Genetic Analysis Identifies DDR2 as a Novel Gene Affecting Bone Mineral Density and Osteoporotic Fractures in Chinese Population

Yan Guo1, Tie-Lin Yang1*, Shan-Shan Dong1, Han Yan1, Ruo-Han Hao1, Xiao-Feng Chen1, Jia-Bin Chen1, Qing Tian2, Jian Li2, Hui Shen2, Hong-Wen Deng2*

1 Key Laboratory of Biomedical Information Engineering of Ministry of Education, and Institute of Molecular Genetics, School of Life Science and Technology, Xi’an Jiaotong University, Xi’an, Shaanxi, P. R. China, 2 School of Public Health and Tropical Medicine, Tulane University, New Orleans, Louisiana, United States of America

* yangtielin@mail.xjtu.edu.cn (TLY); hdeng2@tulane.edu (HWD)

Abstract

DDR2 gene, playing an essential role in regulating osteoblast differentiation and chondrocyte maturation, may influence bone mineral density (BMD) and osteoporosis, but the genetic variations actually leading to the association remain to be elucidated. Therefore, the aim of this study was to investigate whether the genetic variants in DDR2 are associated with BMD and fracture risk. This study was performed in three samples from two ethnicities, including 1,300 Chinese Han subjects, 700 Chinese Han subjects (350 with osteoporotic hip fractures and 350 healthy controls) and 2,286 US white subjects. Twenty-eight SNPs in DDR2 were genotyped and tested for associations with hip BMD and fractures. We identified 3 SNPs in DDR2 significantly associated with hip BMD in the Chinese population after multiple testing adjustments, which were rs7521233 ($P = 1.06 \times 10^{-4}$, $\beta = -0.018$ for allele C), rs7553831 ($P = 1.30 \times 10^{-4}$, $\beta = -0.018$ for allele T), and rs6697469 ($P = 1.59 \times 10^{-3}$, $\beta = -0.015$ for allele C), separately. These three SNPs were in high linkage disequilibrium. Haplotype analyses detected two significantly associated haplotypes, including one haplotype in block 2 ($P = 9.54 \times 10^{-4}$, $\beta = -0.016$) where these three SNPs located. SNP rs6697469 was also associated with hip fractures ($P = 0.043$, OR: 1.42) in the Chinese population. The effect on fracture risk was consistent with its association with lower BMD. However, in the white population, we didn’t observe significant associations with hip BMD. eQTL analyses revealed that SNPs associated with BMD also affected DDR2 mRNA expression levels in Chinese. Our findings, together with the prior biological evidence, suggest that DDR2 could be a new candidate for osteoporosis in Chinese population. Our results also reveal an ethnic difference, which highlights the need for further genetic studies in each ethnic group.
Introduction

Osteoporosis is a common disease characterized by low bone mineral density (BMD) and microarchitectural deterioration of bone tissue leading to increased risk of low-trauma osteoporotic fractures. Hip fractures are the most common and severe form of osteoporotic fractures associated with high mortality and tremendous health care costs [1]. The incidence of hip fractures continues to increase rapidly in developing countries [2]. One third of the world’s hip fractures now occur in Asia, mostly in China, and this rate will rise to 52% by 2050 [3]. Clinically, osteoporosis is defined through the measurement of BMD, which is the single best predictor of osteoporotic fractures [4,5].

Genetic factors have important influence on BMD and osteoporotic fractures, with heritability estimates of 0.6–0.8 for BMD [6] and ~0.5 for osteoporotic fractures [7]. However, the genetic variants actually involved remain largely unknown. In recent years, a number of genome-wide association studies (GWASs) on BMD have been successfully performed and more than 60 genome-wide significant loci have been reported [8]. Nevertheless, all the identified loci so far have modest effects on BMD and account for only a small proportion of the variance in liability to osteoporosis, suggesting that many more associated loci remain to be discovered.

DDR2 gene (discoidin domain receptor 2) is located in chromosome 1q23.3 and encodes a member of the receptor tyrosine kinase (RTK) family that binds collagens and regulates cellular responses to the extracellular matrix [9]. DDR2 is demonstrated to be expressed in osteocytes, osteoblastic cells in subchondral bone, bone lining cells, tibia and cartilage [10,11,12]. In vivo studies provide functional evidence that DDR2 is required for normal bone development. Knockout of DDR2 in mice leads to dwarfism, manifested by shortening of long bones [13,14]. In humans, mutations in DDR2 gene are responsible for spondylometaphyseal dysplasia, which is manifested by short limbs with abnormal calcification [15]. Moreover, in vitro studies have shown that DDR2 plays an essential role in osteoblast differentiation and chondrocyte maturation via a signaling pathway that involves MAP kinases and leads to the activation of the transcription factor RUNX2 [11,16]. Given the biological function of DDR2 involved in bone, it is reasonable to hypothesize that DDR2 may influence BMD and could be a new candidate gene for osteoporosis risk estimation, but the genetic variations actually leading to the association remain to be elucidated. Therefore, the aim of this study was to investigate the role of genetic variations of DDR2 in BMD and fracture risk. Our study was performed in three samples from two ethnicities, including two Chinese Han populations and a US white population, in order to see whether the variants identified are common or ethnic-specific.

Materials and Methods

Subjects

The study was approved by the Research Administration of School of Life Science and Technology at Xi’an Jiaotong University and Institutional Review Board at Creighton University and University of Missouri-Kansas City. Signed informed consent documents were obtained from all study participants before entering the study. The basic characteristics of the study samples are summarized in Table 1, with additional descriptions below.

Chinese samples. We have two Chinese samples, one is BMD sample from a population-based cohort, and the other is fracture sample from a case-control cohort. All the subjects were unrelated northern Chinese Han adults living in the city of Xi’an and its neighboring areas. The BMD sample comprised 1,300 unrelated Chinese Han subjects. The inclusion and exclusion criteria have been detailed in our previous publication [17]. BMD (g/cm²) values at total...
were measured using Hologic 4500W machines (Hologic Inc., Bedford, MA, USA). The coefficient of variation (CV) value of the hip BMD was approximately 1.33%.

The fracture sample consisted of 350 cases with osteoporotic hip fractures and 350 elderly healthy controls. Hip fracture was diagnosed by orthopedic surgeons/radiologists according to radiological reports and X-rays. Healthy control subjects were selected from our established database as a ratio of 1:1 to cases and geography-matched. The inclusion and exclusion criteria for cases and controls have been described in detail in our earlier publication [17].

**Caucasian sample.** The Caucasian sample came from a population-based cohort, which consisted of 2,286 unrelated adults. All of the subjects were US Caucasians of Northern European origin living in Midwestern area. The sample description has been detailed in our previous study [18]. BMD values at hip were measured using the same model Hologic 4500W machines (Hologic Inc., Bedford, MA, USA) under the same strict protocols used in the Chinese sample. The CV value of the hip BMD was approximately 1.87%.

**SNP selection and genotyping**

Genomic DNA was extracted from peripheral blood leukocytes using a commercial isolation kit (Gentra systems, Minneapolis, MN, USA) following the protocol of the kit. Selection of SNPs was done on data of the CHB population (Han Chinese in Beijing, China) provided by the HapMap project (Rel28 PhaseII + III, August10, on NCBI B36 assembly, dbSNP b126). We initially selected 30 SNPs in DDR2 according to the following criteria: (i) minor allele frequency (MAF) > 0.05 in the CHB (Han Chinese in Beijing, China) population; (ii) an average density of 1 SNP per 5 kb; (iii) tagging SNP information.

For the Chinese BMD sample, SNP genotyping was carried out using MALDI-TOF mass spectrometry on a MassARRAY system (Sequenom, Inc., San Diego, CA) with iPLEX assay. Genotype calling was performed in real time with MassARRAY RT software version 3.0.0.4 and analyzed using the MassARRAY Typer software version 3.4 (Sequenom). Genotyping quality control procedures leading to SNP exclusion were call rate < 90%, MAF < 0.05 and \( P < 0.001 \) for deviations from Hardy–Weinberg equilibrium (HWE). The average call rate was 98% and the duplicate concordance rate was 99%. Of the 30 SNPs attempted, 28 were genotyped successfully. The basic characteristics of all the qualified SNPs are summarized in Table 2. For the Chinese fracture sample, SNP genotyping was performed using the Affymetrix Human Mapping 500K array set, which has been finished in our previous experiments [17]. For the Caucasian sample, SNP genotyping was performed using Genome-Wide Human SNP Array 6.0 (Affymetrix, Santa Clara, CA, USA), which has been detailed before [18]. For SNPs

| Trait          | Chinese BMD sample | Chinese fracture sample | Caucasian sample |
|----------------|--------------------|-------------------------|------------------|
|                | Cases              | Controls                |                  |
| Number         | 1,300              | 350                     | 350              | 2,286             |
| Male/Female    | 600/700            | 124/226                 | 173/177          | 558/1,727         |
| Age (years)    | 33.42 (11.32)      | 69.35 (7.41)            | 69.54 (6.09)     | 51.37 (13.76)     |
| Weight (kg)    | 59.63 (10.41)      | 59.15 (10.05)           | 59.61 (10.84)    | 75.27 (17.54)     |
| Height (cm)    | 163.94 (8.11)      | 162.84 (8.31)           | 159.41 (9.20)    | 166.35 (8.47)     |
| Hip BMD (g/cm²)| 0.92 (0.13)        | –                       | –                | 0.97 (0.18)       |

Note: Data are shown as mean (standard deviation, SD).

doi:10.1371/journal.pone.0117102.t001
missing in the Affymetrix 500K or 6.0 arrays, we imputed genotypes using the IMPUTE program [19] to facilitate comparison of associations at the same SNPs. To guarantee the reliability of imputation, all imputed SNPs reached a calling threshold of 0.90, i.e., there was a 90% probability that an imputed genotype is true.

Statistical analyses

Before association analyses, to correct for potential population stratification that may result in spurious association results, we applied principal component analysis implemented in EIGENSTRAT [20] to all available genotypic data in the Chinese fracture and Caucasian samples, retaining the top ten principal components. For the BMD samples, significant parameters ($P < 0.05$) such as age, sex, height and weight were used as covariates to adjust for the raw BMD values. The distribution of the resulting residuals was tested for normality by Kolmogorov-Smirnov test. The residuals were used for association analyses. Linear regression implemented in PLINK [21] was fitted to examine association for each SNP assuming an additive inheritance model. Population haplotypes and their frequencies were inferred using Haplovie

| No. | SNP        | Physical position | Genic position | Allele 1 | Allele 2 | MAF  |
|-----|------------|-------------------|----------------|----------|----------|------|
| 1   | rs4518873  | 162603729         | Intron1        | C        | T        | 0.372|
| 2   | rs6671267  | 162608768         | Intron1        | G        | A        | 0.095|
| 3   | rs10917577 | 162613975         | Intron1        | A        | G        | 0.385|
| 4   | rs10799854 | 162619828         | Intron1        | C        | T        | 0.113|
| 5   | rs3795641  | 162625020         | Exon2          | G        | A        | 0.113|
| 6   | rs10799858 | 162630279         | Intron2        | G        | A        | 0.175|
| 7   | rs12044881 | 162635875         | Intron2        | A        | G        | 0.261|
| 8   | rs4657221  | 162642183         | Intron2        | G        | A        | 0.168|
| 9   | rs7521233  | 162649471         | Intron2        | C        | T        | 0.266|
| 10  | rs4657226  | 162653274         | Intron2        | G        | A        | 0.388|
| 11  | rs6697469  | 162658773         | Intron2        | C        | G        | 0.253|
| 12  | rs7553831  | 162661011         | Intron2        | T        | G        | 0.264|
| 13  | rs10737487 | 162667331         | Intron2        | G        | A        | 0.407|
| 14  | rs3923572  | 162671589         | Intron2        | T        | C        | 0.237|
| 15  | rs10917588 | 162671619         | Intron2        | C        | T        | 0.398|
| 16  | rs4559477  | 162681151         | Intron2        | T        | G        | 0.365|
| 17  | rs16843872 | 162686362         | Intron2        | G        | A        | 0.129|
| 18  | rs1039873  | 162692668         | Intron3        | C        | G        | 0.226|
| 19  | rs2805034  | 162695997         | Intron3        | A        | G        | 0.229|
| 20  | rs1510310  | 162703160         | Intron3        | C        | G        | 0.232|
| 21  | rs2806423  | 162711989         | Intron3        | T        | A        | 0.247|
| 22  | rs2684865  | 162721688         | Intron3        | T        | C        | 0.222|
| 23  | rs2684866  | 162726281         | Intron7        | A        | C        | 0.412|
| 24  | rs10799870 | 162733123         | Intron9        | T        | C        | 0.173|
| 25  | rs2298258  | 162737116         | Exon11         | G        | C        | 0.389|
| 26  | rs3738807  | 162741794         | Intron12       | T        | C        | 0.227|
| 27  | rs1780007  | 162748025         | Intron16       | A        | C        | 0.420|
| 28  | rs1779999  | 162753092         | 3'UTR          | C        | T        | 0.418|

Note: MAF, minor allele frequency for allele A1 in the Chinese BMD sample.

doi:10.1371/journal.pone.0117102.t002

DDR2 Gene for BMD and Fractures

PLOS ONE | DOI:10.1371/journal.pone.0117102 February 6, 2015
software [22]. Haplotypes with estimated frequencies greater than 5% were included for association analyses by PLINK [21]. A raw \( P \) value of < 0.05 was considered nominally significant, which was further subjected to Bonferroni correction to account for multiple comparisons. The significance threshold was set at a \( P \) value of less than 0.0018 for single SNP test (0.05/28 SNPs that were included in the association analyses) and 0.0038 for haplotype analysis (0.05/13 haplotypes that were included in the association analyses). For the validation analyses in the fracture sample, SNPTEST [19] was used to test for associations between genotypes and hip fractures. The covariates included age, sex, height, and weight.

**Expression quantitative trait locus (eQTL) analysis**

We examined associations between SNPs and mRNA expression levels of the \( DDR2 \) gene, to ascertain whether the SNPs associated with BMD affected expression of their transcript. Gene expression information was obtained from human lymphoblastoid cell lines (LCLs) of 210 unrelated individuals from HapMap populations in the NCBI Gene Expression Omnibus [23,24]. The sample included 60 white individuals from the CEU (Utah residents with northern and western European ancestry) and 45 Han Chinese individuals. SNP genotype data were derived from the corresponding HapMap Phase III dataset. The linear regression model implemented in PLINK [21] was used to determine associations between expression levels and genotypes.

**Results**

**Association of SNPs in \( DDR2 \) with BMD in the Chinese sample**

The basic characteristics of the study subjects are provided in Table 1. The single SNP association results are summarized in Table 3. For the Chinese sample, there were 18 SNPs showing nominally significant associations with hip BMD (\( P < 0.05 \)). After Bonferroni correction for multiple testing, 3 SNPs remained significant, which were rs7521233 (\( P = 1.06 \times 10^{-4} \)), rs7553831 (\( P = 1.30 \times 10^{-4} \)), and rs6697469 (\( P = 1.59 \times 10^{-3} \)), separately. Each minor allele of these 3 SNPs was associated with reduced hip BMD values with the effect size (\( \beta \)) of -0.018 (rs7521233-C), -0.018 (rs7553831-T), and -0.015 (rs6697469-C), respectively. All of these 3 SNPs are located at the intron 2 of \( DDR2 \). We further characterized the linkage disequilibrium (LD) block and haplotypes for \( DDR2 \). Four blocks in high LD were identified, which ranged in size from 11–60 kb (Fig. 1A). In these blocks, a total of 20 haplotypes were identified and 13 of them were with frequencies more than 0.05 (Fig. 1B). Table 4 listed the major association results for common haplotypes within each block. All of the 4 blocks showed nominally significant associations. After multiple testing adjustment, 2 haplotypes remained significant, which were haplotypes “CGCT” in block 2 (\( P = 9.54 \times 10^{-4} \), \( \beta = -0.016 \)) and “CACTACGTAC” in block 4 (\( P = 2.63 \times 10^{-3} \), \( \beta = -0.015 \)). Block 2 included the above 3 significant SNPs (rs7521233, rs7553831, and rs6697469), presenting the consistent results between single SNP and haplotype analyses. As shown in Fig. 1, these 3 SNPs were in high LD with each other (pairwise LD \( r^2 \) and \( D' > 0.9 \)).

**Association of the BMD associated SNPs in \( DDR2 \) with fracture in the Chinese sample**

We also examined associations between fracture and the above 18 SNPs associated with BMD (Table 3). SNP rs6697469 was found to be associated with hip fractures (\( P = 0.043 \)), and the odds ratio (OR) was estimated to be 1.42 (95% confidence interval (CI): 1.04–1.95). Subjects with the heterozygote genotype CG had an increased risk of fracture, in comparison with those bearing GG genotype. Besides rs6697469, there were 2 additional SNPs showed marginally
significant association signals with hip fractures, which were rs10917588 (\(P = 0.050\)) and rs7553831 (\(P = 0.058\)). These 2 SNPs also associated with increased risk of fractures, and the ORs were estimated to be 1.42 (95% CI: 1.02–1.97, genotypes CT versus TT at rs10917588), and 1.33 (95% CI: 0.97–1.81, genotypes TG versus GG at rs7553831), respectively. The effects on fracture risk for these 3 SNPs were totally consistent with their associations with lower BMD values.

Replication in the white samples

We further checked the association results for the above 18 SNPs in our Caucasian sample (Table 3). However, we didn’t observe significant association signals between the above SNPs and hip BMD.

eQTL analysis results

To investigate the functional relevance of the identified SNPs associated with BMD in DDR2, we performed eQTL analysis in Chinese and white samples from HapMap data, respectively. Three SNPs were significantly associated with DDR2 mRNA expression levels in LCLs of 45 Chinese individuals (Table 3). However, no significant association was found in 60 white individuals (data not shown).
Previous studies have established the important role of DDR2 in regulating osteoblast differentiation and chondrocyte maturation [11,16]. Deletions of DDR2 in mice and humans have been found to be associated with dwarfism and short limbs [13,14,15]. Motivated by the functional role of DDR2 involved in bone, we first performed an association study to investigate whether the common variants in DDR2 influence BMD and fracture risk. We identified a cluster of SNPs in DDR2 significantly associated with hip BMD and fractures in Chinese populations. The effects of DDR2 on BMD and fracture were totally consistent. Taking into account of those biological evidence and our statistical findings, we suggest that DDR2 might have potential role in BMD regulation and fracture pathogenesis.
To further evaluate the functional role of the identified SNPs in DDR2, we exploited eQTL analysis in human LCLs. Generally, analysis of cis-eQTL in tissue type relevant to phenotype is more informative than that in unrelated tissues. However, disease-related human tissues with large sample size are usually difficult to obtain for research purposes. Recent studies have shown that cis-eQTLs are conserved across tissues, and that tissue specific genetic variations comprise only a small proportion of gene expression diversity [25,26,27]. As for bone tissue, there is a large overlap in the transcriptomic effects of genetic variation between human osteoblasts and LCLs [28,29], suggesting that if genes are expressed across tissues, their allele-specific expression can be preserved and highly correlated across tissues. Therefore, we examined cis-eQTL associations to ascertain whether the SNPs associated with BMD affected expression of DDR2 in human LCLs using public HapMap data. We found that the SNPs associated with BMD also affected the expression of DDR2 in the Chinese individuals, but not in the white individuals, suggesting that the eQTLs we identified might be ethnic specific. Additionally, although the top 3 significant SNPs associated with BMD were not the significant eQTLs, another SNP, rs4657226, which was in high LD with these 3 SNPs and belonged to one block, was significantly associated with DDR2 expression levels. The sample size of HapMap data is relatively small. Limited power could influence the detection of additional associations. Nevertheless, our results confirm the relevance of the association data and provide supplementary insights that are indirectly informative for osteoporosis.

Most of BMD associated loci are reported in white populations. Genes predisposing to the risk of osteoporosis may vary between white and Asian populations [30]. Our study found significant association for DDR2 with hip BMD and fractures in Chinese populations. However, the significant associations were not replicated in our Caucasian population. Such results implied an ethnic differentiation. The minor allele of SNP rs6697469 was much common in Chinese than Caucasians, which may contribute to the overall effect and in part explain the ethnic difference. Conversely rs7521233 and rs7553831 showed similar MAF in both ethnic groups. The LD patterns for these 3 SNPs were differ between Chinese (pairwise LD $r^2 > 0.9$) and Caucasian ($r^2 < 0.6$) samples, which could also explain the different results partly. Nevertheless,
our findings support that it is interesting and valuable to conduct genetic susceptibility studies in Asian populations, which could expand our knowledge of the genetic basis of osteoporosis.

We estimated the statistical power of our study using the Genetic Power Calculator program (http://pngu.mgh.harvard.edu/~purcell/gpc/qtlassoc.html). The conservative significance level was set at $P = 0.0018$. Assuming that a marker has a MAF of 0.05 and is in strong LD ($D' = 0.98$) with a functional variant that accounts for 1.0% variation of a phenotype, our Chinese BMD sample can achieve $>65\%$ statistical power. For the 3 significant SNPs we identified, the proportion of BMD variation explained by each SNP was 1.15% (rs7521233), 1.15% (rs7553831), and 0.81% (rs6697469), respectively. We acknowledge that our study is not powerful to detect genetic variants with low effect size.

In summary, our study provides novel evidence that DDR2 is associated with hip BMD and fractures in Chinese populations. Considering the biological function of DDR2, we suggest that DDR2 could be a new candidate in determining the risk of osteoporosis and fractures. Our results also reveal some extent of ethnic difference for DDR2, which highlight the need for further genetic studies in each ethnic group. Our findings further advance our understanding of the genetic architecture of osteoporosis. Whether DDR2 could be exploited as a novel therapeutic agent for osteoporosis warrants further investigation.

Author Contributions

Conceived and designed the experiments: YG TLY. Performed the experiments: HY RHH XFC JBC. Analyzed the data: TLY SSD. Contributed reagents/materials/analysis tools: QT JL HS HWD. Wrote the paper: YG.

References

1. Cooper C, Campion G, Melton LJ 3rd (1992) Hip fractures in the elderly: a world-wide projection. Osteoporos Int 2: 285–289. PMID: 1421796
2. Lau EM, Lee JK, Suriwongpaisal P, Saw SM, Das De S, et al. (2001) The incidence of hip fracture in four Asian countries: the Asian Osteoporosis Study (AOS). Osteoporos Int 12: 239–243. PMID: 11315243
3. Gullberg B, Johnell O, Kanis JA (1997) World-wide projections for hip fracture. Osteoporos Int 7: 407–413. PMID: 9425497
4. Johnell O, Kanis JA, Oden A, Johansson H, De Laet C, et al. (2005) Predictive value of BMD for hip and other fractures. J Bone Miner Res 20: 1185–1194. PMID: 15940371
5. Kanis JA, Oden A, Johnell O, Johansson H, De Laet C, et al. (2007) The use of clinical risk factors enhances the performance of BMD in the prediction of hip and osteoporotic fractures in men and women. Osteoporos Int 18: 1033–1046. PMID: 17323110
6. Peacock M, Turner CH, Econs MJ, Foroud T (2002) Genetics of osteoporosis. Endocr Rev 23: 303–326. PMID: 12050122
7. Michaelsson K, Melhus H, Ferm H, Ahlbom A, Pedersen NL (2005) Genetic liability to fractures in the elderly. Arch Intern Med 165: 1825–1830. PMID: 16157825
8. Richards JB, Zheng HF, Spector TD (2012) Genetics of osteoporosis from genome-wide association studies: advances and challenges. Nat Rev Genet 13: 576–586. doi: 10.1038/nrg3228 PMID: 22805710
9. Vogel W, Gish GD, Alves F, Pawson T (1997) The discoidin domain receptor tyrosine kinases are activated by collagen. Mol Cell 1: 13–23. PMID: 9659899
10. Karn T, Holtzch U, Brauning A, Bohme B, Wolf G, et al. (1993) Structure, expression and chromosomal mapping of TKT from man and mouse: a new subclass of receptor tyrosine kinases with a factor VIII-like domain. Oncogene 8: 3433–3440. PMID: 8247548
11. Lin KL, Chou CH, Hsieh SC, Hwa SY, Lee MT, et al. (2010) Transcriptional upregulation of DDR2 by ATF4 facilitates osteoblastic differentiation through p38 MAPK-mediated Runx2 activation. J Bone Miner Res 25: 2489–2503. doi: 10.1002/jbmr.199 PMID: 20564243
12. Xu L, Peng H, Glasson S, Lee PL, Hu K, et al. (2007) Increased expression of the collagen receptor discoïd domain receptor 2 in articular cartilage as a key event in the pathogenesis of osteoarthritis. Arthritis Rheum 56: 2663–2673. PMID: 17665456
13. Labrador JP, Azcoitia V, Tuckermann J, Lin C, Olaso E, et al. (2001) The collagen receptor DDR2 regulates proliferation and its elimination leads to dwarfism. EMBO Rep 2: 446–452. PMID: 11375938
14. Kano K, Marin de Evsikova C, Young J, Wnek C, Maddatu TP, et al. (2008) A novel dwarfism with gonadal dysfunction due to loss-of-function allele of the collagen receptor gene, Ddr2, in the mouse. Mol Endocrinol 22: 1866–1880. doi: 10.1210/me.2007-0310 PMID: 18483174
15. Bargal R, Cormier-Daire V, Ben-Neriah Z, Le Merrer M, Sosna J, et al. (2009) Mutations in DDR2 gene cause SMED with short limbs and abnormal calcifications. Am J Hum Genet 84: 80–84. doi: 10.1016/j.ajhg.2008.12.004 PMID: 1910212
16. Zhang Y, Su J, Yu J, Bu X, Ren T, et al. (2011) An essential role of discoïd domain receptor 2 (DDR2) in osteoblast differentiation and chondrocyte maturation via modulation of Runx2 activation. J Bone Miner Res 26: 604–617. doi: 10.1002/jbmr.225 PMID: 20734453
17. Guo Y, Tan LJ, Lei SF, Yang TL, Chen XD, et al. (2010) Genome-wide association study identifies ALDH7A1 as a novel susceptibility gene for osteoporosis. PLoS Genet 6: e1000806. doi: 10.1371/journal.pgen.1000806 PMID: 20072603
18. Yang TL, Guo Y, Liu YJ, Shen H, Liu YZ, et al. (2012) Genetic variants in the SOX6 gene are associated with bone mineral density in both Caucasian and Chinese populations. Osteoporos Int 23: 781–787. doi: 10.1007/s00198-011-1626-x PMID: 21625884
19. Price AL, Patterson NJ, Plenge RM, Weinblatt ME, Shadick NA, et al. (2006) Principal components analysis corrects for stratification in genome-wide association studies. Nat Genet 38: 904–909. PMID: 17572673
20. Stranger BE, Nica AC, Forrest MS, Dimas A, Bird CP, et al. (2007) Population genomics of human gene expression. Nat Genet 39: 1217–1224. PMID: 17873874
21. Purcell S, Neale B, Todd-Brown K, Thomas L, Ferreira MA, et al. (2007) PLINK: a tool set for whole-genome association and population-based linkage analyses. Am J Hum Genet 81: 559–575. PMID: 17701901
22. Barret JC, Fry B, Maller J, Daly MJ (2005) Haploview: analysis and visualization of LD and haplotype maps. Bioinformatics 21: 263–265. PMID: 15297300
23. Stranger BE, Forrest MS, Dunning M, Ingle CE, Beazley C, et al. (2007) Relative impact of nucleotide and copy number variation on gene expression phenotypes. Science 315: 848–853. PMID: 17289997
24. Stranger BE, Nica AC, Forrest MS, Dimas A, Bird CP, et al. (2007) Population genomics of human gene expression. Nat Genet 39: 1217–1224. PMID: 17873874
25. Cookson W, Liang L, Abecasis G, Moffatt M, Lathrop M (2009) Mapping complex disease traits with global gene expression. Nat Rev Genet 10: 184–194. doi: 10.1038/nrg2537 PMID: 19223927
26. Zhang K, Li JB, Gao Y, Egli D, Xie B, et al. (2009) Digital RNA allelotyping reveals tissue-specific and allele-specific gene expression in human. Nat Methods 6: 613–618. doi: 10.1038/nmeth.1357 PMID: 19620972
27. Lee JH, Park IH, Gao Y, Li JB, Li Z, et al. (2009) A robust approach to identifying tissue-specific gene expression regulatory variants using personalized human induced pluripotent stem cells. PLoS Genet 5: e1000718. doi: 10.1371/journal.pgen.1000718 PMID: 19911041
28. Kwan T, Grundberg E, Koka V, Ge B, Lam KC, et al. (2009) Tissue effect on genetic control of transcript isoform variation. PLoS Genet 5: e1000608. doi: 10.1371/journal.pgen.1000608 PMID: 19880542
29. Hsu YH, Zillikens MC, Wilson SG, Farber CR, Demissie S, et al. (2010) An integration of genome-wide association study and gene expression profiling to prioritize the discovery of novel susceptibility Loci for osteoporosis-related traits. PLoS Genet 6: e1000977. doi: 10.1371/journal.pgen.1000977 PMID: 20548944
30. Dvornyk V, Liu XH, Shen H, Lei SF, Zhao LJ, et al. (2003) Differentiation of Caucasians and Chinese at bone mass candidate genes: implication for ethnic difference of bone mass. Ann Hum Genet 67: 216–227. PMID: 12914574