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Optimizing bone health in anorexia nervosa and hypothalamic amenorrhea: new trials and tribulations

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Anorexia nervosa (AN) is an eating disorder characterized by refusal to maintain a healthy body weight and a fear of food, leading to a state of chronic energy deprivation. Anorexia nervosa is more common in young women, where the prevalence is about 0.3% to 1%, although it may be as high as 3.7% if less stringent diagnostic criteria are used [1,2]. A range of neuroendocrine abnormalities is observed in patients with AN, including amenorrhea, one of the diagnostic criteria of AN [3]. Functional hypothalamic amenorrhea (HA) is the cessation of menstrual periods due to the absence of hypothalamic signals to the pituitary gland resulting in absent stimulation of the ovaries and anovulation. This occurs not only in the context of AN but also in every case of chronically low energy availability, either from excessive exercise or weight loss, as well as psychological stress [4,5]. Therefore, HA is more common, with prevalence rates of 2% to 5% [6]; and it accounts for more than 30% of amenorrhea in reproductive-age women [7]. Most women with HA, such as competitive athletes, who do not have AN have less reduction in food intake, less severe weight loss, and less concomitant psychiatric problems compared with those with AN [8].

Amenorrhea, present in the majority of women with AN and in all women with HA [9], is associated with infertility that usually resolves with resolution of the amenorrhea, although rates of obstetrical complications may remain higher [10]. In addition to HA, there is also a range of comorbidities in women with AN that include cardiovascular complications

[11,12]; electrolyte abnormalities including hypokalemia, hypomagnesemia, and hypophosphatemia [11]; gastrointestinal complications including gastroparesis and constipation; elevated liver enzymes; skin changes including dry skin and development of lanugo hair; respiratory muscle weakness; cytopenias; and brain atrophy, in addition to endocrine dysfunction [11,13,14].

The endocrine abnormalities of AN are closely linked to the adverse effects on bone health. Both AN and HA in general are associated with dysfunction of the hypothalamic-pituitary-gonadal axis. The amenorrhea observed in these conditions is due to a reduction in amplitude and/or frequency of gonadotropin-releasing hormone pulse secretion [15], leading to loss or dysregulation of normal luteinizing hormone (LH) pulsatility [16] along with dysregulation of other hypothalamic-pituitary-peripheral axes. The lack of the stimulus from the gonadotropic hormones leads to reduced ovarian production of estradiol [17,18], with well-known adverse effects on bone health in women [19]. There is also a reduction in circulating androgen levels [17] in AN. Testosterone, which in women predominantly originates from the adrenal gland (25%), the ovary (25%), and the peripheral conversion of androstenedione (50%) [20], is markedly lower in women with AN [17]. Dehydroepiandrosterone sulfate (DHEAS) is the major androgen in a normal-cycling woman and is converted to other active androgens such as androstenedione, androstenediol, testosterone, and 5-dihydrotestosterone and to estrogens including estradiol and estrone [21]. Whether or not DHEAS levels are affected in women with AN is currently unclear. One large cross-sectional study found that DHEAS levels were not lower in women with AN compared with healthy, normal-weight controls [17], whereas another study reported decreased DHEAS levels in AN [22]. However, various studies have conclusively demonstrated that levels of DHEAS are reduced in women taking the oral contraceptive pill, including those with AN [17,23–25]. Androgens have putative effects on bone too [26,27], and androgen levels have been shown to be associated with both cortical and trabecular bone mineral density (BMD) [28,29]. The hypoandrogenemia in women with AN, whether secondary to the hypothalamic-pituitarygonadal dysfunction or to a suppressive effect of estrogen treatment in women with AN, may therefore result in further deleterious effects on bone health above that imposed by hypoestrogenemia.

Other endocrine axes are also affected in AN. Growth hormone levels increase, but insulin-like growth factor–1 (IGF-1) levels do not increase consequently and are frequently low [30–32], suggesting a state of growth hormone resistance [31]. Insulin-like growth factor–1 is a nutritionally dependent bone trophic factor [33,34] with critical endocrine and paracrine actions to stimulate osteoblast function and collagen formation [35,36], and low IGF-1 levels have been found to be strong predictors of abnormal bone microarchitectural parameters [37]. Anorexia nervosa is also complicated by hypothalamic-pituitary-adrenal dysregulation, resulting in hypercortisolemia [18,38]. Cortisol has well-known deleterious effects on bone health [39], and hypercortisolemia is associated with the severity of bone loss in AN [40].

Besides the multitude of endocrine dysfunctions, nutritional deficiency in general is known to impair peak bone accretion in adolescence [41], with subsequent risk of osteoporosis in later life. Chronic severe undernutrition is a hallmark of AN, and the resulting medical

complications are directly attributable to the caloric restriction and weight loss seen in this condition.

The combination of the poor nutritional status, low estrogen levels, and the other endocrine abnormalities discussed above leads to adverse effects on the bone health of these women. Bone density is markedly lower in women with AN at both trabecular and cortical bone sites [11,42], although trabecular bone, especially in the lumbar spine, tends to be more severely affected [43]. The annual rate of decline in BMD at the spine and hip is approximately 2.5% in adults with AN [44]. Loss of bone density occurs early in the disease course [45] and can be quite severe, with osteoporosis seen in 35% of patients with AN [11,42] due to both decreased bone formation and increased bone resorption, leading to an overall reduction in the BMD [46]. In addition, assessment of bone microarchitecture has shown marked abnormalities in bone volume and trabecular thickness [47]. Importantly, this insult to bone accumulation occurs at an age when bone mass should be markedly increasing to achieve peak bone mass, therefore leading to a reduced BMD later in life [48]. This leads to a fracture rate reported to be as high as 30% in AN [11,49] and an incidence of fractures of up to 61% in ballet dancers with HA [50]. Most of these fractures are atraumatic, stress fractures or fractures resulting from aminor injury that would not be expected normally to result in a fracture [11].

Hence, a major focus of treating women with AN is to prevent bone loss and lower fracture risk. Multiple treatment options have been investigated in AN, including weight gain, calcium and vitamin D supplementation, estrogen supplementation, androgen supplementation, IGF-1 replacement, and traditional osteoporosis treatments.

A study examining the effect of a behavioral weight-gain protocol involving nutritional and psychiatric intervention on bone turnover and BMD reported significant percentage increases in BMD with 2.2 months of treatment, although N-telopeptide, a bone resorption marker, remained elevated. A fall of N-telopeptide and a shift from the dominant resorptive state, which the authors postulate to imply full recovery, occurred only in the subgroup of subjects who regained menses with weight recovery. This suggests that normalization of bone metabolism is likely biphasic, involving a primary nutritional mechanism that stimulates bone formation and a hormonal mechanism that decreases bone resorption [51]. Weight gain through nutritional rehabilitation may therefore improve some or most of the neuroendocrine dysfunction [52], with associated improvement but perhaps not normalization of bone density [53].

Ensuring adequate intake of calcium (1200–1500 mg/d of elemental calcium) and vitamin D (600–1000 U/d) is also recommended and is considered standard treatment for patients with AN [54], and may have other beneficial effects [55]. However, studies have consistently failed to show any improvement in bone health with calcium and vitamin D supplementation alone in AN [56,57]. In a separate study, there was no correlation between calcium intake and BMD in adolescents with AN [58]. Therefore, although calcium and vitamin D are important determinants of developing bone density, they do not appear by themselves to be major contributing factors to restoration of bone mineral content in AN [43].

Because of the crucial role of estrogen in bone physiology, several trials have attempted to treat women with AN with estrogen, usually in the form of the combined oral contraceptive pill. Results have been largely unsatisfactory. Klibanski et al [56] studied the use of combined estrogen and progesterone vs calcium and vitamin D in adult women with AN. They found no increase in BMD in either group after 1 year, although subgroup analyses showed a significant increase in BMD in women with the lowest weight. Similarly, Strokosch et al [59] studied oral estrogen and progesterone vs placebo in adolescents with AN. After 1 year, they found no significant increase in BMD after controlling for weight gain. Other studies have found similar results [60,61]. However, a trial using 18 months of a more physiologic low-dose estrogen replacement regimen mimicking pubertal estrogen levels resulted in improvement of BMD *z*-score in the spine and hip when compared with placebo recipients (change in *z*-score of −0.026 and −0.236 in estrogen and placebo group, respectively, at the lumbar spine; corresponding numbers at the hip were −0.177 and −0.331, respectively) [62], without any evidence of estrogen-induced decrease in levels of IGF-1.

Exogenous estrogen suppresses IGF-1 secretion [58], which is a potential explanation as to why low-dose estrogen may be more beneficial than higher-dose estrogen. However, trials have also examined the effect of recombinant human IGF-1 on bone health, whether given alone or in combination with oral contraceptive pills. These have shown some promising results, with an increase in bone formation markers when recombinant IGF-1 was given alone [46]. However, a significant improvement in BMD was only shown when recombinant IGF-1 was given in combination with combined oral contraceptive pills for 9 months, when compared with recombinant IGF-1 alone, oral contraceptive pills alone, or no therapy [63].

As androgens may play a role in bone health, it has been suggested that androgen therapy could be a potential treatment of osteoporosis in AN. Dehydroepiandrosterone sulfate has been studied in the elderly, with mixed results. Postmenopausal women showed stabilization of BMD during treatment with DHEAS [64], but elderly men did not have any beneficial effect [65]. Although it is unclear whether women with AN have subnormal DHEAS levels, prior trials have looked at the effect of either DHEAS or testosterone supplementation in women with AN. Aprior 3-month–long trial of DHEAS found an increase in bone formation markers [22], but a follow-up study comparing DHEAS to oral estrogen-progesterone found no increase in BMD after 1 year with either treatment after controlling for weight gain [66]. One study found that testosterone treatment also increased bone formation markers in women with AN, but BMD data were not obtained [67]. In another study, treatment with testosterone did not lead to a significant increase in BMD whether with or without the addition of risedronate [68]. Therefore, although androgens may increase bone formation markers, they do not appear to improve BMD when used as monotherapy.

Other traditional medications such as antiresorptives used for osteoporosis have also been evaluated in AN, with varying results. For example, treatment with alendronate led to no increase in BMD after controlling for body weight [69], whereas treatment with risedronate for 9 months showed a 3% to 5% increase of BMD at the spine [68,70]. However, none of these medications are approved by the Food and Drug Administration for use in women with AN.

The modest results of the above studies and the lack of approved medications for prevention or treatment of osteoporosis in women with AN highlight an unmet need. They also suggest that treatment approaches aiming at improving bone health in AN subjects will likely require correcting the multitude of pathophysiological factors simultaneously rather than addressing isolated nutritional or hormonal deficiencies. Moreover, specific drawbacks of these treatments do exist. As illustrated above, exogenous estrogen monotherapy are generally unable to restore bone mass to the level of age-matched healthy controls [71], in contrast to unequivocal benefits seen in postmenopausal osteoporosis [72]. The lack of long-term safety data and clear-cut indications of DHEAS, as well as the lack of standardization of commercially available compounds, has led to recommendation against their use in women in general [74]. Although recombinant IGF-1 treatment improved bone formation markers when given in isolation, improvement in BMD was only seen in combination with estrogen replacement [63], suggesting likely insubstantial effect of recombinant IGF-1 on bone health when used as a monotherapy [46]. Concerns about long-term effects of suppressing bone turnover in young women with the use of bisphosphonates and the potential for teratogenic effects in pregnancy have limited the utility of these medications in AN subjects who are usually in the reproductive-age group [73]. Most importantly, most of these treatments entail either treating in isolation or only targeting specific deficiencies without addressing the wide spectrum of neuroendocrine, reproductive, and bone abnormalities evident in HA and AN, leading to inconsistent and unclear outcomes in terms of achieving peak bone mass or reducing fracture risk in the long term [74].

An article in this issue of *Metabolism* [75] evaluated the effect of combined DHEA with estrogen/progestin in preserving skeletal health over a duration of 18 months in subjects with AN. This study reported the preservation of spinal and whole-body BMD *z*-scores in the intervention group receiving DHEA and estrogen/progestin treatment, in contrast to the downward trend in the placebo group, resulting in a placebosubtracted improvement in bone density z-scores in the interventional group of 0.021 standard deviations (SD) in the spine, 0.017 SD in the hip, and 0.024 SD in the total body (placebo-corrected change in bone mineral density from baseline was 0.27%, 0.22% and 0.16%, respectively) [75]. This illustrates how treatment using both the anabolic bone-forming effect of DHEA and the antiresorptive effect of estrogen might yield better outcomes on BMD compared with previously reported treatment regimen targeting only an isolated defect in bone metabolism [66]. The therapy was well tolerated, with few androgen or estrogen-associated adverse effects. This presents a reasonably convenient, safe, and effective complementary treatment option to subjects with AN to improve bone health, while other interventions, including psychiatric and nutritional interventions, could still be applied concomitantly to further improve and provide a more permanent solution to the underlying medical problem. A longer duration of intervention and follow-up in future studies would be helpful to determine the optimal duration of therapy in maximizing bone health in this group of subjects with AN and to elucidate the long-term effects and/or adverse effects of this combination treatment. Future studies should also evaluate the efficacy on bone health of similar treatment regimens in the milder group of women with HA. Dehydroepiandrosterone sulfate is currently available as a dietary supplement in many countries, and the purity and potency of commercially available preparations are either inconsistent or not known [76]. Importantly,

purity and potency were standardized in this study [75], which may not be the case if patients obtain the medication outside of a research setting. Future studies using different doses and preparations of DHEAS in the treatment of AN will be helpful to clarify if the current inconsistency in commercial available preparations will affect treatment outcome. Clinical trials addressing potential adverse effects also need to be performed [77,78].

These above treatment regimens have largely been studied in subjects with AN. The utility of these treatments in subjects with HA is less clear. Both AN and HA are characterized by a common pathophysiological state of chronic energy deprivation, a condition marked by hypoleptinemia [79–81]. Circulating leptin levels reflect the amount of energy stores, and hypoleptinemia may well mediate many of the neuroendocrine disturbances associated with AN and HA [5,82]. Recent studies have highlighted the therapeutic potential of leptin as a treatment option in women with hypoleptinemic HA [83,84]. In a prospective open-label study, women with HA administered metreleptin for 3 months had increased mean LH levels and LH pulse frequency after 2 weeks, leading to an increase in maximal follicular diameter, the number of dominant follicles, ovarian volume, and estradiol levels [85], providing proof of concept that hypoleptinemia may be responsible for the reproductive and neuroendocrine abnormalities associated with HA. A randomized, double-blinded, placebo-controlled trial of metreleptin in replacement doses over a longer duration of 36 weeks in women with HA increased estradiol, progesterone, free triiodothyronine levels, IGF-1 to insulin-like growth factor–binding protein 3 ratio, and osteocalcin level while decreasing cortisol level, further validating the key role of hypoleptinemia in the multiple neuroendocrine, reproductive, and bone abnormalities associated with HA [86]. Another study examining the effect of longer-term metreleptin replacement in subjects with HA for up to 2 years found significantly increased bone mineral content by 5.1% and BMD by 8.6% at the lumbar spine, demonstrating the novel therapeutic potential of leptin replacement in the optimization of bone health in hypoleptinemic amenorrheic women [87]. These data herald new promises in the treatment of HA, as leptin results in an improvement of bone health not only through treating isolated hormonal abnormalities but possibly also through a direct effect on bones [88] while at the same time resolving several other neuroendocrine [89] and reproductive dysfunctions [90–94]. Although studies in AN with leptin replacement have not been performed, the effect of leptin in women with HA elucidates mechanisms that are operational in both states and highlights the importance of a coordinated correction of all abnormalities [95]. Further studies evaluating the role of leptin in optimizing peak bone mass accrual and the actual reduction of fracture risk in the long term in both groups of AN and HA subjects remain to be fully elucidated.

Although these novel treatment options bring promises, one could argue that the correction of the underlying energy deficits state via nutritional intervention should be the mainstay of treatment in all HA and AN subjects, as weight regain leads to the concomitant full normalization of all neuroendocrine axes. Although this is true, we should bear in mind that such intervention takes time; and while important, psychiatric interventions in AN, including pharmacotherapy and behavioral interventions, might yield varying degree of success [96]. In the meantime, the hormonal and neuroendocrine abnormalities continue to take a toll on bone health, resulting in the failure to achieve peak bone mass in these subjects who are usually at a critical stage of peak bone mass accretion, with possible long-term

consequences on bone health and fracture risk [97,98]. Therefore, we could envision a complementary role of these novel treatments in the management of AN as a means to optimize bone health while awaiting adequate weight regain to be achieved.

Moving forward, further longer-term studies including comparative studies are necessary to evaluate which of these novel therapies, either in isolation or in combination, will be the most ideal in optimizing bone health in women with AN and HA. Treatment aimed at achieving peak bone mass in this group of often young female patients who are at a critical stage of bone mass accrual might be more important than that merely preventing continual bone lost. In addition, larger trials focusing on stress fractures are needed. Besides bone and reproductive health, the possibility that AN may result in multiple health consequences including cardiovascular complications and increased mortality [99,100] suggests that the impact of these conditions on health may involve multiple pathophysiological abnormalities. Therefore, treatment addressing the entire multitude of hormonal, reproductive, neuroendocrine, and bone abnormalities will likely yield more significant outcomes in contrast to targeting isolated hormonal deficiencies. Lastly, as the target subjects are usually young and active women, treatment regimen with convenient dosing without subcutaneous or other invasive route of administration will be helpful in improving acceptance and compliance. Future research will hopefully bring about better treatment options that will allow these two conditions to be treated more appropriately and effectively.

References

1. Hoek HW, van Hoeken D. Review of the prevalence and incidence of eating disorders. Int J Eat Disord. 2003; 34:383–96. [PubMed: 14566926]
2. Walters EE, Kendler KS. Anorexia nervosa and anorexic-like syndromes in a population-based female twin sample. Am J Psychiatry. 1995; 152:64–71. [PubMed: 7802123]
3. Diagnostic and statistical manual of mental disorders : DSM-IV. Washington (DC): American Psychiatric Association; 1994.
4. Miller KK, Parulekar MS, Schoenfeld E, et al. Decreased leptin levels in normal weight women with hypothalamic amenorrhea: the effects of body composition and nutritional intake. J Clin Endocrinol Metab. 1998; 83:2309–12. [PubMed: 9661600]
5. Chan JL, Mantzoros CS. Role of leptin in energy-deprivation states: normal human physiology and clinical implications for hypothalamic amenorrhoea and anorexia nervosa. Lancet. 2005; 366:74–85. [PubMed: 15993236]
6. Highet R. Athletic amenorrhoea. An update on aetiology, complications and management. Sports Med. 1989; 7:82–108. [PubMed: 2646673]
7. Reindollar RH, Novak M, Tho SP, et al. Adult-onset amenorrhea: a study of 262 patients. Am J Obstet Gynecol. 1986; 155:531–43. [PubMed: 3529965]
8. Ahima RS. Body fat, leptin, and hypothalamic amenorrhea. N Engl J Med. 2004; 351:959–62. [PubMed: 15342803]
9. Poyastro Pinheiro A, Thornton LM, Plotonicov KH, et al. Patterns of menstrual disturbance in eating disorders. Int J Eat Disord. 2007; 40:424–34. [PubMed: 17497704]
10. Bulik CM, Sullivan PF, Fear JL, et al. Fertility and reproduction in women with anorexia nervosa: a controlled study. J Clin Psychiatry. 1999; 60:130–5. [quiz 5–7]. [PubMed: 10084645]
11. Miller KK, Grinspoon SK, Ciampa J, et al. Medical findings in outpatients with anorexia nervosa. Arch Intern Med. 2005; 165:561–6. [PubMed: 15767533]
12. Cooke RA, Chambers JB, Singh R, et al. QT interval in anorexia nervosa. Br Heart J. 1994; 72:69–73. [PubMed: 8068473]

13. Treatment of patients with eating disorders, third edition. American Psychiatric Association. Am J Psychiatry. 2006; 163:4–54. [PubMed: 16925191]
14. Mitchell JE, Crow S. Medical complications of anorexia nervosa and bulimia nervosa. Curr Opin Psychiatry. 2006; 19:438–43. [PubMed: 16721178]
15. Santoro N, Filicori M, Crowley WF Jr. Hypogonadotropic disorders in men and women: diagnosis and therapy with pulsatile gonadotropin-releasing hormone. Endocr Rev. 1986; 7:11–23. [PubMed: 3082615]
16. Boyar RM, Katz J, Finkelstein JW, et al. Anorexia nervosa. Immaturity of the 24-hour luteinizing hormone secretory pattern. N Engl J Med. 1974; 291:861–5. [PubMed: 4412035]
17. Miller KK, Lawson EA, Mathur V, et al. Androgens in women with anorexia nervosa and normal-weight women with hypothalamic amenorrhea. J Clin Endocrinol Metab. 2007; 92:1334–9. [PubMed: 17284620]
18. Misra M, Miller KK, Almazan C, et al. Alterations in cortisol secretory dynamics in adolescent girls with anorexia nervosa and effects on bone metabolism. J Clin Endocrinol Metab. 2004; 89:4972–80. [PubMed: 15472193]
19. Khosla S, Melton LJ 3rd, Riggs BL. The unitary model for estrogen deficiency and the pathogenesis of osteoporosis: is a revision needed? J Bone Miner Res. 2011; 26:441–51. [PubMed: 20928874]
20. Longcope C. Adrenal and gonadal androgen secretion in normal females. Clin Endocrinol Metab. 1986; 15:213–28.
21. Kroboth PD, Salek FS, Pittenger AL, et al. DHEA and DHEA-S: a review. J Clin Pharmacol. 1999; 39:327–48. [PubMed: 10197292]
22. Gordon CM, Grace E, Emans SJ, et al. Changes in bone turnover markers and menstrual function after short-term oral DHEA in young women with anorexia nervosa. J Bone Miner Res. 1999; 14:136–45. [PubMed: 9893076]
23. Coenen CM, Thomas CM, Borm GF, et al. Changes in androgens during treatment with four low-dose contraceptives. Contraception. 1996; 53:171–6. [PubMed: 8689882]
24. White T, Jain JK, Stanczyk FZ. Effect of oral versus transdermal steroidal contraceptives on androgenic markers. Am J Obstet Gynecol. 2005; 192:2055–9. [PubMed: 15970897]
25. Carlstrom K, Karlsson R, Von Schoultz B. Diurnal rhythm and effects of oral contraceptives on serum dehydroepiandrosterone sulfate (DHEAS) are related to alterations in serum albumin rather than to changes in adrenocortical steroid secretion. Scand J Clin Lab Invest. 2002; 62:361–8. [PubMed: 12387582]
26. Colvard DS, Eriksen EF, Keeting PE, et al. Identification of androgen receptors in normal human osteoblast-like cells. Proc Natl Acad Sci U S A. 1989; 86:854–7. [PubMed: 2915981]
27. Gordon CM, LeBoff MS, Glowacki J. Adrenal and gonadal steroids inhibit IL-6 secretion by human marrow cells. Cytokine. 2001; 16:178–86. [PubMed: 11814313]
28. Khosla S, Riggs BL, Robb RA, et al. Relationship of volumetric bone density and structural parameters at different skeletal sites to sex steroid levels in women. J Clin Endocrinol Metab. 2005; 90:5096–103. [PubMed: 15998772]
29. Steinberg KK, Freni-Titulaer LW, DePuey EG, et al. Sex steroids and bone density in premenopausal and perimenopausal women. J Clin Endocrinol Metab. 1989; 69:533–9. [PubMed: 2527242]
30. Stoving RK, Veldhuis JD, Flyvbjerg A, et al. Jointly amplified basal and pulsatile growth hormone (GH) secretion and increased process irregularity in women with anorexia nervosa: indirect evidence for disruption of feedback regulation within the GH-insulin-like growth factor I axis. J Clin Endocrinol Metab. 1999; 84:2056–63. [PubMed: 10372710]
31. Misra M, Miller KK, Bjornson J, et al. Alterations in growth hormone secretory dynamics in adolescent girls with anorexia nervosa and effects on bone metabolism. J Clin Endocrinol Metab. 2003; 88:5615–23. [PubMed: 14671143]
32. Laughlin GA, Yen SS. Nutritional and endocrine-metabolic aberrations in amenorrheic athletes. J Clin Endocrinol Metab. 1996; 81:4301–9. [PubMed: 8954031]

33. Kaklamani VG, Linos A, Kaklamani E, et al. Dietary fat and carbohydrates are independently associated with circulating insulin-like growth factor 1 and insulin-like growth factor–binding protein 3 concentrations in healthy adults. J Clin Oncol. 1999; 17:3291–8. [PubMed: 10506632]
34. Kaklamani VG, Linos A, Kaklamani E, et al. Age, sex, and smoking are predictors of circulating insulin-like growth factor 1 and insulin-like growth factor–binding protein 3. J Clin Oncol. 1999; 17:813–7. [PubMed: 10071271]
35. Hock JM, Centrella M, Canalis E. Insulin-like growth factor I has independent effects on bone matrix formation and cell replication. Endocrinology. 1988; 122:254–60. [PubMed: 3335207]
36. Ohlsson C, Bengtsson BA, Isaksson OG, et al. Growth hormone and bone. Endocr Rev. 1998; 19:55–79. [PubMed: 9494780]
37. Lawson EA, Miller KK, Bredella MA, et al. Hormone predictors of abnormal bone microarchitecture in women with anorexia nervosa. Bone. 2010; 46:458–63. [PubMed: 19747572]
38. Loucks AB, Mortola JF, Girton L, et al. Alterations in the hypothalamic-pituitary-ovarian and the hypothalamic-pituitary-adrenal axes in athletic women. J Clin Endocrinol Metab. 1989; 68:402–11. [PubMed: 2537332]
39. Chyun YS, Kream BE, Raisz LG. Cortisol decreases bone formation by inhibiting periosteal cell proliferation. Endocrinology. 1984; 114:477–80. [PubMed: 6690287]
40. Lawson EA, Donoho D, Miller KK, et al. Hypercortisolemia is associated with severity of bone loss and depression in hypothalamic amenorrhea and anorexia nervosa. J Clin Endocrinol Metab. 2009; 94:4710–6. [PubMed: 19837921]
41. Gordon CM. Normal bone accretion and effects of nutritional disorders in childhood. J Womens Health (Larchmt). 2003; 12:137–43. [PubMed: 12737711]
42. Rencken ML, Chesnut CH III, Drinkwater BL. Bone density at multiple skeletal sites in amenorrheic athletes. J Am Med Assoc. 1996; 276:238–40.
43. Mehler PS, MacKenzie TD. Treatment of osteopenia and osteoporosis in anorexia nervosa: a systematic review of the literature. Int J Eat Disord. 2009; 42:195–201. [PubMed: 18951456]
44. Miller KK, Lee EE, Lawson EA, et al. Determinants of skeletal loss and recovery in anorexia nervosa. J Clin Endocrinol Metab. 2006; 91:2931–7. [PubMed: 16735492]
45. Bachrach LK, Guido D, Katzman D, et al. Decreased bone density in adolescent girls with anorexia nervosa. Pediatrics. 1990; 86:440–7. [PubMed: 2388792]
46. Grinspoon S, Baum H, Lee K, et al. Effects of short-term recombinant human insulin-like growth factor I administration on bone turnover in osteopenic women with anorexia nervosa. J Clin Endocrinol Metab. 1996; 81:3864–70. [PubMed: 8923830]
47. Walsh CJ, Phan CM, Misra M, et al. Women with anorexia nervosa: finite element and trabecular structure analysis by using flat-panel volume CT. Radiology. 2010; 257:167–74. [PubMed: 20713613]
48. Biller BM, Saxe V, Herzog DB, et al. Mechanisms of osteoporosis in adult and adolescent women with anorexia nervosa. J Clin Endocrinol Metab. 1989; 68:548–54. [PubMed: 2493036]
49. Marcus R, Cann C, Madvig P, et al. Menstrual function and bone mass in elite women distance runners. Endocrine and metabolic features. Ann Intern Med. 1985; 102:158–63. [PubMed: 3966752]
50. Warren MP, Brooks-Gunn J, Hamilton LH, et al. Scoliosis and fractures in young ballet dancers. Relation to delayed menarche and secondary amenorrhea. N Engl J Med. 1986; 314:1348–53. [PubMed: 3451741]
51. Dominguez J, Goodman L, Sen Gupta S, et al. Treatment of anorexia nervosa is associated with increases in bone mineral density, and recovery is a biphasic process involving both nutrition and return of menses. Am J Clin Nutr. 2007; 86:92–9. [PubMed: 17616767]
52. Jacoangeli F, Masala S, Staar Mezzasalma F, et al. Amenorrhea after weight recover in anorexia nervosa: role of body composition and endocrine abnormalities. Eat Weight Disord. 2006; 11:e20–6. [PubMed: 16801735]
53. Herzog W, Minne H, Deter C, et al. Outcome of bone mineral density in anorexia nervosa patients 11.7 years after first admission. J Bone Miner Res. 1993; 8:597–605. [PubMed: 8511987]
54. Golden NH, Katzman DK, Kreipe RE, et al. Eating disorders in adolescents: position paper of the Society for Adolescent Medicine. J Adolesc Health. 2003; 33:496–503. [PubMed: 14642712]

55. Al-Daghri NM, Alkharfy KM, Al-Saleh Y, et al. Modest reversal of metabolic syndrome manifestations with vitamin D status correction: a 12-month prospective study. Metabolism. 2012; 61:661–6. [PubMed: 22075268]
56. Klibanski A, Biller BM, Schoenfeld DA, et al. The effects of estrogen administration on trabecular bone loss in young women with anorexia nervosa. J Clin Endocrinol Metab. 1995; 80:898–904. [PubMed: 7883849]
57. Soyka LA, Misra M, Frenchman A, et al. Abnormal bone mineral accrual in adolescent girls with anorexia nervosa. J Clin Endocrinol Metab. 2002; 87:4177–85. [PubMed: 12213868]
58. Soyka LA, Grinspoon S, Levitsky LL, et al. The effects of anorexia nervosa on bone metabolism in female adolescents. J Clin Endocrinol Metab. 1999; 84:4489–96. [PubMed: 10599707]
59. Strokosch GR, Friedman AJ, Wu SC, et al. Effects of an oral contraceptive (norgestimate/ethinyl estradiol) on bone mineral density in adolescent females with anorexia nervosa: a double-blind, placebo-controlled study. J Adolesc Health. 2006; 39:819–27. [PubMed: 17116511]
60. Golden NH, Lanzkowsky L, Schebendach J, et al. The effect of estrogen-progestin treatment on bone mineral density in anorexia nervosa. J Pediatr Adolesc Gynecol. 2002; 15:135–43. [PubMed: 12106749]
61. Munoz MT, Morande G, Garcia-Centenera JA, et al. The effects of estrogen administration on bone mineral density in adolescents with anorexia nervosa. Eur J Endocrinol. 2002; 146:45–50. [PubMed: 11751066]
62. Misra M, Katzman D, Miller KK, et al. Physiologic estrogen replacement increases bone density in adolescent girls with anorexia nervosa. J Bone Miner Res. 2011; 26:2430–8. [PubMed: 21698665]
63. Grinspoon S, Thomas L, Miller K, et al. Effects of recombinant human IGF-I and oral contraceptive administration on bone density in anorexia nervosa. J Clin Endocrinol Metab. 2002; 87:2883–91. [PubMed: 12050268]
64. Labrie F, Diamond P, Cusan L, et al. Effect of 12-month dehydroepiandrosterone replacement therapy on bone, vagina, and endometrium in postmenopausal women. J Clin Endocrinol Metab. 1997; 82:3498–505. [PubMed: 9329392]
65. von Muhlen D, Laughlin GA, Kritz-Silverstein D, et al. Effect of dehydroepiandrosterone supplementation on bone mineral density, bone markers, and body composition in older adults: the DAWN trial. Osteoporos Int. 2008; 19:699–707. [PubMed: 18084691]
66. Gordon CM, Grace E, Emans SJ, et al. Effects of oral dehydroepiandrosterone on bone density in young women with anorexia nervosa: a randomized trial. J Clin Endocrinol Metab. 2002; 87:4935–41. [PubMed: 12414853]
67. Miller KK, Grieco KA, Klibanski A. Testosterone administration in women with anorexia nervosa. J Clin Endocrinol Metab. 2005; 90:1428–33. [PubMed: 15613421]
68. Miller KK, Meenaghan E, Lawson EA, et al. Effects of risedronate and low-dose transdermal testosterone on bone mineral density in women with anorexia nervosa: a randomized, placebo-controlled study. J Clin Endocrinol Metab. 2011; 96:2081–8. [PubMed: 21525157]
69. Golden NH, Iglesias EA, Jacobson MS, et al. Alendronate for the treatment of osteopenia in anorexia nervosa: a randomized, double-blind, placebo-controlled trial. J Clin Endocrinol Metab. 2005; 90:3179–85. [PubMed: 15784715]
70. Miller KK, Grieco KA, Mulder J, et al. Effects of risedronate on bone density in anorexia nervosa. J Clin Endocrinol Metab. 2004; 89:3903–6. [PubMed: 15292325]
71. Warren MP, Brooks-Gunn J, Fox RP, et al. Persistent osteopenia in ballet dancers with amenorrhea and delayed menarche despite hormone therapy: a longitudinal study. Fertil Steril. 2003; 80:398–404. [PubMed: 12909505]
72. Ettinger B, Pressman A, Sklarin P, et al. Associations between low levels of serum estradiol, bone density, and fractures among elderly women: the study of osteoporotic fractures. J Clin Endocrinol Metab. 1998; 83:2239–43. [PubMed: 9661589]
73. McNicholl DM, Heaney LG. The safety of bisphosphonate use in pre-menopausal women on corticosteroids. Curr Drug Saf. 2010; 5:182–7. [PubMed: 19534636]
74. Vescovi JD, Jamal SA, De Souza MJ. Strategies to reverse bone loss in women with functional hypothalamic amenorrhea: a systematic review of the literature. Osteoporos Int. 2008; 19:465–78. [PubMed: 18180975]

75. DiVasta AD, Feldman HA, Giancaterino C, et al. The effect of gonadal and adrenal steroid therapy on skeletal health in adolescents and young women with anorexia nervosa. Metabolism. 2012; 61:1010–20. [PubMed: 22257645]
76. Thompson RD, Carlson M. Liquid chromatographic determination of dehydroepiandrosterone (DHEA) in dietary supplement products. J AOAC Int. 2000; 83:847–57. [PubMed: 10995111]
77. Pappa T, Vemmos K, Saltiki K, et al. Severity and outcome of acute stroke in women: relation to adrenal sex steroid levels. Metabolism. 2012; 61:84–91. [PubMed: 21820139]
78. Creatsa M, Armeni E, Stamatelopoulos K, et al. Circulating androgen levels are associated with subclinical atherosclerosis and arterial stiffness in healthy recently menopausal women. Metabolism. 2012; 61:193–201. [PubMed: 21820132]
79. Mantzoros C. Role of leptin in reproduction. Ann N Y Acad Sci. 2000; 900:174–83. [PubMed: 10818404]
80. Audi L, Mantzoros CS, Vidal-Puig A, et al. Leptin in relation to resumption of menses in women with anorexia nervosa. Mol Psychiatry. 1998; 3:544–7. [PubMed: 9857982]
81. Mantzoros CS, Cramer DW, Liberman RF, et al. Predictive value of serum and follicular fluid leptin concentrations during assisted reproductive cycles in normal women and in women with the polycystic ovarian syndrome. Hum Reprod. 2000; 15:539–44. [PubMed: 10686193]
82. Bluher S, Mantzoros CS. Leptin in humans: lessons from translational research. Am J Clin Nutr. 2009; 89:991S–7S. [PubMed: 19176740]
83. Brennan AM, Mantzoros CS. Drug Insight: the role of leptin in human physiology and pathophysiology—emerging clinical applications. Nat Clin Pract Endocrinol Metab. 2006; 2:318–27. [PubMed: 16932309]
84. Mantzoros CS, Flier JS. Editorial: leptin as a therapeutic agent—trials and tribulations. J Clin Endocrinol Metab. 2000; 85:4000–2. [PubMed: 11095422]
85. Welt CK, Chan JL, Bullen J, et al. Recombinant human leptin in women with hypothalamic amenorrhea. N Engl J Med. 2004; 351:987–97. [PubMed: 15342807]
86. Chou SH, Chamberland JP, Liu X, et al. Leptin is an effective treatment for hypothalamic amenorrhea. Proc Natl Acad Sci U S A. 2011; 108:6585–90. [PubMed: 21464293]
87. Sienkiewicz E, Magkos F, Aronis KN, et al. Long-term metreleptin treatment increases bone mineral density and content at the lumbar spine of lean hypoleptinemic women. Metabolism. 2011; 60:1211–21. [PubMed: 21741057]
88. Iwamoto I, Fujino T, Douchi T. The leptin receptor in human osteoblasts and the direct effect of leptin on bone metabolism. Gynecol Endocrinol. 2004; 19:97–104. [PubMed: 15624271]
89. Chan JL, Matarese G, Shetty GK, et al. Differential regulation of metabolic, neuroendocrine, and immune function by leptin in humans. Proc Natl Acad Sci U S A. 2006; 103:8481–6. [PubMed: 16714386]
90. Chan JL, Bullen J, Stoyneva V, et al. Recombinant methionyl human leptin administration to achieve high physiologic or pharmacologic leptin levels does not alter circulating inflammatory marker levels in humans with leptin sufficiency or excess. J Clin Endocrinol Metab. 2005; 90:1618–24. [PubMed: 15613407]
91. Chan JL, Moschos SJ, Bullen J, et al. Recombinant methionyl human leptin administration activates signal transducer and activator of transcription 3 signaling in peripheral blood mononuclear cells in vivo and regulates soluble tumor necrosis factor-alpha receptor levels in humans with relative leptin deficiency. J Clin Endocrinol Metab. 2005; 90:1625–31. [PubMed: 15613409]
92. Lee JH, Chan JL, Sourlas E, et al. Recombinant methionyl human leptin therapy in replacement doses improves insulin resistance and metabolic profile in patients with lipoatrophy and metabolic syndrome induced by the highly active antiretroviral therapy. J Clin Endocrinol Metab. 2006; 91:2605–11. [PubMed: 16636130]
93. Moon HS, Matarese G, Brennan AM, et al. Efficacy of metreleptin in obese patients with type 2 diabetes: cellular and molecular pathways underlying leptin tolerance. Diabetes. 2011; 60:1647–56. [PubMed: 21617185]
94. Matarese G, Moschos S, Mantzoros CS. Leptin in immunology. J Immunol. 2005; 174:3137–42. [PubMed: 15749839]

95. Kelesidis T, Kelesidis I, Chou S, et al. Narrative review: the role of leptin in human physiology: emerging clinical applications. Ann Intern Med. 2010; 152:93–100. [PubMed: 20083828]

96. Bulik CM, Berkman ND, Brownley KA, et al. Anorexia nervosa treatment: a systematic review of randomized controlled trials. Int J Eat Disord. 2007; 40:310–20. [PubMed: 17370290]

97. Barrack MT, Rauh MJ, Nichols JF. Cross-sectional evidence of suppressed bone mineral accrual among female adolescent runners. J Bone Miner Res. 2010; 25:1850–7. [PubMed: 20200979]

98. Christo K, Prabhakaran R, Lamparello B, et al. Bone metabolism in adolescent athletes with amenorrhea, athletes with eumenorrhea, and control subjects. Pediatrics. 2008; 121:1127–36. [PubMed: 18519482]

99. Friday KE, Drinkwater BL, Bruemmer B, et al. Elevated plasma low-density lipoprotein and high-density lipoprotein cholesterol levels in amenorrheic athletes: effects of endogenous hormone status and nutrient intake. J Clin Endocrinol Metab. 1993; 77:1605–9. [PubMed: 8263148]

100. Olivares JL, Vazquez M, Fleta J, et al. Cardiac findings in adolescents with anorexia nervosa at diagnosis and after weight restoration. Eur J Pediatr. 2005; 164:383–6. [PubMed: 15909184]