0021-972X/97/$03.00/0
Journal of Clinical Endocrinology and Metabolism

Vol. 82, No. 2
Printed in U.S.A.

Cross Genotype Sex Hormone Treatment in Two Cases of Hypogonadal Osteoporosis

IRIS VERED, IGOR KAISERMAN, BEN-AMI SELA, AND JOSEPH SACK

Pediatric Endocrinology Unit, Institute of Endocrinology, Chaim Sheba Medical Center, Tel Hashomer, Israel; Sackler School of Medicine, Tel-Aviv University, Tel-Aviv, Israel

ABSTRACT

Background: Sex hormone deficiency is the most common cause of bone loss. Reduced bone mass and an increased risk for osteoporotic fractures have been described in hypogonadal subjects of both sexes. We present here the results of treating two patients showing abnormal sexual differentiation (an XX male and an XY female), who suffered from bone loss related to sex hormone deficiency, with cross genotype sex hormones.

Subjects and Methods: Patient 1 was an asymptomatic 39-yr-old XY female with complete androgen insensitivity. Her testes had been removed, and she later discontinued estrogen treatment. Patient 2, a 37-yr-old XX male, had congenital adrenal hyperplasia, which led to a masculine phenotype. He was ovariectomized and reared as a male. He was treated with glucocorticoids but refused androgen treatment for many years. We treated both patients with phenotypically matched sex hormones (patient 1 received conjugated estrogens 1.25 mg/day, and patient 2 received 250 mg testosterone every 4 weeks) and followed their bone mineral density (BMD) using dual-energy X-ray absorptiometry, urine calcium, and hydroxyproline excretion.

Results: Before treatment both patients had low sex hormones and highly elevated gonadotropins. As a result of treatment urine hydroxyproline excretion decreased from 45 and 26.7 mg/g creatinine to 15 and 15.9 mg/g creatinine in patients 1 and 2 respectively. In patient 1, lumbar BMD rose from 0.912gr/cm^2 to 0.976gr/cm^2 and femoral neck BMD rose from 0.716gr/cm^2 to 0.836gr/cm^2 after 4 years of treatment. In patient 2, lumbar BMD rose from 0.717gr/cm^2 to 0.815gr/cm^2 and the femoral neck BMD rose from 0.509gr/cm^2 to 0.635gr/cm^2 after 27 months of treatment.

Conclusions: Phenotypically-matched sex hormone therapy in patients with abnormal sexual differentiation is essential not only to maintain external appearance but also for the preservation of bone mass. (*J Clin Endocrinol Metab* **82:** 576–578, 1997)

SEX HORMONE deficiency is the most common cause of bone loss. Reduced bone mass and an increased risk of osteoporotic fractures have been described in hypogonadal subjects of both sexes in various age groups (1). Replacement of sex hormones is the treatment of choice for hypogonadal osteoporosis, and its role in prevention of osteoporosis has been well established (1). For obvious reasons, hormone replacement treatment for osteoporosis in humans is usually based on the use of sex-matched sex hormones. Thus, limited information exists regarding the effects of cross-genotype sex hormones on human bone.

Estrogens and androgens bind to specific nuclear receptors in the bone cells of both sexes (2, 3). Estrogen and testosterone elicit similar biologic responses both in cultured bone cells and in castrated animals of both sexes. Testosterone is converted into dihydro-testosterone and estradiol in bone (4), but the specific impact of each steroid hormone on male or female bone is unknown.

Patients with cross-genotype sex assignment resulting from abnormal sexual development are natural models to study the sex-specific response of the skeleton to hormones. We had the opportunity to follow up two patients with abnormal sexual differentiation, an XY female and an XX male, in whom bone loss related to sex hormone deficiency was diagnosed. We present here the clinical and laboratory data on these patients and their responses to cross-genotype sex hormone administration.

Received July 17, 1996. Revision received September 16, 1996. Accepted September 24, 1996.

Address correspondence and reprint requests to: Professor Joseph Sack, Department of Pediatrics, Sheba Medical Center, Tel-Hashomer, 52621 Israel. Email: Igork@cc.huji.ac.il

Subjects and Methods

Patient 1

Patient 1 was an asymptomatic 39-yr-old XY female, self-referred, who was concerned about osteoporosis. Complete androgen insensitivity had been diagnosed at age 17, when she was investigated for primary amenorrhea. The abdominal testes were removed at age 32, and estrogen treatment was initiated but then discontinued by the patient after 4 months. She was a heavy smoker, and her calcium intake was estimated at 1200 mg/day. After initial evaluation, conjugated estrogens were prescribed (1.25 mg/day), and we followed her up for 4 yr.

Patient 2

Patient 2 was a 37-yr-old XX male, referred for evaluation of his bone density. Congenital adrenal hyperplasia (11β hydroxylase deficiency) had been diagnosed at the age of 1 yr, following the onset of pseudoprecocious puberty. He was ovariectomized and reared as a male, but was not given glucocorticoids. At age 10 he suffered a hypertensive crisis. At that time he was 139 cm tall, fully virilized, hyperpigmented, with bone age over 18 yr. XX karyotype and 11 β-hydroxylase deficiency were documented. Since then, he has been treated with glucocorticoids, thiazide diuretics and β-blockers. Androgen treatment was suggested many times but refused. The patient was a nonsmoker, and his dietary calcium intake was estimated at 500 mg/day. After initial evaluation he was given testosterone-depot injections, 250 mg every 4 weeks, and 600 mg/day calcium supplement. He was followed up for 27 months.

During the follow-up of these patients, serum FSH, LH, 17β-Estradiol, DHEAS, TSH, and T_4 were measured using RIA kits (DPC, LA Ca). Urinary hydroxyproline was measured by a refinement of a specific assay (5), and bone mineral density was measured using dual energy X-ray absortiometry (DXA) (LUNAR, Madison, WI). The *in vivo* precision in our laboratory is 1% for L2-L4 BMD, and 1.8% for femoral neck BMD.

Results

Patient 1

Patient 1 was 181 cm tall, a slender female, weighing 67 kg, with well-developed breasts, scanty pubic and axilary hair, and normal external genitalia.

Her initial laboratory tests revealed normal kidney and liver functions, normal blood count, and normal levels of serum calcium, phosphorus, alkaline phosphatase, PTH, and thyroid hormones. Serum gonadotropins were elevated (FSH 40 IU/L, LH 46 IU/L), while 17-β estradiol (<20 pg/mL) and testosterone (0.1 ng/mL) were below the normal range. DHEAS was mildly elevated (3.6 μg/mL, normal <3). Table 1 presents the decrease in urine calcium and hydroxyproline with estrogen therapy. Bone density in the lumbar spine and proximal femur was originally lower than expected for a female of that age. Fig. 1 presents the bone mineral density (BMD) at 1 yr intervals up to 4 yr after the estrogen treatment was started. As can be seen, the lumbar spine density rose from 0.912 gr/cm^2 (81% of young normal mean) to 0.976 gr/cm^2 (84%) and the femoral neck bone density rose from 0.716 gr/cm^2 (78% of young normal mean) to 0.836 gr/cm^2 (85%).

Patient 2

Patient 2 was a 137 cm tall male, weighing 67 kg, with little facial hair, feminine hair distribution, small penis, and empty scrotum. His initial laboratory tests revealed normal kidney and liver functions, normal blood count, and normal levels of serum calcium, phosphorus, alkaline phosphatase, PTH, and thyroid hormones. The gonadotropins were high (FSH 40.4 IU/L, LH 27.7 IU/L), 17-β estradiol (<20 pg/mL), testosterone (<0.1 ng/mL) and DHEAS (<0.05 pg/mL) were very low. Compound S was adequately suppressed (0.1 μg/L). Table 1 presents the changes in urine calcium and hydroxyproline with testosterone therapy. Bone mineral density at the lumbar spine and proximal femur was markedly decreased at baseline and increased at both sites during the 27 months of therapy. Fig. 2 presents the bone mineral density up to 27 months after the initiation of testosterone treatment. As can be seen, the lumbar spine density rose from 0.717 gr/cm^2 (58% of young male normal mean) to 0.815 gr/cm^2 (66%), and the femoral neck bone density rose from 0.509gr/cm^2 (51% of young male normal mean) to 0.635 gr/cm^2 (59%).

Discussion

The aim of this study was to learn about the effects of cross-genotype sex hormone treatment in human subjects.

TABLE 1. Urine calcium and hydroxyproline levels before and during treatment with cross-genotype sex hormones

Patient number	Time during cross-genotype sex hormone treatment	Urine calcium mg/24 hours	Urine hydroxyproline mg/g creatinine (normal <25)
1	Pre-treatment	92	49
	1 year	55	15
2	Pre-treatment	266	26.7
	1 year	322*	15.9

* Treatment included calcium supplement (600 mg/day).

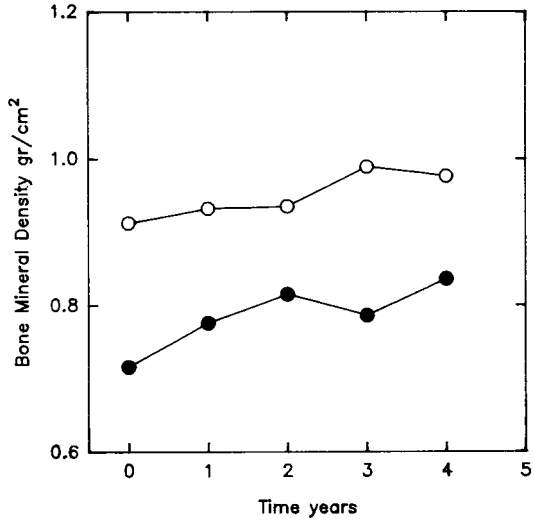

FIG. 1. The bone mineral density of patient 1 during 4 yr of treatment with estrogen. ○, lumbar spine L2-L4; ●, femoral neck.

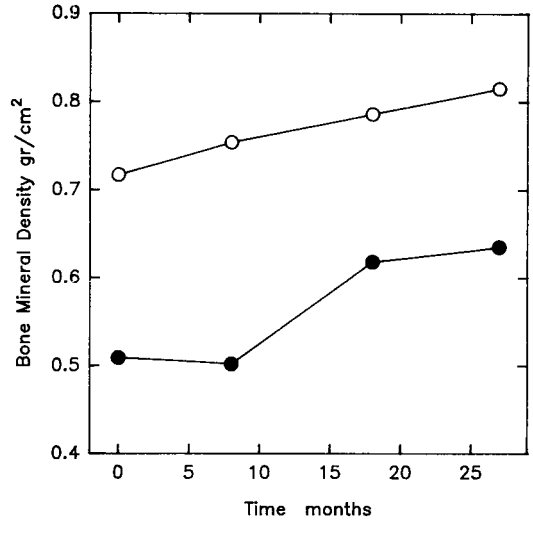

FIG. 2. The bone mineral density of patient 2 during 27 months of treatment with testosterone. ○, lumbar spine L2-L4; ●, femoral neck.

Both estrogens and androgens elicit similar biological responses in bone cells and in animal models (2–4). Both affect bone cell proliferation and alkaline phosphatase production (6, 7), increase type I collagen mRNA (6), decrease PTH-stimulated PGE_2 production (8, 9), decrease cAMP production (6, 10), increase mRNA for TGF-β (11, 12), and inhibit the increase in bone turnover and the resultant bone loss in castrated male rats (13).

In human subjects, the anabolic steroid nandrolone seems to increase bone mass in osteoporotic post-menopausal women (14), and in one case-controlled study there was a lower, albeit nonsignificant, risk for hip fracture in post-menopausal women taking anabolic steroids (15). Bone mass was maintained in young women treated with danazol for endometriosis despite a low-estrogen state (16), and male to female transsexuals treated with estrogens and antiandrogens showed suppressed bone turnover and normal bone mass compared with age-matched healthy males (17).

We present here two cases of cross-genotype sex assignment who were not treated with sex hormones following gonadectomy. Both showed accelerated bone resorption and compromised bone density. The first patient, a genetic male with feminine phenotype resulting from lack of testosterone receptor activity, responded well to estrogen with a resultant increase in bone density. Thus, we might assume she had active estrogen receptors. The importance of the estrogen receptor in the male is demonstrated by the findings that lack of estrogen activity in the male, secondary to a defective estrogen receptor, leads to decreased mineral density and skeletal maturity even if testosterone concentration is normal (18).

The second patient, a genetic female with masculine phenotype caused by increased adrenal androgens, had a low bone density because of lack of estrogen (ovariectomy), and the suppression of adrenal androgens as well as the glucocorticoid treatment. Treatment with testosterone was successful in increasing bone density. This could be because of a dual sensitivity of the bone steroid receptor to both estrogen and testosterone or because of aromatization of testosterone to estradiol. The latter is supported by findings that P450 aromatase deficiency can lead to a dissociation between skeletal growth and the accretion of bone density and mass (19). The lack of *in situ* aromatization in the male results in tall stature, delayed epiphyseal closure, and osteoporosis.

In both our patients, phenotype-matched sex hormone therapy resulted in an increase in bone density. We assume that in both sexes it is probably the estrogen that causes a favorable effect on bone density when administered as conjugated estrogen or derived from testosterone by aromatization.

In conclusion, long-term, phenotype matched, sex hormone treatment is recommended in castrated patients with abnormal sexual differentiation, not only for sexual appearance and function, but also to preserve skeletal integrity. Thus, even patients who are not interested in therapy for external sexual appearance should be advised to continue treatment to maintain normal bone density.

References

1. **Turner RT, Riggs BI, Spelsbery TC.** 1994 Skeletal effects of estrogen. Endocr Rev. 15:275–300.
2. **Colvard DS, Erikson EF, Keeting PE, et al.** 1989 Identification of androgen receptors in normal human osteoblast-like cells. Proc Natl Acad Sci USA. 86:854–857.
3. **Erikson EF, Colvard DS, Berg NJ, et al.** 1988 Evidence of estrogen receptors in normal human osteoblast-like cells. Science. 241:84–86.
4. **Schweikert H, Ruff W, Niederle N, Schafer HE, Keck E, Kruck F.** 1980 Testosterone metabolism in human bone. Endocrinology. 94:325–329.
5. **Sela BA, Doolman R.** 1991 Refinement of a specific assay for hydroxyproline in urine. Clin Chim Acta. 203:91–94.
6. **Enrnst M, Heath JK, Rodan GA.** 1989 Estradiol effects on proliferation messenger ribonucleic acid for collagen and insulin-like growth factor-I and parathyroid-stimulated adenylate cyclase activity in osteoblastic cells from calvariae and long bone. Endocrinology. 125:825–833.
7. **Kasperk CH, Wergedal JE, Farkey JK, Linkhart TA, Turner RT, Baylink DJ.** 1989 Androgens directly stimulate proliferation of bone cells *in vitro.* Endocrinology. 124:1576–1578.
8. **Pilbeam CC, Klein-Nulend J, Raisz LG.** 1989 Inhibition by 17-β-estradiol of PTH-stimulated resorption and prostaglandin production in cultured neonatal mouse calvariae. Biochem Biophys Res Commun. 163:1319–1324.
9. **Pilbeam CC, Raisz LG.** 1990 Effects of androgens on parathyroid hormone and interleukin-1 stimulated prostaglandin production in cultured neonatal mouse calvariae. J Bone Miner Res. 5:1183–1188.
10. **Fukayama H, Tashjian AHJ.** 1989 Direct modulation by androgens of the response of human bone cells (SAOS-2) to human parathyroid hormone (PTH) and PTH-related protein. Endocrinology. 125:1789–1794.
11. **Komm BS, Terpening CM, Bentz DJ, et al.** 1988 Estrogen binding receptor mRNA and biologic responses in osteoblast-like osteosarcoma cells. Science. 241:81–84.
12. **Kasperk C, Fitzsimmons R, Strong D, et al.** 1990 Studies of the mechanism by which androgens enhance mitogenesis and differentiation in bone cells. J Clin Endrocrinol Metab. 71:1322–1329.
13. **Vanderschueren D, VanHerck E, Suiker AMH, Visser WJ, Schot LPC, Bouillon R.** 1992 Bone and mineral metabolism in aged male rats: short and long term effects of androgen deficiency. Endocrinology. 130:2906–2916.
14. **Need AG, Horowitz M, Walker CJ, Chatterton BE, Chapman IC, Nordin BEC.** 1989 Cross-over study of fat-corrected forearm mineral content during Mandrolone decanoate therapy for osteoporosis. Bone. 10:3–6.
15. **Kanis JA, Johnell O, Gullbery B, Allander E, Dilsen G, Gennari C, et al.** 1992 Evidence for efficacy of drug affecting bone metabolism in preventing hip fractures. Brit Med J. 305:1124–8.
16. **Somjen D, Weisman Y, Harell A, Berger E, Kay AM.** 1989 Direct and sex-specific stimulation by sex steroids of creatine kinase activity and DNA synthesis in rat bone. Proc Natl Acad Sci USA. 86:3361–3365.
17. **Lips P, Asscheman H, Unitewall P, Netelenbos JC, Gooren L.** 1989 The effects of cross-gender hormonal treatment on bone metabolism in male-to-female transsexuals. J Bone Miner Res. 4:657–662.
18. **Smith EP, Boyd J, Frank GR, et al.** 1994 Estrogen resistance caused by a mutation in the estrogen-receptor gene in a man. N Engl J Med. 331:1056–1061.
19. **Morishima A, Grumbach MM, Simpson ER, Fisher C, Qin K.** 1996 Aromatase deficiency in male and female siblings caused by a novel mutation and the physiological role of estrogen. J Clin Endocrinol Metab. 80:3689–3697.