THE GENETICS OF OSTEOPOROSIS

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Summary

Osteoporosis is highly influenced by genetic factors. Bone mineral density (BMD) has also been shown to be highly heritable, as are other risk factors for osteoporotic fracture such as bone quality, femoral neck geometry and bone turnover. Susceptibility to osteoporosis is mediated, in all likelihood, by multiple genes each having small effect and a number of different approaches are being employed to identify the genes involved. Study methods include linkage studies in both humans and experimental animals as well as candidate gene and gene expression studies. Linkage studies have identified multiple quantitative trait loci (QTL) for regulation of BMD and, along with twin studies, have indicated that the effects of these loci on BMD are site-dependent and sex-specific. On the whole, the genes responsible for BMD regulation at these QTL have not been identified. Most studies have used the candidate gene approach, based on what is known of bone metabolism. The vitamin D receptor gene (VDR), the collagen type I alpha I gene (COLIA1) and estrogen receptor gene (ER) alpha have been widely investigated and found to play a role in regulating BMD. A recent meta-analysis suggests, however, that VDR plays no significant role, and the effects of the other 2 genes are modest – probably accounting for less than 3% of the genetic contribution to BMD. Cost-effective large scale genetic testing is becoming available and lends itself to combining large multi-national populations for candidate gene analysis, meta-analyses, DNA pooling studies and gene expression studies.

Keywords: Osteoporosis; Gene; QTL; Bone Mineral Density,

Resumo

A osteoporose é fortemente influenciada por factores genéticos. Do mesmo modo, a densidade mineral óssea (DMO) e outros factores de risco para fracturas, como a baixa qualidade óssea, a geometria do colo do fémur e o turnover ósseo são grandemente hereditários. A susceptibilidade para a osteoporose é, com grande probabilidade, mediada por múltiplos genes, com um pequeno contributo cada. Várias abordagens têm sido utilizadas para identificar os genes envolvidos, incluindo estudos de *linkage* em humanos e em modelos animais, estudo de genes candidatos e de expressão de genes. Os estudos de *linkage* identificaram múltiplos *quantitative trait loci* (QTL) para a regulação da DMO e, juntamente com estudos em gémeos, indicaram que o efeito destes *loci* na DMO é dependente do local e específica para o sexo. No global, os genes responsáveis pela regulação da DMO nestes QTL não foram identificados. A maioria dos estudos utilizou genes candidatos, baseando-se no metabolismo ósseo. O gene do receptor da vitamina D (RVD), do colagénio tipo I alfa1 (COLIA1) e do receptor dos esterogénios (RE) alfa têm sido amplamente investigados e concluiu-se que têm um papel na regulação da DMO. Uma meta-análise recente sugere contudo que o RVD não desempenha um papel relevante e que o efeito dos outros 2 genes é modesto – provavelmente contribuindo em menos de 3% para a componente genética da DMO. Os testes genéticos em larga escala mais acessíveis permitirão grandes estudos populacionais multinacionais de análise de genes candidatos, meta-análises, *pooling* de DNA e estudos de expressão de genes.

Palavras-chave: Osteoporose; Gene; QTL; Densidade Mineral Óssea

Osteoporosis Genes and their Identification

Osteoporosis is characterized by diminished bone mineral density and deterioration in bone microarchitecture. The main clinical endpoint is fracture. It is common and costly, both financially and in

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social terms. Genetic factors have long been recognised to play an important role in both osteoporosis and its associated phenotypes, including bone mineral density (BMD), bone mass, broadband ultrasound attenuation (BUA), velocity of sound (VOS). Twin and family studies have estimated that 50-85% of the variance in bone mass is genetically determined.[1-4] Similar studies have shown evidence of significant genetic effects on other determinants of fracture risk, including quantitative ultrasound,[5] several aspects of femoral neck geometry,[5] muscle strength,[6] bone turnover markers,[7;8] body mass index[9] and age at menopause.[10]

Unfortunately, there are few data describing the heritability of osteoporotic fracture, mainly because recruiting adequate numbers of study subjects with fracture is difficult and expensive. Several studies have shown that a family history of fracture is a risk factor for fracture, and importantly, this is independent of BMD.[11-14] Two studies (one of sib-pairs, one of twins) have shown wrist fracture to be clearly heritable[15;16] and suggest that the genes involved may be separate to those influencing BMD.[17] This illustrates the important difference between associated phenotypes, osteoporosis and fracture: associated phenotypes have been found to be highly heritable but finding the genes responsible for them does not necessarily identify genes causative for other phenotypes or fracture. Another such example is that of genes influencing bone density and speed of sound in bone. A UK twin study reported both wrist ultrasound and BMD to be heritable. However only a modest genetic overlap was found between genes influencing BMD/VOS properties of bone and genes influencing fracture.[18]

Several approaches are being employed currently in the search for genes which contribute to osteoporosis in the general population (reviewed by Huang *et al.*).[19] Rare monogenic conditions affecting bone have already been used to cast light on genes which may influence population osteoporosis. Going forward, the most important approaches include association studies and, to a lesser extent, linkage studies.

Methods of Identifying Genes in Osteoporosis

Linkage Studies

Linkage disequilibrium (LD) refers to the phenomenon whereby genes lying close together tend to be inherited together. Evidence suggests that LD varies greatly and variably according to both chromosomal region and human population studied, but can extend to 350kb or further.[20] Linkage studies are a well validated method for the identification of genes responsible for monogenic diseases and have been applied to the identification of chromosomal regions which harbour genes regulating quantitative traits such as bone mass, in so-called quantitative trait loci (QTL). An advantage of linkage-based studies is that they offer the prospect of identifying new molecular pathways that regulate bone metabolism. In addition they are not influenced by population admixture. A major disadvantage is their low statistical power to detect genes having modest effects and so they require family samples of considerable size (several thousand). An independent validation group is now recognised as providing important confirmatory replication data – many previous findings have not been replicated. This type of study is waning in popularity as the genome-wide association scans become available (see below).

Linkage studies in Animals

Linkage studies in experimental animals have also been used in the identification of genes responsible for complex traits. This approach has several advantages: optimal control over the animals' environment minimises the influence of confounding factors; and large numbers of progeny may be generated,providing excellent statistical power. In addition, fine mapping of loci identified may be achieved using a technique known as 'back crossing'. The most obvious drawback of the approach is that genes/loci regulating BMD in mice may not be influential in regulating BMD in man.

A recent study has combined genetic and genomic approaches in mice to provide evidence of a role for the *Alox15* gene. Earlier work had identified a region on mouse chromosome 11 as influencing peak BMD.[21] A congenic mouse model was then constructed using the area of interest on chromosome 11 and shown to have increased BMD.[22] Microarray analysis identified *Alox15* as the differentially expressed gene which encodes 12/15 lipoxygenase (12/15-LO), and other studies confirmed that this overexpression had biological impact (increased expression of CD36 and reduced osteocalcin). A 12/15-LO knock out mouse model also confirmed the findings, as did pharmacological inhibitors of 15-lipoxygenase.[22] Work in humans

has also shown linkage to a region on human chromosome 17 containing the genes for 12 and 15 lipoxygenase, suggesting that the findings in mice may be of direct relevance to human BMD regulation[23] and further evidence for both forms of lipoxygenase is emerging, if somewhat conflicting.[24;25]

Linkage Studies in Humans

Linkage studies in sib-pairs and extended families having osteoporosis have also been used to identify loci linked to BMD. Early studies identified loci on chromosomes 1p36, 2p23-p24 and 4q32-34,[23] with subsequent work in a second sample confirming linkage to the 1p36 locus.[26] A genome wide search in a Chinese sample for loci regulating forearm BMD[27] also revealed the highest LOD score at 2p23-24. Koller and co-workers conducted a whole genome search in a series of 595 healthy Caucasian and African-American female sib-pairs, finding LOD = +3.86 at chromosome 1q21-23[28] and an area suggestive of linkage at 5q33-35. Linkage studies in the same population identified multiple loci for regulation of femoral neck geometry on chromosome 5q and 4q and 17q.[29] Karasik and colleagues[30] have reported a genome scan on 330 families (Framingham study) and identified QTL suggestive of linkage on chromosome 6 and 20. Of interest, a subsequent analysis using the same population suggested that QTL regulation of BMD differs between men and women, and different QTL were found for the phenotypes peak bone mass and bone loss.[31] Wilson *et al.* have performed one of the largest linkage studies with 1100 dizygous UK twin pairs, defining two regions of suggestive linkage on chromosomes 1p36 and 3p21. Linkage to the 3p21 region was confirmed in a validation sample of 254 extreme discordant or concordant affected sib pairs having low BMD.[32]

Most linkage studies have examined BMD as the associated phenotype of interest. However in a study of Icelandic families, Styrkasdottir and colleagues detected significant linkage of osteoporosis to chromosome 20p12 (LOD = 5.1) using a novel classification system.[33] In this study, subjects were scored as "affected" if they had reduced BMD (Z-score less than –1.0 at spine or hip) or if they had a history of fragility fractures, or if they were undergoing bisphosphonate treatment for osteoporosis. The Icelandic study also suggested linkage of spine and hip BMD to chromosome 20p12, with LOD scores of around +3.0 on the genome wide scan and LOD scores of between +3.4 and +4.0 on fine mapping. Further analysis showed that part of the linkage signal was due to an association between osteoporosis and a polymorphism in the BMP2 gene which results in a serine-alanine amino acid change at codon 37, but of note this was not replicated in the large Rotterdam cohort.[34]

Associated osteoporosis risk phenotypes other than BMD have also been examined. Using ultrasound to generate two associated phenotypes, BUA and VOS, Wilson *et al.* have performed a genome-wide screen of dizygous twin pairs using 737 highly polymorphic microsatellite markers. Evidence was found of linkage to chromosome 2q33-37 (BUA, LOD 2.1-5.1) and 4q12-21 (VOS, LOD2.2-3.4). LOD scores >2 were also identified on chromosomes 1,2,13,14 and X.[35] Similar work on the Framingham study sample showed quantitative ultrasound to be linked to chromosomal regions 1p36.1.[36] Subsequently, this group has used combined bone phenotypes to see if more information may be obtained. Using BMD and quantitative ultrasound they per-

Table I. Summary of main Quantitative Trait Loci findings for BMD in humans

Author (date)	locus	LOD score	phenotype
Devoto et al. (1998)	1p36	3.51	Hip BMD
	2p23	2.29	Hip BMD
	4q33	2.95	Hip BMD
Nui et al. (1999)	2p21	2.15	Wrist BMD
Koller et al. (2000)	1q21	3.86	Spine BMD
	5q33	2.23	Hip BMD
Karasik et al. (2002)	6p21	2.93	Spine BMD
	21q22	3.14	Hip BMD
Wilson et al. (2003)	3p21	2.7	Spine BMD
	1p36	2.4	Hip BMD
Iceland (2003)	20p12	3.18	Hip BMD
	20p12	2.89	Spine BMD
Ralston et al. (2005)	10q21 men	4.42	Hip BMD
	20q13 women	3.2	Spine BMD
Tang et al. (2007)	a number of shared regions		Body fat mass and BMD

formed principal components analysis: linkage to 1q21.3 and 8q24.3 was found with the first prinicipal component (LODs 2.5, 2.4) while 1p36 was found with the second (LOD 2.1).[37]

Given the polygenic nature of BMD regulation and osteoporosis susceptibility, most linkage studies performed to date have probably been underpowered, although the two large studies mentioned included over 1000 subjects in each.[32;33] Overall, results from different linkage studies show more discrepancy than agreement, probably because of differing study populations and differing criteria for subject enrolment. It is partly for these reasons and easier access to clinical samples that association studies are becoming more widely used.

Candidate Gene Studies

Candidate gene studies have been widely used. Candidate gene association studies are relatively easy to perform and may have sufficient power to detect small allelic effects. They may be disadvantaged, however, by the effects of confounding factors, genetic heterogeneity and population stratification. Furthermore, demonstration of an association between a candidate gene and BMD does not necessarily mean that the gene is causally responsible for the effect observed, as there may be linkage disequilibrium with a nearby causal gene. The transmission desequilibrum test (TDT) can help by testing candidate genes for both association and linkage.

Candidate genes investigated thus far have included genes influencing cytokines and growth factors which regulate bone turnover; genes that encode components of bone matrix; and genes that encode receptors for calciotropic hormones. Individual candidate genes that have been implicated in the regulation of bone mass or osteoporotic fractures have been reviewed elsewhere.[38] Recent advances in knowledge are discussed in more detail below. Candidate genes have been suggested on the basis on what is known about bone metabolism. In future, novel genes may be identified by genome-wide association scans (see below).

Vitamin D Receptor (VDR)

Vitamin D interacts with its receptor to play an important role in calcium homeostasis by regulating bone cell growth and differentiation, intestinal calcium absorption and parathyroid hormone secretion. The VDR was therefore a natural place to begin looking for genetic variation that might account for osteoporosis. The original finding that VDR alleles played a role in BMD is over 10 years old.[39] Other studies of VDR in relation to bone mass have since been conflicting, and it is likely that the VDR genotype is associated with relatively modest effects on bone mass. Various different polymorphisms have been described, in different populations,[40;41] although the mechanisms by which these polymorphisms modulate VDR function remain unclear: some 3' polymorphisms may influence RNA stability, and isoforms of VDR encoded by different alleles may possess different functions.[40] In addition there are data to suggest that an interaction between 5' and 3' polymorphisms is involved in regulating VDR function, and that the risk allele involving such polymorphisms may result in lower levels of VDR mRNA.[42] However, a recent giant study of 26,000 participants could not find a relationship with either BMD or fracture, which seriously calls into question the results of the smaller studies.[43]

Type I collagen

The genes encoding type I collagen (COLIA1 and COLIA2) are important, well studied candidates for the pathogenesis of osteoporosis. A common polymorphism affecting the transcription factor Sp1 binding site has been shown to have increased prevalence in osteoporosis patients.[48] Positive associations between the COLIA1 Sp1 polymorphism and bone mass or osteoporotic fractures were subsequently reported in several populations, and meta-analysis also supported the COLIA1 genotype conferring differences in BMD.[44] Ethnic differences have been reported in population prevalence of COLIA1 Sp1 alleles with the polymorphism being common in Caucasian populations, but rare in Africans and Chinese.[45] Overall the data suggest that the COLIA1 Sp1 polymorphism is a functional variant which has adverse effects on bone composition and mechanical strength. Haplotype analysis has shown that susceptibility to fracture is driven by the Sp1 polymorphism rather than other known polymorphisms at the COLIA1 locus,[46] although it remains possible that hitherto unidentified polymorphisms in linkage disequilibrium with the Sp1 polymorphism exist and contribute to the observed effects. From a clinical viewpoint, the COLIA1 polymorphism may be of value not as a therapeutic target but as a marker of osteoporotic fracture risk, since it predicts fractures indepen-

dent of BMD and interacts with BMD to enhance fracture prediction.[47]

Estrogen Receptors and Aromatase Genes

In view of the strong relationship between estrogen deficiency and bone loss, the estrogen receptor alpha (ER) gene has long been a strong candidate gene for osteoporosis. An association has been reported between a TA repeat polymorphism in the ER promoter and bone mass in both Japanese and U.S. populations. Other investigators have reported positive associations between haplotypes defined by *PvuII* and *XbaI* polymorphisms in intron 1 of the ER gene and bone mass[48] as well as age at menopause.[49] The molecular mechanisms by which these polymorphisms influence bone mass are as yet unclear, but a meta-analysis of the intron 1 polymorphisms indicated that the association with BMD and fracture is attributable mainly to variation at the *XbaI* site.[50] More recently, a large-scale study comprising 8 European centres has attempted to answer the question more definitively using almost 19,000 subjects. Three common ER gene polymorphisms were studied and none of the polymorphisms was shown to be associated with BMD. The absence of a Xba1 polymorphism recognition site conferred a risk reduction in all fractures of 19% while the risk reduction for vertebral fractures was 35%. The effects on fracture were independent of BMD (but may be associated with BUA).[51] Polymorphisms in PvuII and TA repeats did not appear to have any influence.[52]

Aromatase is the enzyme which converts androgens into estrogens so is likely to be of importance in bone metabolism in men and post-menopausal women. It is encoded by the CYP19 gene. A recent study from Australia has shown the TTTA repeat polymorphism of CYP19 to be associated with higher circulating estradiol, higher BMD at hip and lumbar spine and lower markers of bone turnover, in over 1200 women age 70 years or older.[53] Similar findings have also been reported in elderly Italian men.[54]

Other Genes

Polymorphisms in several other candidate genes have been associated with bone mass and/or osteoporotic fracture including TGFb-1 and the IL-6 locus. The effects of these polymorphisms on IL-6 function are yet to be determined. Two studies have looked at the possible associations between apolipoprotein E (APOE) alleles and osteoporosis but again the mechanisms by which APOE alleles influence susceptibility to osteoporosis remain unclear. Two groups have reported an association between a coding polymorphism of the calcitonin receptor gene and BMD. The osteocalcin gene has been found to be associated and linked to BMD and bone quality.[55] Other candidate genes which have been studied in relation to BMD include; parathyroid hormone; the androgen receptor, aromatase, osteoprotegerin, Klotho and the interleukin-1 receptor antagonist (IL-1ra). Most of the original findings, however, have not been consistently replicated.

In addition to the study of single genes or polymorphisms in isolation, it has been realized that both gene-gene and gene-environment interactions play an important role in influencing the variation of expression of complex traits such as osteoporosis within populations. Such interactions are discussed below.

Gene-Gene and Gene-Environment Interactions

A Dutch study of 1000 postmenopausal women looked at the effects of a combination of both the G to A polymorphism in the COL1A1 Sp1 binding site and the 'baT' haplotype of VDR. They found that there was a significant interaction between the genotypes, both being independent of the effect of BMD.[56] The Danish Osteoporosis Prevention Study has recently reported the influence of polymorphisms within the CYP19 and androgen receptor genes in almost 1800 newly postmenopausal women who were randomized to receive estrogen replacement therapy or no treatment.[57] While perimenopausal bone loss was not associated with either genes' polymorphisms, the BMD response over 5 years to estrogen was influenced by genotype: one CYP19 allele was associated with significantly greater response. While the androgen receptor genotype was not related to BMD, a modifying effect of sex hormone-binding globulin (SHBG) was observed. Thus in the highest quartile of SHBG, androgen receptor genotype was associated with baseline BMD.

These types of study emphasize the importance of both gene-gene and gene-environment interactions and highlight once again the need for large, usually multi-center, studies to recruit sufficient subjects to enable well-powered studies to be performed. They also create new difficulties of their own, particularly problems associated with multiple testing of subgroups – which increase the likelihood of spurious positive findings unless they

are taken into account in the analysis.

Gene Expression Studies

A novel approach to the question of osteoporosis genes is that involving gene expression studies. In this type of study, differences in gene expression are explored in tissues derived from subjects expressing and not expressing the trait of interest. Very much greater power is obtained if the genetic background of the trait-discordant subjects is similar or the same, as in the case of identical twins. One such small study has used osteoblast-like culture from 2 pairs of monozygotic twins discordant for BMD and one concordant pair. Genome-wide gene expression of the cell culture derived from bone marrow aspirates suggests the following genes were differentially expressed: chondroitin beta 1,4 N-acetylgalactosaminyltransferase, inhibin beta A, interleukin 1 beta and colony stimulating factor 1 macrophage.[58] These genes are known to play a part in bone physiology. Although the numbers studied were small this study highlights both the potential of the emerging new technology for examining gene expression and the further benefits that may be derived from the twin registers around the world in providing informative willing subjects for intensive study .

Pooling studies

Another newer method being used for increasing the power and cost-effectiveness of studies to detect genes associated with osteoporosis is that of pooling. This type of association study contrasts DNA pools from 200-300 subjects with and without the trait of interest, for example BMD. One such study has used 25,000 SNPs in 16,000 genes from women divided into study groups by expression of the traits high and low BMD. Because of the loss of power with multiple testing, the findings were verified by individual genotyping in two further case control groups. The differences in allele frequency between the two trait expression groups suggested a candidate locus in the phosphodiesterase 4D (PDE4D) gene on chromosome 5q12. This was fine mapped using 80 SNPs within 50 kB of the marker SNP.[59] This study also produced evidence in support of the association with the Ser37Val polymorphism in *BMP2,* a gene known to interact with PDE4D (and implicated in Icelandic studies). These data illustrate the potential of these methods but also highlight the need for several replication groups.

Overlapping phenotypes

In addition to the associated phenotypes and traits which may be used as surrogates of the main clinical outcome of interest, other bone diseases may also shed light on genes of importance in osteoporosis. Studies have shown that perhaps 30% of genes involved in bone metabolism overlap with those influencing osteoarthritis – a disease of bone as well as cartilage. Genes believed to be common to both include the VDR, the COLIA1 and possibly the ER genes.[60] A recent example of an association study of OA progression by Valdes et al implicated several bone genes such as BMP-2[61] and genes involved in inflammation and cytokines have been found, somewhat surprisingly, to be associated with chronic diseases such as disc degeneration.[62] With the finding that the LRP-5 gene is associated with osteoporosis comes the realization that genes controlling pathways such as lipid metabolism and inflammation may be important in what were considered non-inflammatory bone conditions. Thus the choice of potential candidate genes is getting considerably larger and genetic researchers have increasingly to cross the traditional disease boundaries.

In addition to the overlap with osteoarthritis, osteoporosis is also associated with obesity. The tools are now available to us to dissect this relationship using a variety of methods. It would appear, however, that the negative association with body mass index is not independent of the loading expected on bone,[63] thus advising people to gain weight will not be appropriate. It is likely that shared genes operate to account for this association.[64]

Genes of Rare Monogenic Diseases

Osteoporosis and fragility fractures are features of several rare monogenic diseases, and provide an obvious place to start the search for genes influencing osteoporosis in the general population. Such conditions are not always informative, however. They include osteogenesis imperfecta (OI), the osteoporosis-pseudoglioma syndrome (OPS) and syndromes associated with inactivating mutations of the oestrogen receptor alpha and aromatase genes.

OI describes a heterogeneous group of monogenic disorders characterised by multiple bone fractures which, in most forms, is caused by mu-

tations in the type I collagen genes COLIA1 and COLIA2. The genes which encode type I collagen possess mutations in many different places, accounting for the heterogenous nature of the disorder – from mild to extremely severe. OPS is a rare, autosomal recessive disorder characterised by juvenile onset osteoporosis and blindness due to persistent vascularisation of the eye. Initial linkage studies mapped OPS to chromosome 11q12--13.[65] Subsequent work showed the disease to be caused by inactivating mutations in the low density lipoprotein-related receptor-5 (LRP-5).[66] Another phenotype, autosomal dominant high-bone-mass, maps to the same region[67] and independently was reported to be caused by an activating mutation of the same receptor.[68] Osteoporosis has been reported in association with homozygous inactivating mutations of the estrogen receptor and aromatase genes, emphasising the importance of estrogen in the attainment and maintenance of peak bone mass. Mutations in the latency-activating peptide (LAP) domain of the TGF beta 1 gene are associated with Camurati-Engelmann disease – a condition characterised by increased BMD in the diaphysis of long bones.[69] Mutations of the TCIRG1 gene, which encodes a subunit of the osteoclast proton pump, have been shown responsible for the autosomal recessive condition osteopetrosis.[70]

The important question is do the genetic clues obtained from rare diseases cast any light on the osteoporosis and fractures seen in the normal population? There is evidence that some of these genes do contribute to regulation of 'normal' BMD. For example, LRP gene polymorphisms have recently been shown to be associated with bone mineral content, bone area and stature particularly in males.[71;72] Several groups have reported polymorphisms in the TGF beta gene to be associated with BMD and osteoporotic fracture[14;73] and polymorphisms of the TCIRG1 genes (subunit of osteoclast proton pump) have been found to be associated with BMD in normal subjects.[74] Finally, the SOST gene causative in the sclerosteosis/van Buchem disease phenotype has been shown to be associated with BMD in elderly Dutch white volunteers.[75]

Genome wide association scans

At present, some argue, lines of investigation are driven by technology and the availability of new assay techniques handling ever larger numbers of polymorphisms. Although the estimated number of human genes continues to fall (currently around 23,000) the number of recognized SNPs increases – with over 30,000 known non-synonymous SNPs and the possibilty of testing samples with over 250,000 validated SNPs at a cost of less than 1 US cent per SNP. The use of genome wide scans is already beginning to yield exciting new genes in other complex genetic traits, such as diabetes.[76] The new technology will enable increasingly large panels of polymorphisms, as well as gene expression levels and, eventually, proteins and metabolic profiles to be studied simultaneously. Funding for future work should be prioritised for those study proposals demonstrating sufficient power to answer the question being addressed, although the increasing problem of multiple testing and the difficulties in having large numbers of replicate clinical cohorts will make the task no less challenging.

In conclusion, osteoporosis is a perfect example of a complex genetic trait. The associated phenotypes studied thus far have heritabilities of 50--80% and a large number of genes are likely to be involved in its pathogenesis. Several candidate genes have been identified but their individual effects are small. Many genome-wide linkage scans have been performed but the results are inconsistent - underlining some of the difficulties in pinpointing the genes - and suggest that to maximise the chances of gene discovery a full range of phenotypes and methods will need to be utilised. Regardless of the methods employed, combining datasets will be essential to obtain sufficient power. This means national and international collaboration will play a vital role in taking forward the work done so far.

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