Endocrinology. 1981;109:900–7.
144. Kent GN, Price RI, Gutteridge DH, Rosman KJ, Smith M, Allen JR, et al. The efficiency of intestinal calcium absorption is increased in late pregnancy but not in established lactation. Calcif Tissue Int. 1991;48:293–5.
145. Ritchie LD, Fung EB, Halloran BP, Turnlund JR, Van Loan MD, Cann CE, et al. A longitudinal study of calcium homeostasis during human pregnancy and lactation and after resumption of menses. Am J Clin Nutr. 1998;67:693–701.
146. Ardeshirpour L, Brian S, Dann P, VanHouten J, Wysolmerski J. Increased PTHrP and decreased estrogens alter bone turnover but do not reproduce the full effects of lactation on the skeleton. Endocrinology. 2010;151:5591–601.
147. Krebs NF, Reidinger CJ, Robertson AD, Brenner M. Bone mineral density changes during lactation: maternal, dietary, and biochemical correlates. Am J Clin Nutr. 1997;65:1738–46.
148. Naveh-Many T, Raue F, Grauer A, Silver J. Regulation of calcitonin gene expression by hypocalcemia, hypercalcemia, and vitamin D in the rat. J Bone Miner Res. 1992;7:1233–7.
149. Ardeshirpour L, Dann P, Adams DJ, Nelson T, VanHouten J, Horowitz MC, et al. Weaning triggers a decrease in receptor activator of nuclear factor-kappaB ligand expression, widespread osteoclast apoptosis, and rapid recovery of bone mass after lactation in mice. Endocrinology. 2007;148:3875–86.
150. Liu XS, Ardeshirpour L, VanHouten JN, Shane E, Wysolmerski JJ. Site-specific changes in bone microarchitecture, mineralization, and stiffness during lactation and after weaning in mice. J Bone Miner Res. 2012;27:865–75.
151. Wendelboe MH, Thomsen JS, Henriksen K, Vegger JB, Brüel A. Zoledronate prevents lactation induced bone loss and results in additional post-lactation bone mass in mice. Bone. 2016;87:27–36.
152. Hiyaoka A, Yoshida T, Cho F, Yoshikawa Y. Changes in bone mineral density of lumbar vertebrae after parturition in African green monkeys (Cercopithecus aethiops). Exp Anim. 1996;45:257–9.

153. Ott SM, Lipkin EW, Newell-Morris L. Bone physiology during pregnancy and lactation in young macaques. J Bone Miner Res. 1999;14:1779–88.
154. Miller MA, Omura TH, Miller SC. Increased cancellous bone remodeling during lactation in beagles. Bone. 1989;10:279–85.
155. Giesemann MA, Lewis AJ, Miller PS, Akhter MP. Effects of the reproductive cycle and age on calcium and phosphorus metabolism and bone integrity of sows. J Anim Sci. 1998;76:796–807.
156. Liesegang A, Risteli J, Wanner M. The effects of first gestation and lactation on bone metabolism in dairy goats and milk sheep. Bone. 2006;38:794–802.
157. Liesegang A, Eicher R, Sassi ML, Risteli J, Kraenzlin M, Riond JL, et al. Biochemical markers of bone formation and resorption around parturition and during lactation in dairy cows with high and low standard milk yields. J Dairy Sci. 2000;83:1773–81.
158. Holtenius K, Ekelund A. Biochemical markers of bone turnover in the dairy cow during lactation and the dry period. Res Vet Sci. 2005;78:17–9.
159. Laskey MA, Prentice A, Hanratty LA, Jarjou LM, Dibba B, Beavan SR, et al. Bone changes after 3 mo of lactation: influence of calcium intake, breast-milk output, and vitamin D-receptor genotype. Am J Clin Nutr. 1998;67:685–92.
160. Sowers M, Corton G, Shapiro B, Jannausch ML, Crutchfield M, Smith ML, et al. Changes in bone density with lactation. JAMA. 1993;269:3130–5.
161. More C, Bettembuk P, Bhattoa HP, Balogh A. The effects of pregnancy and lactation on bone mineral density. Osteoporos Int. 2001;12:732–7.
162. Møller UK, Vio Streym S, Mosekilde L, Rejnmark L. Changes in bone mineral density and body composition during pregnancy and postpartum. A controlled cohort study. Osteoporos Int. 2012;23:1213–23.
163. Brembeck P, Lorentzon M, Ohlsson C, Winkvist A, Augustin H. Changes in cortical volumetric bone mineral density and thickness, and trabecular thickness in lactating women postpartum. J Clin Endocrinol Metab. 2015;100:535–43.
164. Suntornsaratoon P, Wongdee K, Goswami S, Krishnamra N, Charoenphandhu N. Bone modeling in bromocriptine-treated pregnant and lactating rats: possible osteoregulatory role of prolactin in lactation. Am J Physiol Endocrinol Metab. 2010;299:E426–36.
165. Boyce BF, Xing L. Biology of RANK, RANKL, and osteoprotegerin. Arthritis Res Ther. 2007;9:S1.
166. Cross NA, Hillman LS, Allen SH, Krause GF. Changes in bone mineral density and markers of bone remodeling during lactation and postweaning in women consuming high amounts of calcium. J Bone Miner Res. 1995;10:1312–20.
167. Prentice A, Jarjou LM, Stirling DM, Buffenstein R, Fairweather-Tait S. Biochemical markers of calcium and bone metabolism during 18 months of lactation in Gambian women accustomed to a low calcium intake and in those consuming a calcium supplement. J Clin Endocrinol Metab. 1998;83:1059–66.
168. Ardeshirpour L, Dumitru C, Dann P, Sterpka J, VanHouten J, Kim W, et al. OPG treatment prevents bone loss during lactation but does not affect milk production or maternal calcium metabolism. Endocrinology. 2015;156:2762–73.
169. Lorget F, Kamel S, Mentaverri R, Wattel A, Naassila M, Maamer M, et al. High extracellular calcium concentrations directly stimulate osteoclast apoptosis. Biochem Biophys Res Commun. 2000;268:899–903.
170. Boyce BF. Advances in the regulation of osteoclasts and osteoclast functions. J Dent Res. 2013;92:860–7.
171. Miller SC, Bowman BM. Rapid inactivation and apoptosis of osteoclasts in the maternal skeleton during the bone remodeling reversal at the end of lactation. Anat Rec (Hoboken). 2007;290:65–73.
172. Onal M, Galli C, Fu Q, Xiong J, Weinstein RS, Manolagas SC, et al. The RANKL distal control region is required for the increase in RANKL expression, but not the bone loss, associated with hyperparathyroidism or lactation in adult mice. Mol Endocrinol. 2013;26:341–8.
173. Belanger LF. Osteocytic osteolysis. Calcif Tissue Res. 1969;4:1–12.
174. Bonewald LF. The amazing osteocyte. J Bone Miner Res. 2011;26:229–38.
175. Wysolmerski JJ. Osteocytes remove and replace perilacunar mineral during reproductive cycles. Bone. 2013;54:230–6.
176. Qing H, Ardeshirpour L, Pajevic PD, Dusevich V, Jähn K, Kato S, et al. Demonstration of osteocytic perilacunar/canalicular remodeling in mice during lactation. J Bone Miner Res. 2012;27:1018–29.
177. Tang SY, Herber RP, Ho SP, Alliston T. Matrix metalloproteinase-13 is required for osteocytic perilacunar remodeling and maintains bone fracture resistance. J Bone Miner Res. 2012;27:1936–50.
178. Macica CM, King HE, Wang M, McEachon CL, Skinner CW, Tommasini SM. Novel anatomic adaptation of cortical bone to meet increased mineral demands of reproduction. Bone. 2016;85:59–69.
179. Kaya S, Basta-Pljakic J, Seref-Ferlengez Z, Majeska R, Cardoso L, Bromage T, et al. Lactation-induced changes in the volume of osteocyte lacunar-canalicular space alter mechanical properties in cortical bone tissue. J Bone Miner Res. 2017;32:688–97.
180. Vajda EG, Bowman BM, Cancellous MSC. cortical bone mechanical properties and tissue dynamics during pregnancy, lactation, and post-lactation in the rat. Biol Reprod. 2001;65:689–95.
181. Hens JR, Wysolmerski JJ. Key stages of mammary gland development: molecular mechanisms involved in the formation of the embryonic mammary gland. Breast Cancer Res. 2005;7:220–4.
182. Lee K, Deeds JD, Segre GV. Expression of parathyroid hormone-related peptide and its receptor messenger ribonucleic acids during fetal development of rats. Endocrinology. 1995;136:453–63.
183. Kovacs CS. The role of PTHrP in regulating mineral metabolism during pregnancy lactation, and fetal/neonatal development. Clinic Rev Bone Miner Metab. 2014;12:142–64.
184. Stewart AF, Insogna KL, Burtis WJ, Aminiafshar A. Wu T, Weir EC, Broadus AE. Frequency and partial characterization of adenylate cyclase-stimulating activity in tumors associated with humoral hypercalcemia of malignancy. J Bone Miner Res. 1986;1:267–76.
185. Burtis WJ, Wu T, Bunch C, Wysolmerski JJ, Insogna KL, Weir EC, et al. Identification of a novel 17,000-dalton parathyroid hormone-like adenylate cyclase-stimulating protein from a tumor associated with humoral hypercalcemia of malignancy. J Biol Chem. 1987;262:7151–6.
186. Burtis WJ, Brady TG, Orloff JJ, Ersbak JB, Warrell RP, Olson BR, et al. Immunochemical characterization of circulating parathyroid hormone-related protein in patients with humoral hypercalcemia of cancer. N Engl J Med. 1990;322:1106–12.
187. Stiegler C, Leb G, Kleinert R, Warnkross H, Ramschak-Schwarzer S, Lipp R, et al. Plasma levels of parathyroid hormone-related peptide are elevated in hyperprolactinemia and correlated to bone density status. J Bone Miner Res. 1995;10:751–9.
188. Washam CL, Byrum SD, Leitzel K, Ali SM, Tackett AJ, Gaddy D, et al. Identification of PTHrP(12-48) as a plasma biomarker associated with breast cancer bone metastasis. Cancer Epidemiol Biomarkers Prev. 2013;22:972–83.
189. Thiébaud D, Janisch S, Koelbl H, Hanzal E, Jacquet AF, Leodolter S, et al. Direct evidence of a parathyroid related protein gradient between the mother and the newborn in humans. Bone Miner. 1993;23:213–21.
190. Ardawi MS, Nasrat HA, BA'Aqueel HS. Calcium-regulating hormones and parathyroid hormone-related peptide in normal human

pregnancy and postpartum: a longitudinal study. Eur J Endocrinol. 1997;137:402–9.
191. Lippuner K, Zehnder HJ, Casez JP, Takkinen R, Jaeger P. PTH-related protein is released into the mother's bloodstream during lactation: evidence for beneficial effects on maternal calcium-phosphate metabolism. J Bone Miner Res. 1996;11:1394–9.
192. Sowers MF, Hollis BW, Shapiro B, Randolph J, Janney CA, Zhang D, et al. Elevated parathyroid hormone-related peptide associated with lactation and bone density loss. JAMA. 1996;276:549–54.
193. Powell GJ, Southby J, Danks JA, Stillwell RG, Hayman JA, Henderson MA, et al. Localization of parathyroid hormone-related protein in breast cancer metastases: increased incidence in bone compared with other sites. Cancer Res. 1991;51:3059–61.
194. Wang Y, Lei R, Zhuang X, Zhang N, Pan H, Li G, et al. DLC1-dependent parathyroid hormone-like hormone inhibition suppresses breast cancer bone metastasis. J Clin Invest. 2014;124:1646–59.
195. Wysolmerski JJ. Parathyroid hormone-related protein: an update. J Clin Endocrinol Metab. 2012;97:2947–56.
196. Pioszak AA, Parker NR, Gardella TJ, Xu HE. Structural basis for parathyroid hormone-related protein binding to the parathyroid hormone receptor and design of conformation-selective peptides. J Biol Chem. 2009;284:28382–91.
197. Horwitz MJ, Tedesco MB, Sereika SM, Syed MA, Garcia-Ocaña A, Bisello A, et al. Continuous PTH and PTHrP infusion causes suppression of bone formation and discordant effects on 1, 25(OH)2 vitamin D. J Bone Miner Res. 2005;20:1792–803.
198. Cosman F, Greenspan SL. Parathyroid hormone treatment for osteoporosis. In: Rosen CJ, editor. Primer on the metabolic bone diseases and disorders of mineral metabolism. Washington DC: American Society for Bone and Mineral Research; 2008. p. 244–9.
199. Karaplis AC, Goltzman. PTH and PTHrP effects on the skeleton. Rev Endocr Metab Disord. 2000;1:331–41.
200. Jemtland R, Divieti P, Lee K, Segre GV. Hedgehog promotes primary osteoblast differentiation and increases PTHrP mRNA expression and iPTHrP secretion. Bone. 2003;32:611–20.
201. Mak KK, Bi Y, Wan C, Chuang PT, Clemens T, Young M, et al. Hedgehog signaling in mature osteoblasts regulates bone formation and resorption by controlling PTHrP and RANKL expression. Dev Cell. 2008;14:674–88.
202. Kiuru M, Solomon J, Ghali B, van der Meulen M, Crystal RG, Hidaka C. Transient overexpression of sonic hedgehog alters the architecture and mechanical properties of trabecular bone. J Bone Miner Res. 2003;24:1598–607.
203. Budayr AA, Halloran BP, King JC, Diep D, Nissenson RA, Strewler GJ. High levels of parathyroid hormone-like protein in milk. Proc Natl Acad Sci U S A. 1989;86:7183–5.
204. Onda K, Sato A, Yamaguchi M, Matsuki N, Ono K, Wada Y. Parathyroid hormone-related protein (PTHrP) and Ca levels in the milk of lactating cows. J Vet Med Sci. 2006;68:709–13.
205. VanHouten JN, Dann P, McGeoch G, Brown EM, Krapcho K, Neville M, et al. The calcium-sensing receptor regulates mammary gland parathyroid hormone–related protein production and calcium transport. J Clin Invest. 2004;113:598–608.
206. Mamillapalli R, VanHouten J, Dann P, Bikle D, Chang W, Brown E, et al. Mammary-specific ablation of the calcium-sensing receptor during lactation alters maternal calcium metabolism, milk calcium transport, and neonatal calcium accrual. Endocrinology. 2013;154:3031–42.
207. Reid IR, Wattie DJ, Evans MC, Budayr AA. Postpregnancy osteoporosis associated with hypercalcemia. Clin Endocrinol (Oxf). 1992;37:298–303.
208. Segal E, Hochberg I, Weisman Y, Ish-Shalom S. Severe postpartum osteoporosis with increased PTHrP during lactation in a patient after total thyroidectomy and parathyroidectomy. Osteoporos Int. 2011;22:2907–11.
209. Ozturk C, Atamaz FC, Akkurt H, Akkoc Y. Pregnancy-associated osteoporosis presenting after severe vertebral fractures. J Obstet Gynaecol Res. 2014;40:288–92.
210. Grizzo FM, da Silva Martins J, Pinheiro MM, Jorgetti V, Carvalho MD, Pelloso SM. Pregnancy and lactation-associated osteoporosis: Bone histomorphometric analysis and response to treatment with zoledronic acid. Calcif Tissue Int. 2015;97:421–5.
211. Kovacs CS. The skeleton is a storehouse of mineral that is plundered during lactation and (fully?) replenished afterwards. J Bone Miner Res. 2017;32:676–80.
212. Pirola CJ, Wang HM, Kaymar A, Wu S, Enomoto H, Sharifi B, et al. Angiotensin II regulates parathyroid hormone-related protein expression in cultured rat aortic smooth muscle cells through transcriptional and post-transcriptional mechanisms. J Biol Chem. 1993;268:1987–94.
213. Zong JC, Wang X, Zhou X, Wang C, Chen L, Yin LJ, et al. Gut-derived serotonin induced by depression promotes breast cancer bone metastasis through the RUNX2/PTHrP/RANKL pathway in mice. Oncol Rep. 2016;35:739–48.
214. Kirby BJ, Ardeshirpour L, Woodrow JP, Wysolmerski JJ, Sims NA, Karaplis AC, et al. Skeletal recovery after weaning does not require PTHrP. J Bone Miner Res. 2011;26:1242–51.
215. Laskey MA, Prentice A. Effect of pregnancy on recovery of lactational bone loss. Lancet. 1997;349:1518–9.
216. America's Bone Health. The State of Osteoporosis and Low Bone Mass in Our Nation. Washington DC: National Osteoporosis Foundation; 2002.
217. Burge R, Dawson-Hughes B, Solomon DH, Wong JB, King A, Tosteson A. Incidence and economic burden of osteoporosis-related fractures in the United States, 2005-2025. J Bone Miner Res. 2007;22:465–75.
218. Weaver CM, Peacock M, Johnston CC. Adolescent nutrition in the prevention of postmenopausal osteoporosis. J Clin Endocrinol Metab. 1999;84:1839–43.
219. Teegarden D, Proulx WR, Martin BR, Zhao J, McCabe GP, Lyle RM, et al. Peak bone mass in young women. J Bone Miner Res. 1995;10:711–5.
220. Wiklund PK, Xu L, Wang Q, Mikkola T, Lyytikäinen A, Völgyi E, et al. Lactation is associated with greater maternal bone size and bone strength later in life. Osteoporos Int. 2012;23:1939–45.
221. Chantry CJ, Auinger P, Byrd RS. Lactation among adolescent mothers and subsequent bone mineral density. Arch Pediatr Adolesc Med. 2004;158:650–6.
222. Paton LM, Alexander JL, Nowson CA, Margerison C, Frame MG, Kaymakci B, et al. Pregnancy and lactation have no long-term deleterious effect on measures of bone mineral in healthy women: a twin study. Am J Clin Nutr. 2003;77:707–14.
223. Hadji P, Ziller V, Kalder M, Gottschalk M, Hellmeyer L, Hars O, et al. Influence of pregnancy and breast-feeding on quantitative ultrasonometry of bone in postmenopausal women. Climacteric. 2002;5:277–85.
224. Forsmo S, Schei B, Langhammer A, Forsén L. How Do Reproductive and Lifestyle Factors Influence Bone Density in Distal and Ultradistal Radius of Early Postmenopausal Women? The Nord-Trøndelag Health Survey, Norway. Osteoporos Int. 2001;12:222–9.
225. Lenora J, Lekamwasam S, Karlsson MK. Effects of multiparity and prolonged breast-feeding on maternal bone mineral density: a community-based cross-sectional study. BMC Womens Health. 2009;9:19.
226. Yazici S, Korkmaz U, Erkan M, Korkmaz N, Erdem Baki A, Alçelik A, et al. The effect of breast-feeding duration on bone mineral density in postmenopausal Turkish women: a population-based study. Arch Med Sci. 2011;7:486–92.

227. Bauer DC, Browner WS, Cauley JA, Orwoll ES, Scott JC, Black DM, et al. Factors associated with appendicular bone mass in older women. The Study of Osteoporotic Fractures Research Group. Ann Intern Med. 1993;118:657–65.
228. Schnatz PF, Barker KG, Marakovits KA, O'Sullivan DM. Effects of age at first pregnancy and breast-feeding on the development of postmenopausal osteoporosis. Menopause. 2010;17:1161–6.
229. Crandall CJ, Liu J, Cauley J, Newcomb PA, Manson JE, Vitolins MZ, et al. Associations of parity, breastfeeding, and fractures in the Women's Health Observational Study. Obstet Gynecol. 2017;130:171–80.
230. Pregnancy SM. lactation as risk factors for subsequent bone loss and osteoporosis. J Bone Miner Res. 1996;11:1052–60.
231. Wysolmerski JJ. Interactions between breast, bone, and brain regulate mineral and skeletal metabolism during lactation. Ann N Y Acad Sci. 2010;1192:161–9.
232. Yeo UH, Choi CJ, Choi WS, Kim KS. Relationship between breast-feeding and bone mineral density among Korean women in the 2010 Korea National Health and Nutrition Examination Survey. J Bone Miner Metab. 2016;34:109–17.
233. Hwang IR, Choi YK, Lee WK, Kim JG, Lee IK, Kim SW, et al. Association between prolonged breastfeeding and bone mineral density and osteoporosis in postmenopausal women: KNHANES 2010-2011. Osteoporos Int. 2016;27:257–65.
234. Kim JH, Kwon H, Oh SW, Lee CM, Joh HK, Kim Y, et al. Breast Feeding is associated with postmenopausal bone loss: findings from the Korea National Health and Nutrition Examination Survey. Korean J Fam Med. 2015;36:216–20.
235. Rojano-Mejía D, Aguilar-Madrid G, López-Medina G, Cortes-Espinosa L, Hernández-Chiu MC, Canto-Cetina T, et al. Risk factors and impact on bone mineral density in postmenopausal Mexican mestizo women. Menopause. 2011;18:302–6.
236. Okyay DO, Okyay E, Dogan E, Kurtulmus S, Acet F, Taner CE. Prolonged breast-feeding is an independent risk factor for postmenopausal osteoporosis. Maturitas. 2013;74:270–5.
237. Bolzetta F, Veronese N, De Rui M, Berton L, Carraro S, Pizzato S, et al. Duration of breastfeeding as a risk factor for vertebral fractures. Bone. 2014;68:41–5.
238. Tsvetov G, Levy S, Benbassat C, Shraga-Slutzky I, Hirsch D. Influence of number of deliveries and total breast-feeding time on bone mineral density in premenopausal and young postmenopausal women. Maturitas. 2014;77:249–54.
239. Mgodi NM, Kelly C, Gati B, Greenspan S, Dai JY, Bragg V, et al. Factors associated with bone mineral density in healthy African women. Arch Osteoporos. 2014;10:206.
240. Petitti DB, Piaggio G, Mehta S, Cravioto MC, Meirik O. Steroid hormone contraception and bone mineral density: a cross-sectional study in an international population. Obstet Gynecol. 2000;95:736–44.
241. Dursun N, Akin S, Dursun E, Sade I, Korkusuz F. Influence of duration of total breast-feeding on bone mineral density in a Turkish population: does the priority of risk factors differ society to society? Osteoporos Int. 2006;17:651–5.
242. Singh R, Gupta S, Awasthi A. Differential effect of predictors of bone mineral density and hip geometry in postmenopausal women: a cross-sectional study. Arch Osteoporos. 2015;10:39.
243. Bjørnerem Å, Ghasem-Zadeh A, Wang X, Bui M, Walker SP, Zebaze R, et al. Irreversible deterioration of cortical and trabecular microstructure associated with breastfeeding. J Bone Miner Res. 2017;32:681–7.
244. Shim RS, Baltrus P, Ye J, Rust G. Prevalence, Treatment, and Control of Depressive Symptoms in the United States: Results from the National Health and Nutrition Examination Survey (NHANES), 2005–2008. J Am Board Fam Med. 2011;24:33–8.
245. Gaynes BN, Gavin N, Meltzer-Brody S, Lohr KN, Swinson T, Gartlehner G, et al. Perinatal depression: prevalence, screening accuracy, and screening outcomes. Evid Rep Technol Assess (Summ). 2005;119:1–8.
246. Dietz PM, Williams SB, Callaghan WM, Bachman DJ, Whitlock EP, Hornbrook MC. Clinically identified maternal depression before, during, and after pregnancies ending in live births. Am J Psychiatry. 2007;164:1515–20.
247. Ko JY, Rockhill KM, Tong VT, Morrow B, Farr SL. Trends in postpartum depressive symptoms – 27 states, 2004, 2008, and 2012. MMWR Morb Mortal Weekly Rep. 2017;66:153–8.
248. Meltzer-Brody S. New insights into perinatal depression: pathogenesis and treatment during pregnancy and postpartum. Dialogues Clin Neurosci. 2011;13:89–100.
249. Guille C, Newman R, Fryml LD, Lifton CK, Epperson CN. Management of postpartum depression. J Midwifery Womens Health. 2013;58:643–53.
250. Henderson JJ, Evans SF, Straton JA, Priest SR, Hagan R. Impact of postnatal depression on breastfeeding duration. Birth. 2003;30:175–80.
251. Pippins JR, Brawarsky P, Jackson RA, Fuentes-Afflick E, Haas JS. Association of breastfeeding with maternal depressive symptoms. J Womens Health (Larchmt). 2006;15:754–62.
252. Woolhouse H, James J, Gartland D, McDonald E, Brown SJ. Maternal depressive symptoms at three months postpartum and breastfeeding rates at six months postpartum: Implications for primary care in a prospective cohort study of primiparous women in Australia. Women Birth. 2016;29:381–7.
253. McKinney CO, Hahn-Holbrook J, Chase-Lansdale PL, Ramey SL, Krohn J, Reed-Vance M, et al. Racial and ethnic differences in breastfeeding. Pediatrics. 2016;138:e20152388.
254. Ogbuanu CA, Probst J, Laditka SB, Liu J, Baek JD, Glover S. Reasons why women do not initiate breastfeeding: A southeastern state study. Womens Health Issues. 2010;19:268–78.
255. Stuebe A. The risks of not breastfeeding for mothers and infants. Rev Obstet Gynecol. 2009;2:222–31.
256. Shema L, Ore L, Ben-Shachar M, Haj M, Linn S. The association between breastfeeding and breast cancer occurrence among Israeli Jewish women: a case control study. J Cancer Res Clin Oncol. 2007;133:539–46.
257. Chowdhury R, Sinha B, Jeeva Sankar M, Taneja S, Bhandari N, Rollins N, et al. Breastfeeding and maternal health outcomes: a systematic review and meta-analysis. Acta Pædiatrica. 2015;104:96–113.
258. Salone LR, Vann WF Jr, Dee DL. Breastfeeding: an overview of oral and general health benefits. J Am Dent Assoc. 2013;144:143–51.
259. Jones G, Riley M, Dwyer T. Breastfeeding in early life and bone mass in prepubertal children: A longitudinal study. Osteoporos Int. 2000;11:146–52.
260. Blanco E, Burrows R, Reyes M, Lozoff B, Gahagan S, Albala C. Breastfeeding as the sole source of milk for 6 months and adolescent bone mineral density. Osteoporos Int. 2017;28:2823–30.
261. World Health Organization. Exclusive breastfeeding for six months best for babies everywhere. 2011. http://www.who.int/mediacentre/news/statements/2011/breastfeeding_20110115/en/. Accessed 6 Dec 2017.
262. American Academy of Pediatrics. Breastfeeding and the use of human milk, section on breastfeeding. Pediatrics. 2012;e827:129.
263. Office of Disease Prevention and Health Promotion. In: Healthy People 2020, Maternal, Infant, and Child Health. 2010. Retrieved from https://www.healthypeople.gov/2020/topics-objectives/topic/maternal-infant-and-child-health/objectives. Accessed 6 Dec 2017.
264. Ozcelik B, Ozcelik A, Debre M. Postpartum depression co-occurring with lactation-related osteoporosis. Psychosomatics. 2009;50(2)

265. Ressler KJ, Nemeroff CB. Role of serotonergic and noradrenergic systems in the pathophysiology of depression and anxiety disorders. Depress Anxiety. 2000;12:2–19.
266. Fabre V, Beaufour C, Evrarad A, Rioux A, Hanoun N, Lesch KP, et al. Altered expression and functions of serotonin 5-HT1A and 5-HT1B receptors in knock-out mice lacking the 5-HT transporter. Eur J Neurosci. 2000;12:2299–310.
267. Sghendo L, Mifsud J. Understanding the molecular pharmacology of the serotonergic system: using fluoxetine as a model. J Pharm Pharmacol. 2012;64:317–25.
268. Kantor ED, Rehm CD, Haas JS, Chan AT, Giovannucci EL. Trends in prescription drug use among adults in the United States from 1999-2012. JAMA. 2015;314:1818–31.
269. Anderson IM. Selective serotonin reuptake inhibitors versus tricyclic antidepressants: a meta-analysis of efficacy and tolerability. J Affect Disord. 2000;58:19–36.
270. Hayes RM, Wu P, Shelton RC, Cooper WO, Dupont WD, Mitchel E, et al. Maternal antidepressant use and adverse outcomes: a cohort study of 228.876 pregnancies. Am J Obstet Gynecol. 2012;207:49.
271. Weissman AM, Levy BT, Hartz AJ, Bentler S, Donohue M, Ellingrod VL, et al. Pooled analysis of antidepressant levels in lactating mothers, breast milk, and nursing infants. Am J Psychiatry. 2004;161:1066–78.
272. Alvarez JC, Gluck N, Fallet A, Grégoire A, Chevalier JF, Advenier C, et al. Plasma serotonin level after 1 day of fluoxetine treatment: a biological predictor for antidepressant response? Psychopharmacology (Berl). 1998;143:97–101.
273. Epperson CN, Jatlow PI, Czarkowski K, Anderson GM. Maternal fluoxetine treatment in the postpartum period: Effects on platelet serotonin and plasma drug levels in breastfeeding mother-infant pairs. Pediatrics. 2003;112:e425-e429.
274. Epperson CN, Czarkowski KA, Ward-O'Brien D, Weiss E, Gueorguieva R, Jatlow P, et al. Maternal sertraline treatment and serotonin transport in breast-feeding mother-infant pairs. Am J Psychiatry. 2001;158:1631–7.
275. Jury NJ. Interaction of lactation and response to SSRI. In: Alterations in peripheral and central serotonin physiologies during lactation: Relevance to mood during the postpartum (Ph.D. Thesis). Cincinnati: University of Cincinnati Neuroscience / Medical Science Scholars Interdisciplinary; 2012. p. 116.
276. Dias CC, Figueiredo B. Breastfeeding and depression: a systematic review of the literature. J Affect Disord. 2015;171:142–54.
277. Marshall AM, Nommsen-Rivers LA, Hernandez LL, Dewey KG, Chantry CJ, Gregerson KA, et al. Serotonin transport and metabolism in the mammary gland modulates secretory activation and involution. J Clin Endocrinol Metab. 2010;95:837–46.
278. Gorman JR, Kao K, Chambers CD. Breastfeeding among women exposed to antidepressants during pregnancy. J Hum Lact. 2012;28:181–8.
279. Hillhouse TM, Porter JHA. brief history of the development of antidepressant drugs: From monoamines to glutamate. Exp Clin Psychopharmacol. 2015;23:1–21.
280. Lattimore KA, Donn SM, Kaciroti N, Kemper AR, Neal CR Jr, Vazquez DM. Selective serotonin reuptake inhibitor (SSRI) use during pregnancy and effects on the fetus and newborn: a meta-analysis. J Perinatol. 2005;25:595–604.
281. Kiryanova V, McAllister BB, Dyck RH. Long-term outcomes of developmental exposure to fluoxetine: A review of the animal literature. Dev Neurosci. 2013;35:437–49.
282. Glover ME, Clinton SM. Of rodents and humans: A comparative review of the neurobehavioral effects of early life SSRI exposure in preclinical and clinical research. Int J Dev Neurosci. 2016;51:50–72.
283. Warden SJ, Robling AG, Sanders MS, Bliziotes MM, Turner CH. Inhibition of the serotonin (5-hydroxytryptamine) transporter reduces bone accrual during growth. Endocrinology. 2005;146:685–93.
284. Ortuño MJ, Robinson ST, Subramanyam P, Paone R, Huang Y, Guo XE, et al. Serotonin reuptake inhibitors act centrally to cause bone loss in mice by counteracting a local antiresorptive effect. Nat Med. 2016;22:1170–9.
285. Hodge JM, Wang Y, Berk M, Collier FM, Fernandes TJ, Constable MJ, et al. Selective serotonin reuptake inhibitors inhibit human osteoclast and osteoblast formation and function. Biol Psychiatry. 2013;74:32–9.
286. Bradaschia-Correa V, Josephson AM, Mehta D, Mizrahi M, Neibart SS, Liu C, et al. The Selective Serotonin Reuptake Inhibitor Fluoxetine directly inhibits osteoblast differentiation and mineralization during fracture healing in mice. J Bone Miner Res. 2017;32:821–33.
287. Bonnet N, Bernard P, Beapied H, Bizot JC, Trovero F, Courteix D, et al. Various effects of antidepressant drugs on bone microarchitecture, mechanical properties and bone remodeling. Toxicol Appl Pharmacol. 2007;221:111–8.
288. Warden SJ, Nelson IR, Fuchs RK, Bliziotes MM, Turner CH. Serotonin (5-hydroxytryptamine) transporter inhibition causes bone loss in adult mice independently of estrogen deficiency. Menopause. 2008;15:1176–83.
289. Schwan S, Hallberg P. SSRIs, bone mineral density, and risk of fractures - a review. Eur Neuropsychopharmacol. 2009;19:683–92.
290. Michelson D, Stratakis C, Hill L, Reynolds J, Galliven E, Chrousos G, et al. Bone mineral density in women with depression. N Engl J Med. 1996;335:1176–81.
291. Yirmiya R, Bab I. Major depression is a risk factor for low bone mineral density: a meta-analysis. Biol Psychiatry. 2009;66:423–32.
292. Atteritano M, Lasco A, Mazzaferro S, Macrì I, Catalano A, Santangelo A, et al. Bone mineral density, quantitative ultrasound parameters and bone metabolism in postmenopausal women with depression. Intern Emerg Med. 2013;8:485–91.
293. Bab I, Yirmiya R. Depression, selective serotonin reuptake inhibitors, and osteoporosis. Curr Osteoporos Rep. 2010;8:185–91.
294. Rauma PH, Honkanen RJ, Williams LJ, Tuppurainen MT, Kröger HP, Koivumaa-Honkanen H. Effects of antidepressants on postmenopausal bone loss - A 5-year longitudinal study from the OSTPRE cohort. Bone. 2016;89:25–31.
295. Rabenda V, Nicolet D, Beaudart C, Bruyére O, Reginster JY. Relationship between use of antidepressants and risk of fractures: a meta-analysis. Osteoporos Int. 2013;24:121–37.
296. Diem SJ, Blackwell TL, Stone KL, Yaffe K, Haney EM, Bliziotes MM, et al. Use of antidepressants and rates of hip bone loss in older women: the study of osteoporotic fractures. Arch Intern Med. 2007;167:1240–5.
297. Ziere G, Dieleman JP, van der Cammen TJ, Hofman A, Pols HA, Stricker BH. Selective serotonin reuptake inhibiting antidepressants are associated with an increased risk of nonvertebral fractures. J Clin Psychopharmacol. 2008;28:411–7.
298. Feuer AJ, Demmer RT, Thai A, Vogiatzi MG. Use of selective serotonin reuptake inhibitors and bone mass in adolescents: An NHANES study. Bone. 2015;78:28–33.
299. Seifert CF, Wiltrout TR. Calcaneal bone mineral density in young adults prescribed selective serotonin reuptake inhibitors. Clin Ther. 2013;35:1412–7.
300. Cauley JA, Fullman RL, Stone KS, Zmuda JM, Bauer DC, Barrett-Connor E, et al. Factors associated with the lumbar spine and proximal femur bone mineral density in older men. Osteoporos Int. 2005;16:1525–37.
301. Ak E, Bulut SD, Bulut S, Akdag HA, Öter GB, Kaya H, et al. Evaluation of the effect of selective serotonin reuptake inhibitors on bone mineral density: an observational cross-sectional study. Osteoporos Int. 2015;26:273–9.

302. Dubnov-Raz G, Hemilä H, Vurembrand Y, Kuint J, Maayan-Metzger A. Maternal use of selective serotonin reuptake inhibitors during pregnancy and neonatal bone density. Early Hum Dev. 2012;88:191–4.
303. Diem SJ, Joffe H, Larson JC, Tsai JN, Guthrie KA, LaCroix AZ, et al. Effects of escitalopram on markers of bone turnover: A randomized clinical trial. J Clin Endocrinol Metab. 2014;99: E1732–7.
304. Calarge CA, Mills JA, Janz KF, Burns TL, Schlechte JA, Coryell WH, et al. The effect of depression, generalized anxiety, and selective serotonin reuptake inhibitors on change in bone metabolism in adolescents and emerging adults. J Bone Miner Res. 2017;32:2367–74.
305. Ham AC, Aarts N, Noordam R, Rivadendeira F, Ziere G, Zillikens MC, et al. Use of selective serotonin reuptake inhibitors and bone mineral density change: a population-based longitudinal study in middle-aged and elderly individuals. J Clin Psychopharmacol. 2017;37:524–30.
306. Saraykar S, John V, Cao B, Hnatow M, Ambrose CG, Rianon N. Association of selective serotonin reuptake inhibitors and bone mineral density in elderly women. J Clin Densitom. 2017; https://doi.org/10.1016/j.jocd.2017.05.016.
307. Bolo NR, Hodé Y, Macher JP. Long-term sequestration of fluorinated compounds in tissues after fluvoxamine or fluoxetine treatmetn: a fluorine magnetic resonance spectroscopy study in vivo. MAGMA. 2004;16:268–76.
308. Tolstykh EI, Shagina NB, Peremyslova LM, Degteva MO, Phipps AW, Harrison JD, et al. Reconstruction of 90Sr intake for breast-fed infants in the Techa riverside settlements. Radiat Environ Biophys. 2008;47:349–57.
309. Bliziotes MM, Eshleman AJ, Zhang XW, Wiren KM. Neurotransmitter action in osteoblasts: expression of a functional system for serotonin receptor activation and reuptake. Bone. 2001;29:477–86.
310. Gustafsson BI, Thommesen L, Stunes AK, Tommeras K, Westbroek I, Waldum HL, et al. Serotonin and fluoxetine modulate bone cell function in vitro. J Cell Biochem. 2006;98:139–51.
311. Dai SQ, Yu LP, Shi X, Wu H, Shao P, Yin GY, et al. Serotonin regulates osteoblast proliferation and function in vitro. Braz J Med Biol Res. 2014;47:759–65.
312. Kode A, Mosialou I, Silva BC, Rached MT, Zhou B, Wang J, et al. FOXO1 orchestrates the bone-suppressing function of gut-derived serotonin. J Clin Invest. 2012;122:3490–503.