CHAPTER 1
Epidemiology and Genetics of Postmenopausal Osteoporosis

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Introduction

Osteoporosis is a skeletal disease characterized by low bone mass and micro-architectural deterioration of bone tissue with a consequent increase in bone fragility and susceptibility to fracture [1]. The term osteoporosis literally means “porous bone” and refers to a condition in which bone is normally mineralized but reduced in quantity. In 1994, a working group of the World Health Organization (WHO) provided a practical definition of osteoporosis as a bone mineral density (BMD) of greater than 2.5 SD below the young normal mean [2]. Earlier definitions had incorporated fracture and so to provide comparability, the subset of women with osteoporosis who had also suffered one or more fragility fractures were deemed to have severe “established” osteoporosis.

The etiology of osteoporotic fractures is complex. Low bone density is not the only risk factor for fracture and there has been a move towards making an assessment of individualized 10-year absolute fracture risk using the WHO FRAX based on multiple clinical risk factors [3]. Family history, and in particular parental hip fracture, is included in the FRAX tool reflecting the hereditary component of the condition. There is growing recognition of a complex interaction between genetic and environmental factors. Only a small number of specific genes contributing to osteoporosis risk have been consistently identified; however, the investigation of gene-environment interactions with developmental plasticity has yielded promising results, raising the possibility of intervening during fetal development or early life to reduce individual fracture risk and the global burden of this disease. It is estimated that around 200 million women worldwide have osteoporosis with an osteoporotic fracture occurring every 3 seconds [4]. This equates to 1 in 3 women over 50 years of age.
Chapter 1

suffering an osteoporotic fracture [5,6]. Fragility fractures make up 0.83% of the worldwide burden of noncommunicable disease. This figure rises to 1.75% in Europe, where fragility fractures also account for more disability adjusted life years (DALYs) than many other chronic diseases [7]. At present the annual cost of all osteoporotic fractures worldwide is in excess of $17 billion and is expected to rise to $25 billion by 2025 [8]. The cost of treating osteoporotic fractures is also increasing in the UK and expected to rise to over £2 billion by 2020 [9]. This chapter will review the genetic and early environmental factors associated with osteoporosis and describe the demographic, global and secular trends in its epidemiology.

Genetics

Heritability estimates in osteoporosis

Peak bone mass is an important factor in determining BMD in later life. It has been suggested by twin and family studies that between 50% and 85% of the variance in BMD is determined by heritable factors [10–12], including both genetics and shared environmental exposures. These estimates do, however, vary depending on the skeletal site, with lumbar spine BMD demonstrating a greater heritable component than the distal forearm BMD [10, 12, 13]. Several studies have suggested that increasing age also influences the extent to which bone outcomes are determined by heritable factors. It has been shown that the heritable component of BMD is lower in postmenopausal compared with premenopausal women [10, 12], probably reflecting the greater role of additional lifestyle, dietary and disease-related factors occurring in postmenopausal women. Similarly, the heritable component of the rate of change in BMD in postmenopausal women is lower than that for peak bone mass, which occurs much earlier in life [14].

In terms of osteoporotic fractures, it is known that the risk is greater in those with a parent who has suffered a hip fracture. There is, however, less evidence for a significant genetic component to this association. A heritable component has also been found in the determination of femoral neck geometry [15], markers of bone turnover [16], age at menopause [17], and muscle strength [18], all of which confer some susceptibility to osteoporotic fracture. These factors, in addition to the associations with BMD, suggest that there is likely to be a role in fracture prediction; however, due to the size of the effect, it has been difficult to demonstrate in epidemiological studies.

Genetic studies in osteoporosis

Having determined that there is a small, but significant, genetic component to the risk of osteoporosis, different types of genetic investigations
Epidemiology and Genetics of Postmenopausal Osteoporosis

have been used to attempt to identify specific genetic loci. Linkage studies are useful in identifying genetic mutations in monogenic disorders and the genes responsible for a number of rare diseases associated with severe osteoporosis, fragility fractures or high bone mass, which result from single gene mutations inherited in classical Mendelian fashion, have been identified through this technique. Osteogenesis imperfecta, for example, is most commonly caused by mutations in the \textit{COL1A1} and \textit{COL1A2} genes resulting in abnormal type 1 collagen formation. Loss of function mutations in the \textit{LRP5} gene, encoding LDL receptor-related protein 5, a key regulator in osteoblastic bone formation, have been implicated in osteoporosis-pseudoglioma syndrome. Conversely gain-of-function mutations in the same gene are associated with familial high bone mass syndrome.

However, postmenopausal osteoporosis has been associated with a large number of common genetic variants each of which imparts only a minor effect. Linkage studies have therefore been of limited success in identifying contributory genes due to the low power to detect these common variants.

Candidate gene association studies (CGAS) and genome wide association studies (GWAS) have successfully identified a number of susceptibility loci. In CGAS, candidate genes are chosen for analysis based on a known role in the regulation of calcium metabolism or bone cell function. Many of the causative genes in monogenic disorders of bone fragility have been investigated. Single nucleotide polymorphisms (SNPs) are common variants which occur in at least 1% of the population. The frequency of these SNPs in candidate genes are compared in unrelated subjects in either a case-control study for categorical outcomes, for example history of an osteoporotic fracture, or as a population study for a quantitative outcome, for example BMD. A number of susceptibility variants have been identified using this method. However, false negative results are not uncommon due to limited power of the studies, and the results of studies in different populations are often conflicting.

With increasing acceptability to undertake genetic studies that are not hypothesis driven, GWAS have been able to clearly and reproducibly identify susceptibility loci for BMD variation. Large numbers (100 000–1 000 000) of common SNPs spread at close intervals across the genome are analyzed rather than focusing on a single candidate gene. A significant observation in the variant site is interpreted to indicate that the corresponding region of the genome contains functional DNA-sequence variants for the disease or trait being studied. These can include sequence variants leading to amino acid alterations in proteins, changes to gene promoter regions or alterations to mRNA degradation. However, a number of potential loci have also been identified, for which the function remains unknown. This might additionally offer the possibility of identifying novel
pathways and mechanisms involved in bone formation and the development of osteoporosis. Due to the large number of tests, GWAS are subject to stringent statistical thresholds. As with CGAS, false negatives are likely. Meta-analysis has been increasingly used to determine the true effects of genetic polymorphisms. The GENOMOS consortium (Genetic Markers of Osteoporosis; www.genomos.eu) was initially formed to undertake prospective meta-analysis of CGAS, and has identified SNP variants in \textit{COL1A1} and \textit{LRP5} associated with femoral and lumbar spine BMD. It has subsequently developed into the GEFOS (Genetic Factors for Osteoporosis; www.gefos.org) consortium which is undertaking meta-analysis of ongoing GWAS, and has identified or confirmed a number of loci associated with lumbar spine or femoral neck BMD [19].

\textbf{Genes involved in osteoporosis}

A number of genes have been identified through CGAS and GWAS as possible candidates for the regulation of bone mass and osteoporotic fracture susceptibility. A substantial number of these can be classified as influencing three biological pathways: the estrogen pathway, the Wnt-\(\beta\)-catenin signaling pathway and the RANKL-RANK-OPG pathway. These are briefly summarized below.

\textbf{The estrogen pathway}

Estrogen is a well-recognized regulator of skeletal growth, bone mass and bone geometry. Estrogen receptor deficiency and aromatase deficiency are monogenic disorders associated with osteoporosis. Genetic variation at a number of SNPs in the estrogen receptor type 1 gene (\textit{ESR1}) have been associated with many osteoporotic traits and risk factors including BMD [19], age at menopause [20] and postmenopausal bone loss [21].

\textbf{Wnt-\(\beta\)-catenin signaling pathway}

The Wnt signaling pathway has a key role in many developmental processes. In bone, the activation of this pathway by Wnt binding to LRP5 or LRP6 transmembrane receptors leads to osteoblast differentiation and proliferation, bone mineralization and reduction in apoptosis. Loss of function mutations of \textit{LRP5} result in osteoporosis-pseudoglioma syndrome, but more subtle polymorphisms have been associated with variance in BMD or fracture risk in the normal population. Some of these variants have been confirmed by meta-analysis [19, 22]. Other osteoporosis susceptibility genes affecting the Wnt-\(\beta\)-catenin signaling pathway have been indentified at genome-wide significance level. These include \textit{SOST} encoding sclerostin, an antagonist of Wnt; \textit{MEF2C}, which may regulate \textit{SOST} expression; \textit{FOXC2}, which activates the signaling pathway; \textit{WLS} encoding a
transmembrane protein which promotes Wnt release; and CTNNB1, which encodes β-catenin, a protein involved in the signaling cascade [23].

**RANKL-RANK-OPG pathway**

RANKL (receptor activator of nuclear factor κB ligand) binds to RANK on osteoclast precursor cells. It stimulates the differentiation of osteoclasts and activates bone resorption. Osteoprotegerin (OPG) has antagonistic actions to RANKL. A number of SNPs in the coding regions and in proximity to the OPG (TNFRSF11B), RANK (TNFRSF11A) and RANKL (TNFRSF11) genes have been associated with BMD and osteoporotic fracture risk through CGAS and GWAS and subsequently confirmed by meta-analysis [19, 24, 25]. Although the variance in BMD explained by these genes is small, the identification of these associations highlights the importance of this pathway in skeletal maintenance.

Additionally a number of candidate osteoporosis susceptibility genes have been identified from GWAS but their function in bone metabolism is yet to be elucidated; and a number of other candidate genes known to have a role in skeletal maintenance have shown inconsistent association with BMD in CGAS and not yet attained genome-wide significance in meta-analysis, including COL1A1 and the vitamin D receptor gene (VDR) [23]. The influence of environmental exposures on the genome might account for these inconsistent findings.

**Early life, gene-environment interactions and epigenetics**

Despite a large number of potential genetic loci suggested through CGAS and GWAS studies, these polymorphisms can explain only a small proportion (1–3%) of the observed variance in BMD in the population. There is, however, increasing recognition that environmental factors influence osteoporosis risk through alterations in gene expression and epigenetic mechanisms. As a result, the phenotype that develops from a specific genotype varies greatly depending on environmental exposures and it is likely to be the significant role of these epigenetic mechanisms that explains why BMD is highly heritable but only a small proportion is accounted for by genetic variation.

A number of examples of gene–environment interaction in both the fetal and early postnatal phases of life are emerging with regards to one’s risk for osteoporosis. For example, in a UK cohort study, no significant associations were identified between either the VDR genotype or birthweight and lumbar spine BMD. However, the relationship between lumbar spine BMD
and VDR genotype varied according to category of birth weight, and a statistically significant interaction between birth weight and VDR genotype as a determinant of lumbar spine BMD was found [26]. As birth weight reflects fetal nutrition, this finding suggests an interaction between the intrauterine environment and genetic influences. A similar study also demonstrated a significant interaction between human growth hormone (GH1) polymorphisms and weight in infancy, a reflection of early life environment, as determinants of rate of bone loss [27]. In the Framingham Offspring Cohort, genetic variation in the interleukin-6 promoter gene was only associated with hip BMD in a subset of women who were not using estrogen replacement therapy, and in those with an inadequate calcium intake [28], demonstrating gene-environment interactions in later life.

Epigenetics refers to stable alterations in gene expression that arise during development and cell proliferation. These changes are heritable and may persist through several generations, but do not involve DNA mutations [29]. Chemical modifications of the DNA and alterations to proteins associated with DNA loci lead to gene repression or increased gene activity. The most studied of these, and now believed to be a major contributor to gene expression, is DNA methylation. This involves the addition of a methyl group to cytosine at carbon-5 position of CpG dinucleotides. When methylation occurs in the promoter region of a gene, it generally leads to gene repression. The patterns of methylation vary with stages of development, but importantly, during fetal development, maternal and environmental factors can alter the pattern of DNA methylation, and subsequently influence gene expression during adult life.

Although no epigenetic mechanisms for osteoporosis have been fully elucidated in humans, the vitamin D response elements and glucocorticoid receptor are potential targets. Lower maternal 25(OH)-vitamin D concentration during late pregnancy has been associated with reduced bone mass in offspring during the neonatal period and mid-childhood [30,31]. This is partly mediated by umbilical venous calcium concentration [31]. Expression of the placental calcium transporter (PMCA3) also determines fetal skeletal growth [32]. It is therefore possible that epigenetic regulation of the PMCA3 gene represents the mechanism by which maternal vitamin D status effects offspring bone mass [33].

Environmental influences in childhood

Longitudinal growth in childhood begins to track shortly after birth, progressively increasing along a centile curve. Recent longitudinal studies have shown that tracking also occurs with bone traits from early childhood, through the pubertal growth spurt and into early adulthood [34]. Despite this, bone mineral accrual in childhood and early adult life can
be influenced by environmental factors and is of paramount importance in achieving optimum peak bone mass, which has a major effect on the risk of osteoporosis in later life [35]. In this same regard, a Finnish cohort study found directional associations between childhood growth rates and the risk of hip fracture in later life [36]. After adjustment for age and sex, the study demonstrated that a low growth rate between the ages of 7 and 15 years was associated with a significantly greater risk of hip fracture. This risk was also elevated in adults who were born short, but who obtained an average height by 7 years of age. In these children it is hypothesized that the skeletal envelope is forced ahead of the capacity to mineralize, a phenomenon which is accelerated during pubertal growth, and subsequently leads to the increased fracture risk. In adult life, several factors, such as diet, lifestyle, medication and comorbidities, are known to influence the risk of low BMD and fracture; these will be discussed in more detail in Chapter 4: Fracture risk assessment.

**Fracture epidemiology**

The incidence of fracture is bimodal, with peaks in childhood and in the elderly [37, 38]. Fractures in the young usually occur due to substantial trauma, are less common in females and tend to affect long bones. Bone mass progressively increases through childhood and usually reaches a peak by 30 years at which point the incidence of fracture is low. There is a progressive decline in BMD thereafter causing the prevalence of osteoporosis to increase with age. Rates of osteoporosis are particularly high in older women due mainly to the development of hypoestrogenemia following menopause. The reduction in bone density is associated with an increase in fracture risk; it has been shown that there is an approximate doubling of fracture risk for every standard deviation drop in BMD [39]. As a result, nearly three-quarters of all hip, vertebral and distal forearm fractures occur in those over 65 years of age [40]. Figure 1.1 clearly shows progressive increases in the incidence of hip, vertebral and wrist fractures with age in women with the exact nature of the relationship dependent on the type of fracture. Once an individual has suffered a fracture, their risk of further fracture is greatly increased and one meta-analysis has shown that the risk is up to 86% higher [41]. This may partly explain the clustering of fractures in some individuals.

In 2004 a report from the US Surgeon General highlighted the huge burden of osteoporosis-related fractures [42]. At that time, it was estimated that 10 million Americans over 50 years of age had osteoporosis and that 1.5 million fragility fractures were occurring each year. A study of fractures in Britain showed the population at risk to be a similar proportion to
Chapter 1

Figure 1.1 Hip, clinical vertebral, radiographic vertebral and wrist fracture incidence by age in men and women.

that in the US [43]. The lifetime risk of a hip fracture for a white woman is 1 in 6 [44]. In Western populations, hip fracture incidence increases exponentially with age with 90% occurring in those over 50 years of age [45]. In this age group, the risk in women is approximately double that in men [46], and as such when combined with greater longevity in females, 75% of hip fractures occur in women [47].

Hip fractures commonly lead to chronic pain, disability, reduced mobility and increased levels of dependence [48]. A significant number of individuals subsequently require long-term nursing care and this proportion increases with age. Hip fractures are also attended by an excess risk of mortality in the years immediately post fracture; survival rates at 5 years were found to be 80% of those expected when compared to age and sex matched individuals without a fracture [49]. Globally, it has been estimated that hip fractures account for around 740,000 deaths per year [50]. They also contribute to over a third of the total economic burden of fractures, reflecting their need for hospital inpatient management and the major costs associated with subsequent residential care. As the numbers of hip fractures are rising, it is estimated that by 2050 the worldwide direct and indirect costs will reach $131.5 billion per year [51].

The majority of vertebral fractures occur due to compressive loading associated with lifting, changing position, or are discovered incidentally. Vertebral fractures are not uncommon in postmenopausal women, with a 50-year-old white woman having a 16% lifetime risk of being affected [5]. Figure 1.1 shows an approximately linear increase in clinical vertebral fractures, and an almost exponential increase in radiographic vertebral fractures, with age. Although only about one third of radiographic vertebral deformities come to clinical attention, symptomatic vertebral fractures cause back pain, loss of height, deformity, immobility, and reduced pulmonary function. As with hip fractures they are also attended by an excess mortality [49].
Epidemiology and Genetics of Postmenopausal Osteoporosis

Distal forearm fractures usually occur as a result of a fall on an outstretched hand. Unsurprisingly, there is a peak in the incidence of these fractures in the winter, most likely to represent an increased frequency of falls on icy surfaces [52]. The gender disparity with this type of fracture is marked with an age-adjusted female to male ratio of 4:1 [43]. Although these fractures can lead to significant disability, particularly when they affect the dominant limb, there is no known associated increase in mortality rate.

**Geography**

Fracture incidence varies greatly across the world. On the whole, regions that are further from the equator have higher rates of fracture. This is thought to be related to less sun exposure resulting in lower levels of vitamin D [53]. However, exceptions to this rule do exist. In countries, such as Iran, where cultural codes encourage covering the majority of the body with clothes, skin exposure to the sun is limited. This practice may explain why, despite being close to the equator, 80% of the population are deficient in vitamin D and high rates of fracture are seen.

In general, fracture rates are similar in all Westernized Caucasian populations such as in Europe, America and Oceania. Within each of these individual regions, however, significant variation can be found. In the US, part of this disparity may be due to ecological factors that have been found to correlate with incidence patterns. These include water fluoride content, urbanization and socioeconomic status. Rates are particularly high in areas with a large proportion of those over the age of 65 years living below the poverty line [54].

In Europe, hip fracture rates are almost seven times lower in parts of southern Europe than in Scandinavia, and in particular Norway, which has some of the highest rates worldwide [55]. In areas where hip fracture risk is high, this tends to be reflected by increased rates of fragility fractures at other anatomical sites [18, 19]. The EVOS study examined the prevalence of vertebral deformities in countries across Europe. They showed a 3-fold difference in prevalence and again the highest frequencies were to be found in Scandinavia [56].

Moderate variation is seen throughout Asia with the highest rates identified in urbanized countries [57]. Due in part to its vast population, hip fractures arising in Asia account for a significant proportion of the world’s burden; recent estimates have put this figure at around 30%. In contrast, fracture rates from populations in Africa appear to be low. However, there is limited data from this region and its validity has been questioned.
Inaccurate case identification may therefore partly explain the low numbers but population demographics may also play a role.

**Secular trends**

Throughout the world, life expectancy is increasing and the number of elderly people is rising, particularly in developing countries. It is predicted that by the year 2050 there will be more than 1500 million people aged 65 years or over worldwide. Between 1990 and 2000, the incidence of fractures worldwide increased by one-quarter [4], and it has been estimated that the number of hip fractures will rise from 1.66 million in 1990 to over 6 million in 2050 [58]. These estimates assume a constant age-specific hip fracture incidence; however, the secular trends found depend markedly on geographical location (Figure 1.2).

It has been shown that in the majority of Western populations, including Oceania, North America and the UK, age-specific fracture rates rose until

![Figure 1.2: Secular trends in fracture incidence throughout the world](Source: Cooper C, et al. (2011) [61]. With kind permission from Springer Science+Business Media).
around 1980. This might have been caused in part by decreasing levels of physical activity, with less time spent outside, and higher rates of vitamin D insufficiency. Furthermore, during this time, a combination of medical and social factors led to improved survival of the frail elderly (i.e., those at greatest risk of fracture). Since this point, rates have either remained constant or started to decline. This trend may be due to a birth cohort effect, an increase in obesity or a specific improvement in the screening and treatment of osteoporosis. In particular, the introduction of bisphosphonates may have played a role although does not provide a full explanation [59, 60]. These changes are in contrast to rates in the developing world which have not declined.

The combination of disparate regional changes in population demographics and age-specific fracture rates is likely to cause a shift in the geographic distribution of fracture burden towards Asia and the developing world. Consequently, it has been estimated that only around 25% of hip fractures will occur in Europe and North America by 2050 [58].

Conclusion

Osteoporosis and osteoporotic fracture is globally a common condition, representing a huge individual and public health burden and associated with increased mortality. Although the clinical outcomes most frequently occur in females in later life, there is increasing evidence to suggest that environmental factors and gene-environment interactions occurring throughout the lifecourse, including prenatal life, childhood and early adulthood, are implicated in osteoporosis and fracture risk. Further identification of and consideration for these factors are important in reducing the currently increasing global burden of osteoporosis.

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References

Chapter 1

Epidemiology and Genetics of Postmenopausal Osteoporosis


Chapter 1