Contribution of genetic variations in estradiol biosynthesis and metabolism enzymes to osteoporosis

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ABSTRACT

The occurrence of osteoporosis is closely related to serum estradiol level. CYP19 is the most important biosynthesis enzyme and CYP1A2, CYP3A4, and 17beta-HSD are important metabolism enzymes. Any changes in these enzymes activity affects estradiol biosynthesis or metabolism, and changes the estradiol serum level. By reviewing the recent literature, it was found that the genetic variations of CYP19, CYP1A2, CYP3A4, and 17beta-HSD are important factors affecting the estradiol serum level, and may be closely related to the development of osteoporosis.

INTRODUCTION

Bone is an estradiol-responsive tissue. Estrogen withdrawal during the menopause causes loss of bone mass and clinically relevant osteoporosis in a third of all women. There are many factors contributing to osteoporosis in postmenopausal women, such as ovary function, estrogen receptor, serum estradiol level, diet habits, and genetic reasons, etc. Among them, the serum estradiol level is the most direct reason for osteoporosis, which is greatly affected by the biosynthesis and metabolism enzymes. Aromatase (CYP19) is an important biosynthesis enzyme, catalyzing the aromatize reaction of dehydroepiandrosterone (DHEA) to form estrone. 17beta-Hydroxysteroid dehydrogenase (17beta-HSD) catalyzing the interconversion of estradiol to estrone exists in six types. 17beta-HSD1 primarily catalyzes the reductive pathway to convert estrone to estradiol, while both 17beta-HSD2 and 17beta-HSD4 catalyze the oxidative pathway to degrade estradiol to estrone. CYP1A2 and CYP3A4 may be the most important enzymes to catalyze estradiol 2-hydroxylation. Increase or decrease in its production, as well as the degradation of estradiol are important mechanisms of alleviation or aggravation of osteoporosis development. The genetic variations in estradiol biosynthesis and metabolism enzymes are major factors to control such changes through changing the enzyme activity. We herein review the contribution of CYP19, 17beta-HSD2, 17beta-HSD4, CYP1A2, and CYP3A4's genetic variation in the development of osteoporosis in postmenopausal women.

CYP19 GENETIC VARIATIONS AND OSTEOPOROSIS

CYP19 catalyzes the conversion of androgens to estrogens in a wide variety of tissues, including ovary, testis, placenta, brain, and adipose tissue. In particular, two recent studies indicate that it can be expressed in osteoblast cells and catalyze the conversion of androgens to estrogen. In these two studies, significant positive correlation between bone mineral density (BMD) and serum dehydroepiandrosterone sulfate (DHEA-S) was found in 120 postmenopausal women (51–99 years old) but no correlation was seen between BMD and serum estradiol (E2). In the subset analysis, strongly positive correlation of serum DHEA-S and estrone (E1) with BMD were observed in the postmenopausal women aged less than 69 years, respectively. Reverse transcription-polymerase chain reaction analysis revealed that phorbol myristate acetate (PMA) was the osteoblast-specific promoter. Dexamethasone and 1α, 25-dihydroxyvitamin D3 synergistically enhanced CYP19 activity and CYP19 mRNA expression. These suggest that decrease of CYP19 activity in case of genetic variation might contribute to the
pathogenesis of osteoporosis\(^2\).

The expression of CYP19 was regulated in a tissue-specific fashion through alternate use of multiple promoter-specific first exons. To date, eight different first exons have been reported in human CYP19, namely 1.1, 1.2, 1.3, 1.4, 1.5, PII, 2a, and 1f\(^3\). CYP19 deficiency caused by a novel mutation was found in male and female siblings, who suffered from severe hyper-androgenic and hypo-estrogenic symptoms\(^4\). Striking osteopenia of the wrist was noted. BMD indexes of the lumbar spine (cancellous bone) and distal radius (cortical bone) were consistent with osteoporosis; the distal radius was \(-4.7 \text{ SD below the mean value for age- and sex-matched normal men.}\). Increased extraglandular aromatization has also been reported as the cause of familial gynecomastia\(^5\). In the studied kindred, the CYP19 excess is inherited in an autosomal dominant manner. The affected males had heterosexual precocity and/or gynecomastia, while the affected females had isolated precocity and/or macromastia. Using the ratio of delta\(^4-\text{[H]}\) androstenedione to \([\text{H}]\) estrone to express the CYP19 activity, markedly increased aromatase activity was found in these patients. The study also found a new 5'-splice variant presented in CYP19 mRNA of patients. A polymorphic tetranucleotide repeats (TTTA)n in intron 5, about 80 nucleotides downstream of exon 4 have previously been described\(^6\). The allele frequencies of the polymorphic repeat were studied in 182 sporadic and 185 familial breast cancer patients as well as in 252 healthy control individuals. Five different alleles containing 7, 8, 9, 11, and 12-TTTA-repeats were detected. A relatively rare allele (A1) containing the longest repeat (TTTA)12 was found significantly more frequently in breast cancer patients than in control individuals. There is no literature reporting the contribution of the polymorphic tetranucleotide repeat (TTTA)n in intron 5 or any other genetic variation to osteoporosis in menopausal women.

17BETA-HSD GENETIC VARIATIONS AND OSTEOPOROSIS

17beta-HSDs have six types playing a crucial role in catalyzing the biosynthesis and/or inactivation of sex steroid hormones. 17beta-HSD1, which is expressed in placenta, granulosa cells, and a number of other tissues, primarily catalyzes the interconversion between weak estrogen estrone and strong estrogen estradiol\(^7\). 17beta-HSD2 is expressed in liver, placenta, endometrium, and small intestine and converts estradiol to estrone, testosterone to androstenedione as well as the inactive progestin, 20-dihydroprogesterone, to the active progestin, progesterone\(^8\). 17beta-HSD3 expressed only in the testes is an enzyme responsible for the conversion of the weak androgen androstenedione to the potent androgen testosterone, but it also catalyzes the conversion of dehydroepiandrosterone (DHEA) to androstenediol (Adiol) and E2 to E1\(^9\). 17beta-HSD4 is expressed in many tissues including liver, testis, ovary, prostate, and heart, but not in placenta, and it preferentially inactivates E2 to E1 and also converts Adiol to DHEA\(^10\). 17beta-HSD5 expressed in prostate mainly catalyzes androstenedione to testosterone\(^11\).

Normand et al studied the polymorphisms in 17beta-HSD2 gene at the EDH17B2 locus on 17q11-q21 and near the region of assignment of the gene BRCA1, which is involved in hereditary breast-ovarian cancer. They revealed a total of 11 allelic variants that were due to single base substitutions. Following study in twenty-six additional unrelated individuals, nine frequent and two rare polymorphisms were found. Seven of the 11 polymorphisms were in complete linkage disequilibrium. These polymorphisms in the 17beta-HSD2 gene provide markers that can be used for the genetic mapping of this locus, and may be used to establish whether 17 beta-HSD2 is a candidate gene for hereditary breast-ovarian cancer or osteoporosis\(^12\). 17beta-HSD4 is a multifunctional enzyme that is localized in the peroxisomes. Its N-terminal part has dehydrogenase activity, the central part has hydratase activity, while the carboxy-terminal part is responsible for sterol transport. Mutations in 17beta-HSD4 cDNA leading to a severe peroxisomal disorder motivated studies to define the genomic organization of this gene mapped to Chromosome (Chr) 5q2. According to these, Leenders et al studied the gene, and found that it consisted of 24 exons and 23 introns with classical intron-exon junctions spanning more than 100 kbp\(^13\). 17beta-HSD4 gene is stimulated by progestrone and ligands of PPARalpha (peroxisomal proliferator activated receptor alpha) such as clofibrate, and is down-regulated by phorbol esters. Mutations in the 17beta-HSD4 can lead to a fatal form of Zellweger syndrome\(^14\). Both 17beta-HSD2 and 17beta-HSD4 are expressed in osteoblasts and osteoblast-like osteosarcomas cells, and play important roles in regulating the action of estrogen through balance of the interconver-
sion of E2 and E1\(^{(15)}\). These imply that any variation of 17\(\beta\)-HSD2 and 17\(\beta\)-HSD4 that changes the enzyme activity may affect the bone metabolism. If the variation increases the enzyme activity, the balance of interconversion of E2 and E1 will be broken resulting in low E2 levels in osteoblasts and hence may cause serious osteoporosis. Studying the contribution of genetic variations in 17\(\beta\)-HSD2 and 17\(\beta\)-HSD4 to osteoporosis development in postmenopausal women is thus warranted.

CYP1A2 AND CYP3A4 GENETIC VARIATIONS AND OSTEOPOOROSIS

Studies with hamster liver microsomes indicate that CYP1A1 and CYP3A1 families catalyze the 2-hydroxylation of Est\(^{(16)}\). Using cDNA-expressed human cytochrome P450 to study mechanism of 2-hydroxylation, it was also found that both CYP1A2 and CYP3A4 catalyze the 2-hydroxylation\(^{(17)}\).

The analysis of urinary caffeine metabolites in human population revealed CYP1A2 phenotype polymorphism\(^{(18)}\). Using 17x/17x as the enzyme activity index to study the phenotype polymorphism in 200 Chinese people Ouyn et al found a 5.4 \% of PM and a 93.6 \% of EM\(^{(19)}\). DNA from 157 Chinese subjects (104 polychlorinated biphenyl-exposed subjects and 53 control subjects) was screened to identify the genetic polymorphism by single-strand conformation polymorphism method and MboII endonuclease digestion. Only 1 of 157 samples showed heterozygous C2866->G mutation. The caffeine breath test value is not significantly higher than the mean value of other polychlorinated biphenyl-exposed subjects. The incidence of the point mutation in these Chinese subjects is less than 1 \%\(^{(20)}\). Nucleotide sequence analysis revealed the existence of a point mutation from guanine (wild type) to adenine (mutated type) at position -2964 in the 5'-flanking region of CYP1A2 gene in the Japanese. This point mutation was detected by PRC-RFLP method using Ddel or BstI restriction enzyme, and was proven to be genetically inherited. Allele frequencies in 116 Japanese subjects showed 0.77 and 0.23 for the wild and mutated types of alleles, respectively. This point mutation caused a significant decrease of CYP1A2 activity measured by the rate of caffeine 3-demethylation in Japanese smokers (\(P < 0.05\))\(^{(21)}\). Sachse et al used caffeine (100 mg oral dose) probing and DNA sequence analysis to study the nucleotide polymorphism in intron 1 of the CYP1A2 gene at position 734 downstream of the first transcribed nucleotide, and the functional significance of this polymorphism in 185 healthy Caucasian non-smokers and 51 smokers\(^{(22)}\). They found that 46 \% of the subjects were homozygous for the variant A (normal type), 44 \% were heterozygous, and 10 \% were homozygous for the variant C (mutated type). The enzyme activity did not differ significantly between the three CYP1A2 genotypes in the 185 healthy non-smokers, whereas differed significantly in the 51 smokers. CYP3A4, the most abundant P450 form in human liver, possesses great inter-individual differences in enzyme activity, but its polymorphism has not been reported so far. Rebbeck et al found a single base change (A->G) in the 5' flanking region of the CYP3A4 gene in 230 Caucasian men with prostate cancer\(^{(23)}\). However it has been suggested that this region of CYP3A4 is highly conserved, and the genetic variation does not influence the enzyme expression in liver to a significant degree\(^{(24)}\).

Michnovicz et al have studied the inhibitory effects on Estradiol 2-Hydroxylation in women\(^{(25)}\). They found that after 1-month course of cimetidine (800 mg, bid oral), the serum estradiol level got significantly increased, and other bone metabolism related biochemical indexes also changed beneficially. These observations suggest that the CYP1A2 activity may be related to osteoporosis in women. Further studies are needed to define the roles of CYP1A2 and CYP3A4 on the metabolism of estradiol 2-hydroxylation and in the pathogenesis of osteoporosis in postmenopausal women.

SUMMARY

Estradiol mainly secreted from ovary is an important estrogen. Many studies have proved it to be closely related to the development of osteoporosis, breast cancer, and atherosclerosis. Estradiol is synthesized and metabolized by CYP19, 17\(\beta\)-HSD, and some other cytochrome P450s. Genetic variants of these enzyme have been reported in the past literature and may cause estradiol levels to change, and hence cause changes in bone metabolism. The genetic polymorphisms of the enzyme, and their contribution to the development of osteoporosis and other diseases need to be further studied.

REFERENCES


雌二醇生物合成和代谢酶的遗传变异对骨质疏松的影响

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关键词：雌二醇；CYP19；17β-HSD；CYP3A4；CYP1A2；骨质疏松