From THE DEPARTMENT OF CLINICAL SCIENCE,
INTERVENTION AND TECHNOLOGY
THE DIVISION OF ORTHOPEDICS AND BIOTECHNOLOGY

Karolinska Institutet, Stockholm, Sweden

ASPECTS OF FRACTURE RISK IN ELDERLY WOMEN

Axel Wihlborg

Stockholm 2018

Published by Karolinska Institutet.
Cover design by Ola Laurin
Printed by AJ E-print AB 2018

ISBN **978-91-7831-112-5**

Institutionen för klinisk vetenskap, intervention och teknik (CLINTEC), Enheten för ortopedi och bioteknologi

Aspects of fracture risk in elderly women

AKADEMISK AVHANDLING
som för avläggande av medicine doktorsexamen vid Karolinska Institutet offentligen försvaras i Föreläsningssalen Solen (4U), Alfred Nobels Allé 8, plan 4. Karolinska Institutet, Huddinge.

Fredagen den 8 juni, kl. 09:00

av
Axel Wihlborg
Leg. läkare

Huvudhandledare:
Docent Paul Gerdhem
Institutionen för klinisk vetenskap, intervention och teknik (CLINTEC)
Enheten för ortopedi och bioteknologi
Karolinska Institutet

Bihandledare:
Docent Ingrid Bergström
Institutionen för klinisk vetenskap, intervention och teknik (CLINTEC)
Enheten för ortopedi och bioteknologi
Karolinska Institutet

Fakultetsopponent:
Docent Anna G Nilsson
Institutionen för medicin,
Sahlgrenska Akademin
Göteborgs Universitet

Betygsnämnd:
Docent Kenneth Jonsson
Institutionen för kirurgiska vetenskaper
Uppsala Universitet

Docent Marketta Henriksson
Institutionen för fysiologi och farmakologi
Karolinska Institutet

Docent Björn Rosengren
Institutionen för kliniska vetenskaper, Malmö
Lunds Universitet

Stockholm 2018

To my parents, sisters and Viktoria

ABSTRACT

The main purpose of this thesis was to identify and further specify factors relevant for the assessment of fracture risk. The studies were conducted on two different cohorts of elderly women. The Osteoporosis Prospective Risk Assessment study (OPRA) is a cohort of women followed for over a decade, from the age of 75. The Distal Forearm Fracture study (DFF) is a cross-sectional cohort of postmenopausal women with a distal forearm fracture and age-matched controls, with a mean age of 65.

Even though the mortality among individuals that declined participation in the OPRA study was increased, it appeared that participants were fairly representative in terms of fracture risk in general. In the OPRA cohort, self-reported history of fall corresponded to increased risk of distal forearm and any osteoporosis-related fracture. Decreased gait speed and a failed balance test corresponded to increased hip fracture risk. Current smokers had an increased risk of vertebral and any osteoporosis-related fracture. Smoking cessation reduced the risk of vertebral fracture. Time as a smoker corresponded to increased vertebral fracture risk. However, amount smoked and time from cessation did not affect fracture risk. In the DFF cohort, women with fracture had decreased site-specific volumetric trabecular and cortical BMD, as well as geometric alterations with increased size and decreased cortical thickness. Weak correlations between parathyroid hormone levels and 25-hydroxy vitamin D with cortical and trabecular bone were observed. Out of 161 women with a distal forearm fracture, 13 women (8%) were diagnosed with primary hyperparathyroidism, suggesting a higher prevalence than in the general population.

In summary, external validity in studies on fracture risk may be satisfactory. It appears to be of great importance to consider physical function and smoking habits in elderly women in the fracture risk assessment. In addition, both trabecular and cortical bone reductions as well as geometric alterations of the forearm may be contributing factors in the pathogenesis of a distal forearm fracture. The occurrence of primary hyperparathyroidism appears to be high in women with a distal forearm fracture, suggesting that further evaluation following a low-energy distal forearm fracture might be beneficial.

SAMMANFATTNING

Osteoporos är en vanlig skelettsjukdom som kännetecknas av minskad skelettstyrka och ökad frakturrisk. Risken för att drabbas av en fraktur ökar med åldern och postmenopausala kvinnor är särskilt drabbade på grund av sjunkande nivåer av östrogen. Osteoporosrelaterade frakturer (handledsfraktur, höftfraktur, kotkompression) är förknippade med mycket lidande och kan även innebära ett förkortat liv. Det finns effektiv behandling som stärker skelettet och därför är det viktigt att identifiera kvinnor som har en ökad frakturrisk. Huvudsyftet med den här avhandlingen var att identifiera och ytterligare specificera faktorer som är relevanta vid frakturriskbedömningen.

Studierna genomfördes på två olika studiegrupper: en observationsstudie i Malmö som består av kvinnor som följts i över ett decennium från 75 års ålder och en tvärsnittsstudie av kvinnor med och utan handledsfraktur med en medelålder på 65 år.

Vi undersökte om kvinnor som valt att delta i observationsstudien var representativa även för de som avstått. Dödligheten bland kvinnorna som avstått att delta var högre än vad den var bland de som deltog, men deltagarna var trots det relativt representativa när det kom till frakturrisken. I en annan studie fann vi att förekomsten av ett fall det senaste året innebar en ökad risk för att drabbas av en handledsfraktur, eller en osteoporosrelaterad fraktur. En nedsatt gånghastighet eller ett misslyckat balansprov medförde en ökad risk för att drabbas av en höftfraktur. Vi såg att rökare hade en ökad risk för att drabbas av en kotkompression, eller en osteoporosrelaterad fraktur. Att ha slutat röka minskade risken för en kotkompression.

Kvinnor med en handledsfraktur hade en nedsatt trabekulär och kortikal skelettdensitet. De hade även geometriska förändringar med en ökad omkrets av strålbenet, kombinerat med en minskad kortikal tjocklek. Både paratyreoideahormon och D-vitamin korrelerade svagt med kortikalt och trabekulärt skelett. Av 161 kvinnor med en handledsfraktur diagnosticerades 13 (8%) med primär hyperparatyreoidism, vilket antyder att det hos denna grupp är vanligare än hos den allmänna befolkningen.

Sammanfattningsvis verkar överförbarheten av resultaten i studier som handlar om frakturrisk vara tillfredsställande. Det förefaller vara viktigt att ta hänsyn till rökvanor och att bedöma den fysiska förmågan hos äldre kvinnor när frakturrisken bedöms. Både minskningar av trabekulärt och kortikala skelett, såväl som geometriska förändringar kan vara bidragande faktorer vid uppkomsten av en handledsfraktur. Förekomsten av primär hyperparatyreoidism

kan vara hög hos kvinnor med en handledsfraktur vilket antyder att det kan vara fördelaktigt med ytterligare uppföljning och utredning efter en handledsfraktur.

LIST OF SCIENTIFIC PAPERS

I. Wihlborg A, Åkesson K, Gerdhem P (2014) External validity of a population-based study on osteoporosis and fracture. Acta Orthopaedica 85(4):433-437.

II. Wihlborg A, Englund M, Åkesson K, Gerdhem P (2015) Fracture predictive ability of physical performance tests and history of falls in elderly women: a 10-year prospective study. Osteoporosis International 26(8): 2101-2109.

III. Thorin M H, Wihlborg A, Åkesson K, Gerdhem P (2016) Smoking, smoking cessation, and fracture risk in elderly women followed for 10 years. Osteoporosis International 27(1): 249-255.

IV. Wihlborg A, Bergström K, Åkesson K, Bergström I, Gerdhem P. Site-specific volumetric skeletal and muscle changes in women with a forearm fracture at different stages of ageing. Manuscript.

V. Wihlborg A, Bergström K, Gerdhem P, Bergström I. Parathyroid hormone disturbances in postmenopausal women with distal forearm fracture. Manuscript.

CONTENTS

LIST OF ABBREVIATIONS

25OHD	25-hydroxy vitamin D
aBMD	Areal bone mineral density
BMC	Bone mineral content
BMD	Bone mineral density
BSIc	Bone strength index
CSA	Cross sectional area
DXA	Dual-energy X-ray absorptiometry
OPG	Osteoprotegerin
PTH	Parathyroid hormone
PHPT	Primary hyperparathyroidism
pQCT	Peripheral quantitative computed tomography
RANK	Receptor Activator of Nuclear factor Kappa-β
RANKL	Receptor Activator of Nuclear factor Kappa-β Ligand
SSI	Strength strain index

1 INTRODUCTION

Osteoporosis is a common systemic bone disease characterized by reduced bone strength and increased fracture risk. The risk of contracting a fracture increases with age and postmenopausal women are particularly affected. A fracture may be associated with short term disability, but may also result in permanent disability or even a shortened life-span. Effective treatment that strengthens bone and reduces fracture risk is available and identifying those at risk is therefore of interest, both for the individual and society at large. Fracture risk is dependent on bone properties, such as mass and structure, but also on factors not related to bone. To introduce the reader to the papers in this thesis here follows a description of bone, osteoporosis, risk factors for fracture and measurements of bone mass and structure.

2 BACKGROUND

2.1 BONE

The main functions of bones in the human body are mechanical and metabolic. Bones provide structure, stability and protection while at the same time remaining low weight and facilitating movement by the attachment of muscles, tendons and ligaments. Furthermore, bone serves as a reservoir for calcium and phosphate and plays an important role in the calcium homeostasis. Bone also interacts with the hematopoiesis in the bone marrow (1, 2).

To comply with the mechanical and metabolic needs, the bone tissue is constantly being remodeled by bone removal and new bone formation. Through the remodeling process, bone is renewed and through modeling, bone is reinforced at skeletal sites with high mechanical load. In addition, the remodeling process allows for regulation of the calcium homeostasis. Through various mediators, low concentration of serum calcium stimulates bone resorption, resulting in a release of calcium from bone, at the expanse of bone mass. Consequently, the integrity of the bone is a result of both mechanical and the metabolic functions (1).

2.1.1 Organization

On a macroscopic level, bone is organized in terms of cortical and trabecular bone (Figure 1). Cortical bone is the more compact outer layer of the bone while trabecular bone is the more porous inner portion. Trabecular bone consists of a network of rods and plates, which is surrounded by bone marrow. Because of different mechanical requirements in different parts of the body, bones vary in size, shape and constitution. The proportion of cortical and

trabecular bone vary in different skeletal sites. The metaphysis and epiphysis of long bones in arms and legs mainly consists of trabecular bone. Trabecular bone is also the main component of the vertebral body and pelvis. The diaphysis of long bones mainly consists of cortical bone with a central hollow part, which is mainly comprised by bone marrow (1, 2).

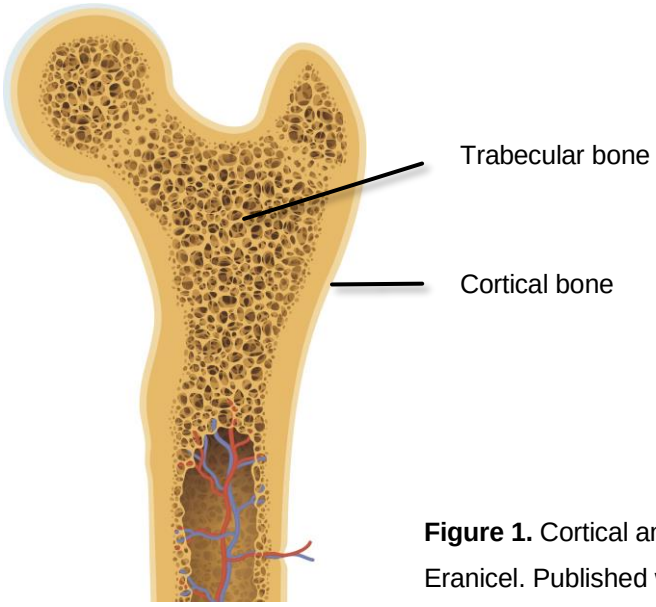


Figure 1. Cortical and Trabecular bone at the hip (By Eranicel. Published with permission from Shutterstock)

The outer contour of the bone, except parts with cartilage or sites for attachment of tendons and ligaments, is covered by a membrane called periosteum. The periosteum consists of an outer fibrous layer that is rich in nerve fibers and blood supply. The inner layer of the periosteum is called cambium and consists mainly of progenitor cells, which have the ability to differentiate into bone forming osteoblasts (Chapter 2.1.3) (1, 2).

2.1.2 Histology

Bone tissue consists of a matrix of non-organic and organic matter, surrounding small amounts of cells. The organic matter of the matrix consists mainly of type I collagen molecules, arranged in fibrils that cross-link and form type I collagen fibers. The non-organic matter of the matrix is comprised of crystalized mineral salts, mainly hydroxyapatite, which is deposited in the collagen fibers in a calcification process. Hydroxyapatite is a crystalline complex of calcium and phosphate with the formula: $Ca_{10}(PO_4)_6(OH)_2$. While the collagen fibers contribute to the flexibility and tensile strength of the bone, the hydroxyapatite provides stiffness. In addition, the matrix consists of water and small amounts non-collagenous proteins (1, 3).

In the adult cortical bone, the calcified collagen fibers are organized into parallel arranged lamellae, which form a cylindrical structural unit called osteon (Figure 2). Each osteon entails a central cavity called, the Haversian canal, which contains blood vessels and nerve fibers,

which is surrounded by concentric rings of lamellae. The lamellae and osteons are mainly arranged in the direction of the bones vertical axis. Additional traversing canals, Volkmann´s canals, facilitate connections between Haversian canals and the cortical surface. The lamellae in trabecular bone do not form osteons but are arranged concentric in line with each trabecula (1).

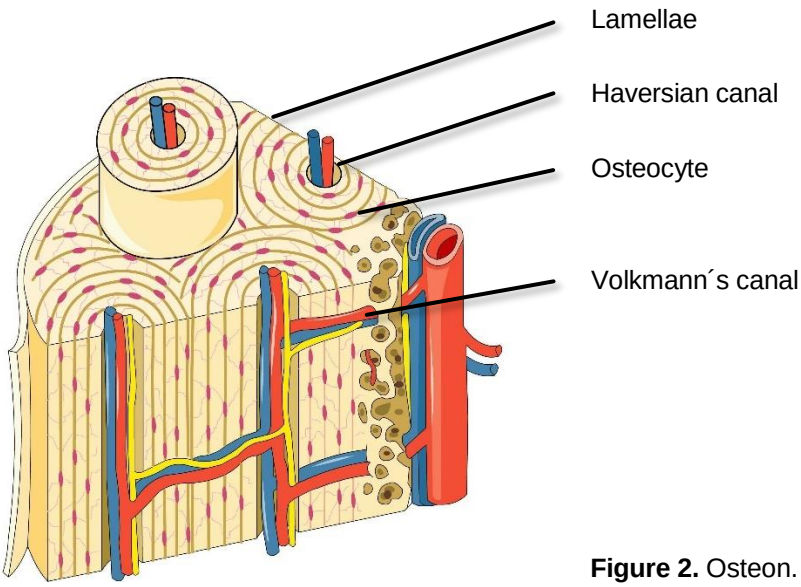


Figure 2. Osteon. (By Mmutlu. Published with permission from Shutterstock)

2.1.3 Cells

The main cells found in bone, responsible for bone formation and remodeling are: osteoblasts, osteocytes, lining cells and osteoclasts (Figure 3).

Osteoblasts are mononucleated cuboid cells derived from the mesenchymal stem cell lineage. The differentiation into osteoblast is controlled by several regulatory factors. Runx2 and osterix are the two most essential transcription factors. Osteoblasts synthesizes and secretes osteoid, consisting predominately of type I collagen and non-collagenous proteins. In addition, osteoblasts produce hydroxyapatite by precipitation of calcium and phosphate (1).

Osteocytes are the continued differentiation of osteoblasts after encasement in bone matrix. They are the most commonly found cells in bone tissue. Osteocytes reside between the lamellae in lacunae, and communicate to surface lining cells and osteoblasts trough dendritic processes in canaliculi (Figure 2). By registration of mechanical stress through integrin receptors, the osteocytes are believed to play an important role in the adaptation of bone remodeling to changes in mechanical forces (4, 5).

Lining cells are flat cells derived from osteoblasts, covering inactive bone. Lining cells are equipped with receptors for various agents and communicate with osteocytes within the bone. When stimulated, the lining cells can induce bone remodulation (1).

Osteoclasts are large multinucleated cells derived from the hematopoietic cell lineage. Osteoclasts are differentiated from macrophage cells and the differentiation is dependent on the presence of the cytokine, Receptor Activator of Nuclear factor Kappa-β Ligand (RANKL). The binding of RANKL to the receptor called, Receptor Activator of Nuclear factor Kappa-β (RANK) on preosteoclasts initiates differentiation and activation. Conversely, Osteoprotegerin (OPG) is a receptor that inhibits osteoclast differentiation and activation by binding to RANKL, and thereby preventing RANKL from binding to RANK.

The osteoclasts adhere to bare bone surface and forms a tight seal with a capsuled compartment between the cell and the bone matrix (resorptive pit). The release of acid into the resorptive pit by proton ATPase dissolves the hydroxyapatite into calcium and phosphate. In addition, proteases are released into the resorptive pit through lysosomes, which degrades the collagen matrix (1, 6).

2.1.4 Remodeling

Bone remodeling refers to the process of bone formation and resorption (Figure 3). By remodeling, the bone maintains strength by removing and replacing old bone tissue with new bone. Trough remodeling, the bone can also maintain systemic calcium homeostasis (1).

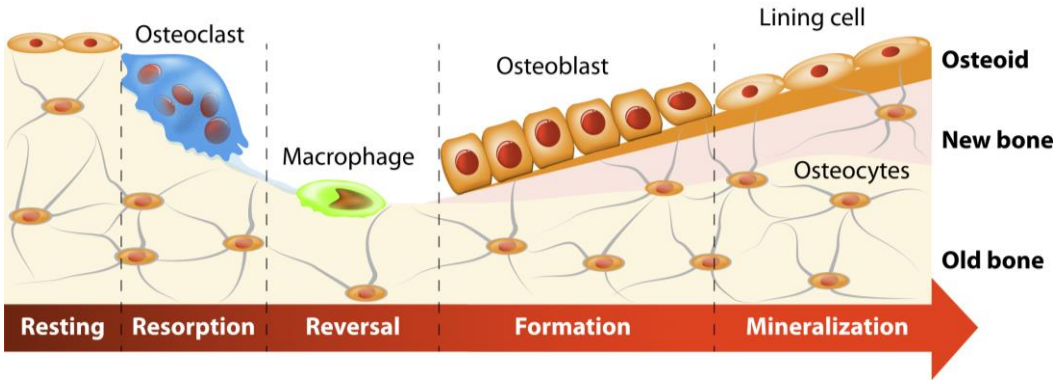


Figure 3. Bone cells and the remodeling process (By Designua. Published with permission from Shutterstock)

Bone remodeling occur at the bone surface which in cortical bone corresponds to either the endosteal or periosteal surface, or within a Haversian and Volkmann canal. Upon activation, preosteoclasts are recruited and differentiated to osteoclasts. Following attachment to the bone surface the osteoclasts resorb bone matrix. In cortical bone, this process creates a

conical hole wherein a capillary vessel grows, supplying osteoblast progenitor cells that differentiate to osteoblasts. The whole is filled with bone matrix and the result is a new osteon. In trabecular bone, remodeling occurs on the trabecular surface (1, 7).

The remodeling process is continuous and the turnover rate (the rate in which bone is being replaced) differs between skeletal sites. Turnover rate is higher in central skeletal sites such as the vertebral body and pelvis compared to peripheral skeletal sites, and is generally higher in trabecular bone compared to cortical bone. This is due to the construction of the trabecular network, which entails a larger surface area for remodeling (1).

Bone modeling is the process in which the bone adapts to changes in mechanical loading and is less continual in adults (1).

2.2 BONE AND THE CALCIUM HOMEOSTASIS

The maintenance of blood and extracellular fluid calcium concentration within a physiological range is referred to as the calcium homeostasis. The concentration of calcium in blood is equivalent to that of most extracellular spaces. However, it is considerably lower intracellularly. In blood, calcium occurs either as ionized calcium, complexed to anions or protein-bound, predominately to albumin. Around half of the total calcium in blood is ionized calcium, which is the biological active form and therefore under regulatory control. Measurements of total blood serum calcium concentration are influenced by the amount of circulating albumin, and when estimating the amount of biological active calcium, adjustments for albumin concentration is required. Physiological concentration of serum ionized calcium is essential for the normal functioning of several biological processes such as blood coagulation, muscle contraction and nerve conduction, as well as in maintaining cellular integrity. Bone is the most important reservoir of calcium in the human body (8, 9).

2.2.1 Regulation of the calcium homeostasis

The calcium homeostasis is regulated by parathyroid hormone (PTH), calcitonin and the active form of vitamin D (1,25 $(OH)_2D$, calcitriol). The three hormones maintain homeostasis by a series of synergic and negative feedback mechanisms, affecting primarily the intestinal tract, kidney and bone.

2.2.2 Parathyroid hormone

The parathyroid glands are sensitive for small changes in ionized calcium concentrations. Calcium sensing receptors on the parathyroid gland cell membrane react immediately to changes in ionized calcium levels and decreasing levels activate a signaling pathway,

resulting in the release of PTH to the circulation. The biological active intact PTH is cleaved, predominately in the liver, producing inactive fragments. The inactive PTH fragments are cleared by the kidneys and have a significant longer half-life than intact PTH (10). There are two main receptors for PTH; the PTH1R receptor is found in bone and kidney tissue, while the PTH2R receptor is found mainly in the nervous system (11, 12).

PTH increases the concentration of ionized calcium by: (i) increasing reabsorption of calcium in the kidneys by a direct effect on the proximal renal tubule, simultaneously increasing the renal phosphate excretion, (ii) stimulating osteoclast activity which increases the release of ionized calcium from bone, and (iii) activating the enzyme 1α-hydroxylase in the proximal renal tubule, which in turn increases the activation of vitamin D (Chapter 2.2.4) (10).

The effects of PTH on osteoclast activity are mediated through an increased RANKL expression, thereby stimulating the differentiation and activity of osteoclasts. Continuously increased PTH levels have catabolic effects on bone (10).

2.2.3 Calcitonin

Calcium sensitive C cells in the thyroid gland releases calcitonin in response to elevated levels of ionized calcium. Calcitonin decreases ionized calcium levels by inhibiting osteoclast activity and decreasing tubular resorption of ionized calcium in the kidneys (13).

2.2.4 Vitamin D

There are three sources of vitamin D; the skin, diet and dietary supplements. The skin is the most important contributor. In the diet, vitamin D is mainly found in fatty fish. In some countries, vitamin D is also supplemented in dairy products. In the skin, *7-dehydrocholesterol* is transformed by UV-B rays from sunlight, causing the formation of *pre-vitamin D*, which in turn under the influence of skin temperature, is converted to *cholecalciferol* (vitamin D). Cholecalciferol is the precursor of active vitamin D (calcitriol) and the activation process involves hydroxylation of carbon atoms in two steps, in the liver and kidney respectively. In the liver, the hydroxylation of carbon 25 is facilitated by the enzyme cytochrome P450, resulting in *25(OH)D*. The second hydroxylation, of carbon 1, is facilitated by the enzyme 1α-hydroxylase (which is activated by PTH) in the kidney, resulting in the active vitamin D hormone *1,25(OH)$_2$D, calcitriol*. In this text, the active hormone calcitriol is simply referred to as vitamin D (14, 15).

Vitamin D circulates bound to vitamin D binding protein (DBP) and the target receptors (VDR) are mainly located in the renal and intestinal epithelia as well as in bone. The main

mechanisms of vitamin D are regulation of calcium and phosphate metabolism, and the furtherance of normal bone mineralization. However, VDR´s have been found in a variety of tissues, emphasizing the possible widespread effects of vitamin D (14, 15).

Vitamin D acts mainly by increasing intestinal absorption of calcium by stimulating calcium channels, calcium-transporting proteins (calbindins) and calcium pumps in the small intestine and duodenum. Furthermore, vitamin D increases the reabsorption of calcium in the kidney tubules with similar mechanism as in the intestine (16).

2.3 AGE-RELATED BONE CHANGES

The skeletal growth during childhood and adolescence results in rapidly increasing bone mass. In the years following puberty, skeletal maturation is achieved and is defined as peak bone mass (Figure 4). A variety of factors influence the degree of bone growth during childhood and adolescence, including heredity, diet, disease, physical activity and smoking. Moreover, influences during intrauterine life seem to have an impact on peak bone quality and mass (17). The peak bone mass is greater in men compared to women, which is primarily explained by an increased bone size in men (17-21).

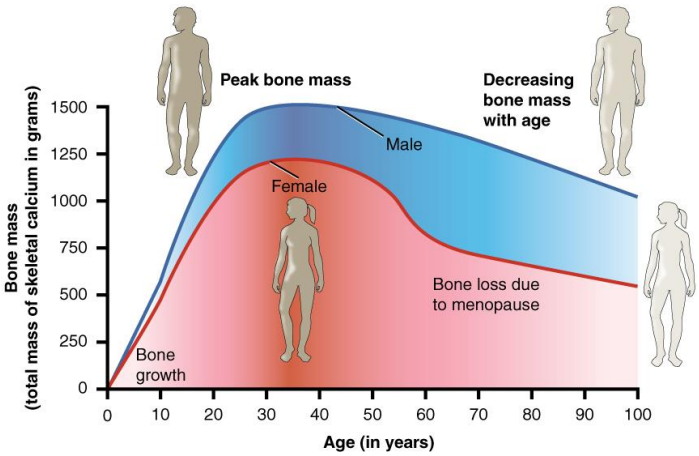


Figure 4. Age and bone mass (By Anatomy & Physiology. Connexions Web site, via Creative Commons)

After adolescence, bone mass decreases continuously with deteriorations of both cortical and trabecular bone. The established peak bone mass and size is therefore important in determining future bone mass. Trabecular network deterioration results in fewer and thinner trabeculae while cortical deterioration entails a reduced cortical thickness and increased cortical porosity. However, the reductions of material properties are somewhat compensated for by geometric alterations. Endosteal resorption in combination with periosteal apposition

of the cortical shell increases the bone size. The resulting increased cross sectional area, with a displacement of the cortical shell from the central axis, provides a greater resistance to load. In men, periosteal apposition is greater, which is believed to be a contributing factor to the greater bone strength in men (18, 19, 21, 22).

2.4 OSTEOPOROSIS

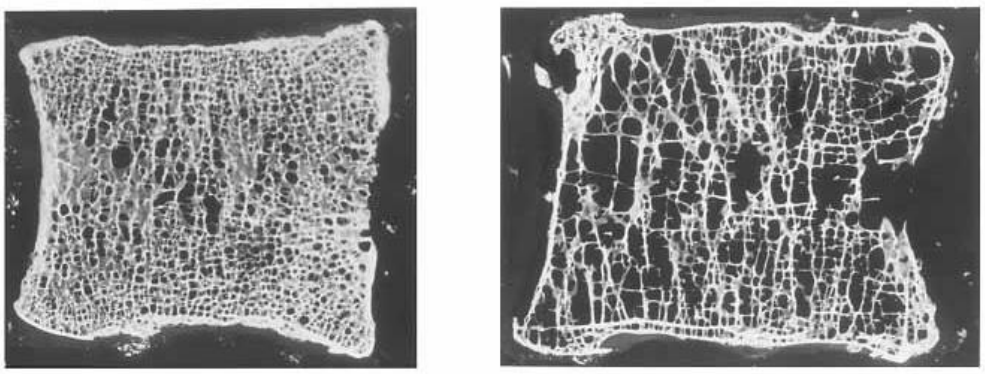


Figure 5. Normal vertebra and osteoporotic vertebra (By Turner Biomechanics Laboratory, via Creative Commons)

Osteoporosis is, according to the National Institutes of Health (NIH), defined as "a skeletal disorder characterized by compromised bone strength predisposing a person to an increased risk of fracture". Furthermore, bone strength is described as dependent on two bone properties; bone density and quality (such as microarchitecture and rate of remodulation) (23). The World Health Organization (WHO) defines osteoporosis solely by areal bone mineral density (aBMD), measured with Dual-energy X-ray absorptiometry (DXA) (Chapter 2.6.1) (24).

As a clinical diagnostic criterion, the measured areal BMD in a specific patient must be compared to a healthy reference population. Due to measurement differences between DXA devices, the actual aBMD measurements are standardized and changes are described as standard deviations (SD) from the mean. In postmenopausal women and men over the age of 50, the measured aBMD is compared to the young (20-29 years) reference population mean, and the SD is referred to as T-score. In premenopausal women and younger men, the aBMD is compared to the age-matched mean, and referred to as Z-score (23-25).

The WHO diagnostic criterion for osteoporosis is a decrease of 2,5 standard deviations or more (T-score $\leq$ -2.5) of the total hip, femoral neck or lumbar spine (23, 25).

2.4.1 Primary osteoporosis

In women, a rapid predominately trabecular bone loss occurs during menopause because of declining estrogen levels. Estrogen has several effects on bone remodulation and is a strong determinant of bone mass. The bone anabolic effect of estrogen is mediated by a suppression of RANKL, caused by a high OPG production, resulting in an inhibition of osteoclast differentiation and activity. In addition, estrogen inhibits osteoclast activity by direct mechanisms and through several other cytokines and growth factors. The rapid postmenopausal bone loss levels out after 4-8 years and is followed by continued gradual deterioration (Figure 4) (18, 19, 22).

2.4.2 Male osteoporosis

Men do not suffer from a rapid bone loss as women do during menopause. However, declining levels of testosterone and estrogen during ageing is believed to contribute to bone loss in elderly men. In men with osteoporosis, 50% suffer from secondary osteoporosis compared to about 25 % in women (18, 19, 22).

2.4.3 Senile osteoporosis

Contributing to bone fragility in the elderly is declining levels of active vitamin D due to less production in the skin and reduced ability to activate vitamin D in the kidneys and liver. The resulting negative calcium balance triggers secondary increments of PTH (Secondary hyperparathyroidism, SHPT) which, combined with decreased nutrition and degenerative muscle loss (sarcopenia), further deteriorates the bone (18, 19, 22, 26-28).

2.4.4 Secondary osteoporosis

Several medical conditions and pharmaceuticals affect bone remodeling and may consequently lead to the development of secondary osteoporosis. Some of the most prevalent are listed in Table 1 (23, 29).

One of the most common causes is glucocorticoid therapy (Cortisol). Glucocorticoids affect bone by; stimulating osteoclast differentiation by increasing the expression of RANKL, decreasing osteoblast activity and inducing apoptosis of osteocytes (23, 29, 30).

Table 1. Secondary causes of osteoporosis

Conditions	- Endocrine disorders (Primary hyperparathyroidism, Thyrotoxicosis, Cushing´s, Hypopituitarism, Prolactinoma) - Hypogonadism (Oophorectomy, Turner syndrome, Klinefelter syndrome, Pregnancy, Lactation, Anorexia, hypothalamic amenorrhea) - Inflammatory disease (Rheumatoid arthritis, Systemic lupus erythematosus, Crohn's disease) - Kidney failure - Liver failure - Malabsorption (Surgery, Celiac disease) - Vitamin D deficiency - Multiple myeloma - Immobility - Organ transplantation
Pharmaceuticals	- Heparin - Glucocorticoids - Immunosuppressant - Aromatase inhibitors - Antiepileptic drugs - Gonadotropin-releasing hormone agonist - Methotrexate

Inflammatory diseases increase bone resorption by a direct effect on osteoclast activity trough inflammatory cytokines. In addition, many inflammatory diseases are treated with glucocorticoids. Malabsorption diseases such as celiac disease may reduce the intestinal absorption of calcium, inducing PTH secretion with subsequent increased bone resorption. Liver and kidney disorders can affect the hydroxylation and activation of vitamin D, which reduces the availability of biological active vitamin D. Vitamin D deficiency may also be caused by insufficient exposure to sunlight (23, 26, 27, 31).

Primary hyperparathyroidism (PHPT) is a common endocrine disorder, characterized by an excessive production of parathyroid hormone (PTH) in combination with hypercalcemia. The increased levels of PTH are most commonly caused by a single parathyroid gland adenoma and rarely by parathyroid gland carcinoma (32, 33). Occurrence of PHPT increases with age and the incidence among postmenopausal women have been reported to range between 1.34 and 3.4% in Scandinavian countries (34-36).

The increased levels of PTH result in hypercalcemia through increased bone resorption combined with increased intestinal absorption and kidney reabsorption of calcium. Severe adverse effects of PHPT include osteoporosis, increased fracture risk, renal stones, peptic ulcers, constipation, neuropsychological and neuromuscular symptoms such as muscle weakness. However, in most cases, the condition is asymptomatic (around 80%) (32, 33).

Nevertheless, progression into symptomatic disease may occur. There seems to be an association between decreased aBMD and increased fracture risk even in mild asymptomatic disease (37-40).

Normocalcemic PHPT is biochemically characterized by calcium levels in the upper reference range combined with increased levels of PTH, in the absence of secondary causes. Normocalcemic PHPT has been associated with decrements of aBMD and the condition has been suggested to progress over time (32, 38).

Surgical removal of the parathyroid glands (parathyroidectomy) has been proven beneficial in terms of osteoporosis, fracture risk, incidence of renal stones and quality of life, even in mild cases of the disease (32, 33, 39, 41-43). Surveillance of patients seems favorable when the surgical indications are not met (38).

2.4.5 Osteoporosis-related fractures

The clinical impact of osteoporosis is fractures and the most common fractures related to osteoporosis are: vertebral fractures, distal forearm fractures and hip fractures. In addition, fractures of the proximal humerus and pelvis may be regarded as osteoporosis-related. Osteoporosis-related fractures are the result of a low-energy trauma, commonly defined as falling from a standing height or a height less than 1 meter. Vertebral fractures, however, may occur without trauma (44).

Distal forearm fractures are more common in women and the incidence rises abruptly in adjunction to menopause, with a plateau around the age of 65. On the contrary, vertebral fracture incidence begins to rise after menopause, around the age of 55, and continues to increase gradually. The hip fracture incidence increases exponentially with increasing age, but the rise begins at higher age compared to forearm and vertebral fractures. In men, the fracture risk generally increases 5-10 years later, compared to women (45, 46).

Distal forearm fractures (Figure 6) cause acute pain and suffering. Many are at need of hospital care where the fracture in most cases is treated with a stabilizing cast or surgery, depending on fracture pattern, individual needs and preferences. A healed forearm fracture may be associated with joint stiffness, pain and functional impairment.

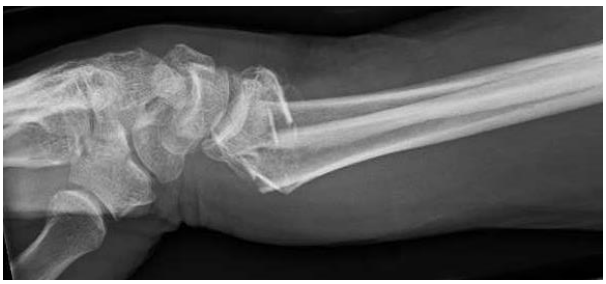


Figure 6. Distal forearm fracture

Vertebral fractures (Figure 7) can entail back pain, length reduction, deformity with increased kyphosis and protruding abdomen, reduced pulmonary function, reduced quality of life and increased mortality. Although many vertebral fractures occur without clinical verification (i.e. are incident radiographic findings), the clinical consequences seem to be similar (47, 48).

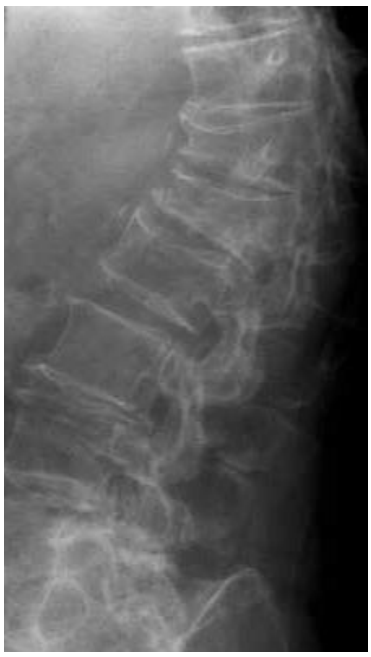


Figure 7. Vertebral fractures

A hip fracture (Figure 8) is an acute condition and the patient needs hospital care and surgery. Mortality following a hip fracture is around 25% within the first year from fracture after which it gradually decreases. Around 50% of those surviving a hip fracture suffer from life-long functional impairment (44).

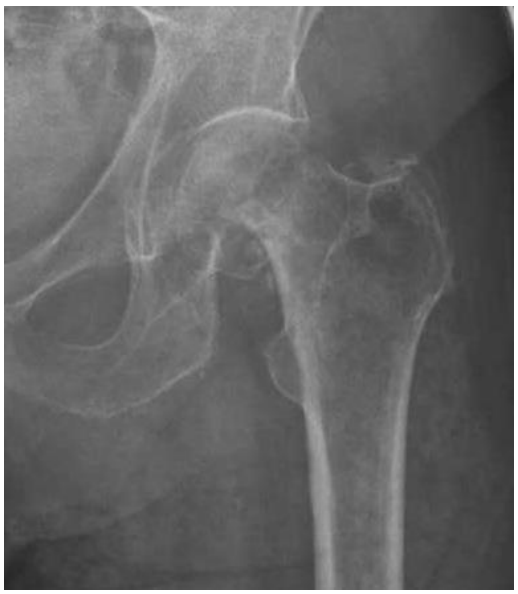


Figure 8. Hip fracture

Besides high individual morbidity and mortality, osteoporosis-related fractures have extensive consequences on society at large in terms of caregiving and costs (49, 50).

2.4.6 Treatment

Clinical considerations regarding the treatment of osteoporosis include addressing secondary causes and estimating the future fracture risk, which is determined by aBMD as well as other risk factors for fracture (Chapter 2.5). Non-pharmaceutical interventions, such as securing adequate nutritional status and reducing the risk of falls seem beneficial, although definite associations with fracture risk remains unclear. Pharmaceutical interventions are focused on reducing the bone loss and several approaches have been proven efficient in terms of reducing fracture risk. Pharmaceutical intervention predominately reduces the risk of vertebral fractures in postmenopausal women with osteoporosis. However, effects have also been seen on the risk of other osteoporosis-related fractures in women with a previous vertebral fracture. Vitamin D and calcium substitution is recommended as a compliment to the pharmaceutical intervention (18, 51, 52).

Selective estrogen-receptor modulators (SERM) reduce bone loss by an estrogen agonist effect on bone. In addition, SERM reduce the risk of breast cancer by estrogen antagonist effects on breast tissue. Adverse drug effects include increased risk of deep venous thromboembolism (18, 51, 52).

Bisphosphonates are analogues of pyrophosphate, which binds to hydroxyapatite. They cause osteoclast apoptosis by various mechanisms in the osteoclast after endocytosis, thereby reducing bone resorption. Bisphosphonates can be administered per os or intravenously. Rare

but severe adverse drug effects include osteonecrosis of the jaw (ONJ) and atypical femoral fractures (AFF) (18, 51, 52).

Denosumab is an antibody, which binds to RANKL and thereby inhibits RANKL from binding to the RANK receptor on osteoclasts. The resulting decrement of osteoclast activity reduces bone resorption. Denosumab is administered by subcutaneous injection every 6 months. Events of ONJ and AFF have been reported but the associations remain unclear (18, 51, 52).

Teriparatide is an artificially produced parathyroid hormone, which when given intermittently, increases bone formation by affecting osteoblasts. Teriparatide is administered by daily subcutaneous injections during 18 months. Treatment is contraindicated if the patient has suffered from malignancy or if concurrent metabolic bone disease or severe renal disease is present (18, 51, 52).

2.5 RISK FACTORS FOR FRACTURE

In Sweden, around 20% of all women over the age of 50 have osteoporosis, compared to 7% in men. In total, around 50 000 osteoporosis-related fractures (distal forearm, hip, vertebral) are reported every year (53). Many of these fractures could have been prevented with adequate treatment interventions that lowers fracture risk (18, 51, 52).

The clinical challenges lie in knowing whom to treat. Low aBMD is a strong predictor for subsequent fracture (54-56), and the fracture predictive ability of aBMD has been proposed to be equivalent to that of high blood pressure and the risk of stroke (54). However, many osteoporosis-related fractures occur in women without osteoporosis defined by aBMD (T-score $\leq$ -2,5) (55, 57). Furthermore, aBMD have demonstrated decreasing fracture predictability with age (56).

This suggests that other factors influence fracture risk, especially in the elderly. Consequently, several clinical risk factors have been identified. Table 2 illustrates the most common risk factors associated with fracture independent of aBMD (57-59).

The clinical risk factors have varied significance and are evaluated together with aBMD in determining the individual fracture risk. Consideration of all risk factors is of importance in targeting treatment of patients at highest risk. Integrating algorithms such as FRAX (Fracture risk assessment tool), have therefore greatly improved the clinical evaluation.

Table 2. Independent risk factors for fracture

Age
BMI
History of osteoporosis-related fracture
Parent with a history of hip fracture
Smoking
Alcohol use
Glucocorticoid treatment
Rheumatoid arthritis
Other secondary causes of osteoporosis
Falls

FRAX was developed by the WHO and provides a standardized estimation of the fracture risk based on a combination of validated clinical risk factors, with or without aBMD (60). The result is a 10-year probability of hip fracture or osteoporosis-related fracture. The probabilities are gender and country specific, and the clinical risk factors included in FRAX are the same as depicted in table 2, apart from falls. Although favorable in a clinical setting, there are limitations with FRAX. The gradient of risk that some of the dose dependent risk factors entails, such as the amount of glucocorticoid, alcohol and tobacco use, is not considered. Measures of bone mass other than aBMD, such as volumetric BMD, are not included. Moreover, the risk of falling and indices physical function is not considered (59, 61).

FRAX is widely used as a diagnostic supplement and is recommended by the International Society for Clinical Densitometry (ISCD) and the International Osteoporosis Foundation (IOF) (62). In Sweden, the Swedish National Board of Health and Welfare includes FRAX in the national guideline regarding assessment of fracture risk (63).

2.6 MEASUREMENTS OF BONE MASS AND STRUCTURE

2.6.1 Dual-energy X-ray absorptiometry

The most validated and widely used method to estimate bone mass is by Dual-energy X-ray absorptiometry (DXA), which measures areal bone mineral density (aBMD).

The principle for DXA in measuring bone mass is quantifying the attenuation, i.e. the reduction of photons from an x-ray beam after passing through the body. The attenuation is determined by the density and the thickness of the tissue, higher density causes higher attenuation. By using x-ray beams with two photon energy peaks, it is possible to separate

bone from soft tissue. The result is bone mineral content (BMC) in grams, which divided by the bone area gives an estimation of the two-dimensional areal bone mineral density (aBMD) in g/cm^2. The effective radiation dose varies depending on the region studied, gender, DXA devise and model. In general, the effective dose of a spine DXA ranges between 13-15 µSv (64-66).

Figure 9. Dual-energy X-ray absorptiometry (DXA)

DXA devices require an x-ray generator, a collimator (beam limiting device), a detector, software and a phantom for calibration. Differences in DXA device composition influence the comparability of aBMD derived from different units (64).

Peripheral DXA refers to measurements of the extremities while central DXA refers to measurements of the lumbar spine and hip. Central DXA has been extensively studied with large reference populations, has a relatively high precision and is considered gold standard in clinical settings. Central aBMD derived from central DXA is a strong predictor of fracture and a suitable variable for monitoring treatment or progression. The effective radiation dose is relatively low. By comparison, the natural background radiation is around 6,6 µSv/day (54, 65, 67, 68).

A limitation with DXA lies within the two-dimensional estimation of aBMD; DXA does not indicate the true volumetric BMD. Areal BMD derived from DXA is reported as areal BMD (denoted aBMD) when compared to true volumetric BMD in mg/cm^3 (denoted BMD) derived from devices capable of three-dimensional estimations (2.6.2). Factors that can impair aBMD estimations include degenerative changes in the hip and spine, fractures and implant devises in bone or other tissues, and calcifications in other tissues such as kidney stones and atherosclerosis (69, 70).

2.6.2 Peripheral quantitative computed tomography

Peripheral quantitative computed tomography (pQCT) is a site-specific volumetric method to evaluate bone mass, which allows for a separate evaluation of cortical and trabecular bone.

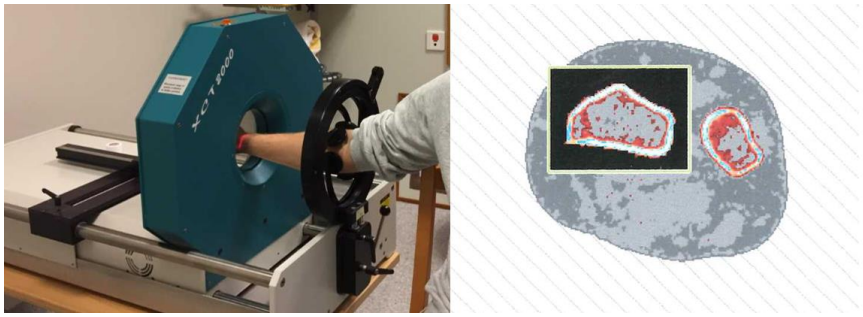


Figure 10. Peripheral quantitative computed tomography (pQCT)

Peripheral quantitative computed tomography is a CT based photon absorptiometry method. In CT images, the amount of attenuation varies depending on the constituents of the tissue. The variation is expressed in Hounsfield Units (HU), which quantifies the constituents by relating to the attenuation of water. The conversion of HU to bone parameters requires calibration with a specific phantom (71, 72). Central QCT refers to measurements of the spine and hip, while peripheral QCT (pQCT) refers to measurements of the radial bone (distal forearm) or tibia (lower leg). As with DXA, the effective radiation dose depends on several variables. Measured effective doses in spine QCT range between 200-1000 µSv, with considerable lower doses in pQCT (<10 µSv) (71, 72).

QCT measures true volumetric BMD in mg/cm^3 and thus provides a more accurate estimation of bone constitution compared to DXA. PQCT distinguishes trabecular bone from cortical bone, which allows for separate analysis of these bone compartments in terms of BMC and BMD. In addition, structural and geometric variables, such as cortical thickness, area and circumference, may be assessed. Furthermore, pQCT has the ability of determining muscle density and area. (71-73).

During a forearm pQCT scan the length of the forearm is initially determined, followed by a coronal scout and the positioning of a reference line, which marks the ulnar side of the radial bone articular surface. Scans are then performed, usually at the distal (4% of the radial bone length) and proximal (60-66% of the radial bone length) parts of the radial bone. These scanning sites are preferred as the distal part of the bone is rich in trabecular bone and the proximal part mainly consists of cortical bone (71, 72).

Forearm pQCT measures has been shown to correlate with subsequent hip and forearm fracture, but not vertebral fracture. On the contrary, spine QCT measures has been shown to correlate with vertebral fracture, but not hip and forearm fracture. However, compared to DXA, QCT measurements are not as widely used and consequently not similarly validated. Few large reference populations are available and differences in scanning procedures, scanning regions, software applications and presented variables impede comparisons between studies. Bone variables from pQCT cannot be applied to the WHO diagnostic classification of osteoporosis, but may be used in monitoring disease and treatment. For these reasons, pQCT is mostly used in research settings and is not widely used among clinicians (25, 71, 72).

3 AIMS OF THE THESIS

The main purpose with this thesis was to identify and further specify factors important in the fracture risk assessment. A deeper understanding of the risk factors for fracture could improve the identification of women at risk.

I. To determine whether women in a study on osteoporosis and risk factors for fracture are representative also for the women that declined participation.

II. To determine whether objective and subjective indices of physical function have a fracture predictive ability in elderly women at different ages.

III. To determine whether smoking and smoking cessation affect fracture risk in elderly women.

IV. To determine whether site-specific musculoskeletal changes in terms of geometry and mass are apparent in women with a distal forearm fracture, and to determine whether PTH and vitamin D affect muscle and bone properties.

V. To determine the prevalence of primary hyperparathyroidism (classical and normocalcemic) in women with a distal forearm fracture.

4 RESEARCH APPROACH

4.1 STUDY POPULATIONS

The studies in this thesis were based on two different populations of elderly women, the OPRA cohort and the DFF cohort.

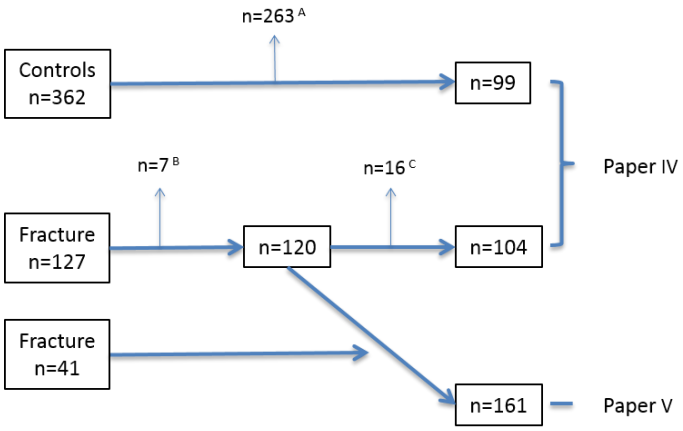
4.1.1 The OPRA cohort

The Osteoporosis Prospective Risk Assessment study (OPRA) was initiated to study risk factors for fractures and osteoporosis in elderly women. The OPRA study is an ongoing population-based study and the cohort consists of elderly women that were recruited at random by the means of population files between the years 1995-1999. During recruitment, women in the city of Malmö, Sweden, received a letter of invitation 1 week after their 75th birthday. In case of no response, reminders were sent by letter or made by telephone. In total, 1604 women were invited. Of the women invited, 1044 accepted participation and were included in the cohort (participants), and 560 women did not participate (non-participants). Out of the 560 non-participants, 376 women were unwilling to partake and 139 women declined because of illness. The remining non-participants were not reached (74).

At baseline, women in the cohort underwent extensive examinations, including blood sampling and DXA. Tests of physical function were performed and the women answered a questionnaire. The procedure was repeated at the 5-year follow-up and at the 10-year follow-up. At the age of 85, the women in addition underwent a pQCT examination of the forearm.

4.1.2 The DFF cohort

The Distal Forearm Fracture study (DFF) was initiated at Karolinska University Hospital, Stockholm, Sweden. Postmenopausal women who sought the emergency ward with a distal forearm fracture between April 2010 and January 2015 were invited to participate. Criteria for participation included fracture due to low-energy trauma and that they were past menopause. No exclusion criteria were applied. The study procedure involved a blood sample at the emergency ward and an invitation to the research facility where they answered a questionnaire and left a supplementary blood sample and, if needed, underwent further examinations. After recruitment of all patients to Paper IV, patients were recruited only to Paper V (Figure 11).



A No response or coincident disease/fracture/treatment
B Declined
C Previous fracture to the other arm or missing data

Figure 11. The recruitment process in Paper IV and V

To the study group in Paper IV, 127 women accepted participation at the emergency ward. Out of these, 7 women were excluded because of unwillingness. The women were examined with DXA and pQCT in addition to the previously mentioned screening procedure (blood screening, questionnaire, supplementary sample). Subsequently, 16 women were withdrawn from analysis because pQCT measurements were conducted on a previously fractured arm (n=13) or missing data (n=3). In total, 104 women with a mean age of 64 were included. Age-matched controls were recruited from the population register. Letters of invitation were sent to 362 women with one reminder if needed. Controls with a known bone remodeling disease, history of an osteoporosis-related fracture, antiresorptive-, estrogen- or oral glucocorticoid treatment were excluded. In all other regards, the examination protocol was identical in cases and controls. In total, 99 controls with a mean age of 65 were included. The total mean age (both cases and controls) in Paper IV was 65 years.

To the study group in Paper V, another 41 patients were recruited to evaluate the PHPT prevalence. In total, 161 women were included in Paper V, all of whom underwent a procedure with blood screening, questionnaire and supplementary sample followed by a complete biochemical and physical examination at the department of Endocrinology. Repeated evaluations were performed if needed.

4.2 DATA COLLECTION

4.2.1 Mortality

Mortality was registered in the OPRA cohort by cross-referencing the 10-digit personal identification number against the population registry. Mortality data were registered for all women in the OPRA cohort, participants as well as non-participants.

4.2.2 Fractures

Fractures in the OPRA cohort were continuously registered by collecting radiology files from the Department of Radiology at Skåne University Hospital using the personal identification number. The radiological files from the hospital can be regarded as near complete, with less than 3% loss (75). In case of uncertainties in the radiology files, the radiology images were reviewed. This was the sole means of fracture registration in Paper I; to avoid discrepancies between participants and non-participants. In Papers II-IV, fracture registration through radiology files was complemented with questionnaire information.

4.2.3 Questionnaire

Participants in the OPRA study and DFF study answered an identical comprehensive questionnaire regarding diet, lifestyle, smoking, physical activity, history of falls, fracture, disease, medication and heredity. Information on smoking habits included: current or former smoker, time as smoker and amount smoked.

4.2.4 Physical performance tests

Gait speed, balance test and knee extension force were performed by the OPRA study participants at baseline (age 75) and at follow-up (age 80). Three attempts of each test were made and the best was registered. One of three physiotherapists supervised and the approach was identical for all participants.

Gait speed was evaluated by the time it took to walk 15 meters back and forth and the women were urged to walk as fast as they could. The balance test was evaluated by the ability to stand on either the left or right leg with eyes open. The test proceeded for 30 seconds. Managing to stand on one leg for more than 5 seconds was considered a passed test. Knee extension force was measured as isometric muscle strength with the knee in 90 degrees of flexion in a dynamometer (Biodex Medical Systems, Version 4.5.0, Biodex Corporation, Shirley, N.Y., USA). Participants in the DFF study did not perform physical performance tests.

4.2.5 Parathyroid hormone and Vitamin D

In Paper IV, PTH and 25-hydroxy vitamin D (25OHD) were analyzed from frozen serum samples by the same laboratory and methodology at Skåne university hospital in Lund (both OPRA and DFF cohort). In Paper V, PTH and 25OHD were analyzed at the Karolinska University Hospital. Serum calcium was corrected for albumin by the formula: Calcium + 0.01 x (39-albumin).

4.2.6 Peripheral Quantitative Computed Tomography

Structural and material properties of the forearm were assessed by peripheral Quantitative Computed Tomography (pQCT). Bone variables assessed were: BMD, BMC, cross-sectional area (CSA), cortical thickness, endosteal and periosteal circumference. Bone strength was assessed by strength strain index (SSI) and bone strength index (BSIc), defined as; total BMD^2 * CSA at the distal 4% site (76).

Stratec XTC-2000 (Stratec Medizintechnik, Pforzheim, Germany) with software version 6.2 provided by the manufacturer was used in both cohorts (OPRA and DFF). The European Forearm Phantom (EFP) was used for calibrations in both cohorts and calibrations were performed for each subject. CV% for the forearm phantom was 0.27. The EFP was tested in both devices by the manufacturer with a Pearson correlation coefficient of 1.00. Measurements included distal radius and radius shaft (4% and 66% of ulnar length proximal to the reference line). Voxel size was 0.5 mm, slice thickness 2.3 mm and scanning speed 20 mm/s. To separate bone from soft tissue, a 180 mg/cm^3 density threshold was applied at the 4% site and a 280 mg/cm^3 threshold at the 66% site. Trabecular bone was evaluated at the 4% site and cortical bone at the 66% (72). 45% of the area at the distal (4%) site was determined as trabecular bone. An inner threshold of 711 mg/cm^3 differentiated cortical bone at the shaft (66%) site. A density threshold of 40 mg/cm^3 distinguished muscle tissue at the 66% site.

4.2.7 Dual energy X-ray Absorptiometry

Areal bone mineral density was determined by Dual energy X-ray Absorptiometry *(*DXA). In the DFF cohort, measurements included lumbar spine, femoral neck, ultra-distal radius (UDR) and 33% of radius length. In the OPRA cohort, measurements included lumbar spine, femoral neck and arm. In the lumbar spine, measurements of the first and second lumbar vertebrae (L1-L2) were chosen to minimize artifacts by degenerative changes (77). The left femoral neck was used for analysis. Calibrations were made with a spine phantom three times every week and automatic calibrations were made daily.

In the DFF cohort, measurements were performed with GE Lunar iDXA (GE Medical systems, Chalfont St. Giles, UK). A correlation coefficient (CV) of 1,5% was determined for the phantom. In the OPRA study, measurements were performed with Lunar DPX-L (Lunar Corporation, Madison, Wi, USA). CV was 1,4% for the phantom.

4.2.8 Site-specific measurements by pQCT and DXA

In the DFF cohort, measurements were performed at the non-fractured forearm with a matching number of sides in the controls. There was no difference between cases and controls regarding right- or left-handedness. The left arm was examined in the OPRA cohort except in three cases. Women with a previous fracture at the examined arm, missing or otherwise invalid measurements were excluded in the analysis.

4.3 STUDY OUTLINE

4.3.1 Paper I

Mortality and fracture rates in the 1044 participants and 560 non-participants in the OPRA study were compared.

Mortality was compared with Kaplan-Meier curves and log-rank test. Fracture incidence was compared with the Mann-Whitney U-test. Cumulative fracture incidence with consideration of competing risks was compared with Grey´s test.

4.3.2 Paper II

The fracture predictive ability of physical function tests (gait speed, standing balance, knee extension force and self-reported history of falls) was evaluated in the 1044 women participating in the OPRA study. Participants were examined at inclusion with repeated examinations after 5 years at the age of 80. The fracture predictive ability of each test during 10 years of follow-up was determined. In addition, the fracture predictive ability at the age of 75 was compared to the fracture predictive ability at the age of 80.

Standing balance and history of falls were dichotomized. Univariate and multivariate hazard ratios with 95% CI, adjusted for known risk factors for fracture, were determined with univariate and multivariate Cox proportional hazards regression (with BMD, BMI, previous fracture, smoking, alcohol-, vitamin D-, glucocorticoid- and bisphosphonate use as covariates).

4.3.3 Paper III

Fracture rates in non-smokers, current smokers and former smokers during 10 years of follow-up in the OPRA study were determined. The fracture rate in current (n=145) and former smokers (n=209) was compared to the rate in non-smokers (n=679). Time as a smoker, amount smoked and time from cessation were analyzed in relation to fracture risk.

Univariate and multivariate cumulative fracture incidence with consideration of competing risks was estimated with univariate and multivariate proportional hazards regression.

4.3.4 Paper IV

Site-specific volumetric material and structural properties of bone and muscle in the forearm was determined by pQCT. Measurements were performed in the DFF cohort (mean age 65) and in a subset of the OPRA cohort (mean age 85). Women with a distal forearm fracture were compared to age-matched controls without fracture within each cohort. Furthermore, the correlation of PTH and 25OHD with volumetric bone and muscle variables was assessed.

Comparisons between cases and controls were made with independent t-tests and analysis of covariance (ANCOVA). In a separate ANCOVA, volumetric pQCT variables were adjusted for site-specific aBMD by DXA. Correlations of PTH and vitamin D with bone and muscle variables were assessed for all women (regardless of fracture status), with multiple linear regression.

4.3.5 Paper V

The prevalence of PHPT was determined by clinical and biochemical examination of 161 women participating in the DFF study. PHPT diagnosis was based on repeated evaluations of PTH and calcium levels in the absence of secondary causes. The biochemical definition of PHPT was based on combinations of PTH and calcium levels, designed with a similar approach as has been reported previously in population prevalence reports. PHPT was subdivided into “classical PHPT” and “normocalcemic PHPT”. Elevated PTH (> 65 ng/L) in combination with elevated serum calcium (> 2.50 mmol/L) was considered classical PHPT. Elevated PTH (> 65ng/L) in combination with serum calcium within, but in the upper part of the reference interval (2.15-2.50 mmol/L), was considered normocalcemic PHPT.

Comparison of PHPT prevalence with population prevalence was made with a binominal two-tailed exact test.

4.4 STATISTICAL CONSIDERATIONS AND POWER

No calculation of power was performed before analysis in Paper I-III, since the sample size was deemed sufficient for the planned analyses.

In Paper IV the following assumption was made: To find a difference in bone density of 10%, i.e. 20 mg/cm^3 at a standard deviation of 45 mg/cm^3, 81 patients and 81 controls ($\alpha = 0.05$ and power 80%) would be needed. If the case-control ratio was 1:4, 53 cases and 159 controls would be needed.

In Paper IV the following assumption was made: Previous population reports have indicated a population prevalence of PHPT around 2%. In our study group, we have previously found prevalence around 6% in postmenopausal women with a distal forearm fracture (78). Considering that the estimated prevalence in the study population is 0.06, it follows that 141 patients are required to find a difference ($\alpha = 0.05$ and power 80%).

A p-value < 0.05 was considered as level of significance in all papers.

4.5 ETHICAL APPROVAL

All parts of the studies were performed with ethics committee approval and informed consent.

Paper I-II. Approved by the Ethics committee of Lund University (LU 200-95).

Paper III. Approved by the Ethics committee of Lund University (LU 200/95).

Paper IV. Approved by the Regional ethical boards in Stockholm and Lund, and the Ethics committee of Lund University (2009/913-31, 2014/804, LU 200-95).

Paper V. Approved by the Regional ethical board in Stockholm (2009/913-31).

4.5.1 Ethical considerations

Increased identification of patients at risk could lead to preventive measures initiated at an earlier state, thus reducing fracture rates. A prerequisite for this is that there are effective and acceptable treatment options for osteoporosis available. Osteoporosis itself is a silent disease, but its consequence, fracture, is associated with increased mortality and mortality. In addition, the socioeconomic consequences are vast. Prevention of fractures could benefit not only the affected patient, but also society at large.

Hyperparathyroidism is proposedly an underdiagnosed condition that, besides osteoporosis, can lead to vague symptoms like fatigue, muscle aches, constipation and kidney stones.

Presuming an increased prevalence of primary hyperparathyroidism in postmenopausal women with forearm fracture, as previously indicated (78, 79), the event of fracture might be an opportunity to identify undiagnosed individuals. This also assumes available and acceptable treatment options.

All study participants and controls in the OPRA and DFF study received information of the planned investigations and risks and gave informed consent. Uncovered diseases and fractures were treated per clinical routines. Participants could at any time, without reason, choose to cancel and withdraw their participation. Records and data files were treated with confidentiality.

A blood sample was taken which may cause discomfort and in rare occasions side effects such as local infections. Radiation doses with DXA and pQCT are generally low (equivalent to less than a standard chest x-ray).

In Paper I, participants and non-participants were studied. We studied fracture rate and mortality through radiological files and population files solely. It involved no risk or inconvenience for the women and the data were not presented on an individual basis. Nevertheless, an ethical dilemma was recognized; informed consent from non-participants was not obtained. This would have been practically difficult since the study is based on comparing the very ones who declined participation. We considered these aspects and concluded that these women would not suffer from the "non-participation". From a utilitarian perspective, it would be beneficial to complete the study.

5 RESULTS

5.1 PAPER I

Mortality was higher in non-participants; 372 out of 560 (66%), when compared to 454 out of 1044 (44%) in participating women (Figure 12). Vertebral and distal forearm fracture incidence rates were lower in non-participants, when compared to participants (IR: 10 vs 13 and 7 vs 11 respectively, $p \leq 0.007$). Multiple fractures were less common among non-participants (IR: 14 vs 20, $p \leq 0.001$). Other specific fractures (proximal humerus, pelvis, hip) and fracture groups (osteoporosis-related, any) were without significant difference between non-participants and participants.

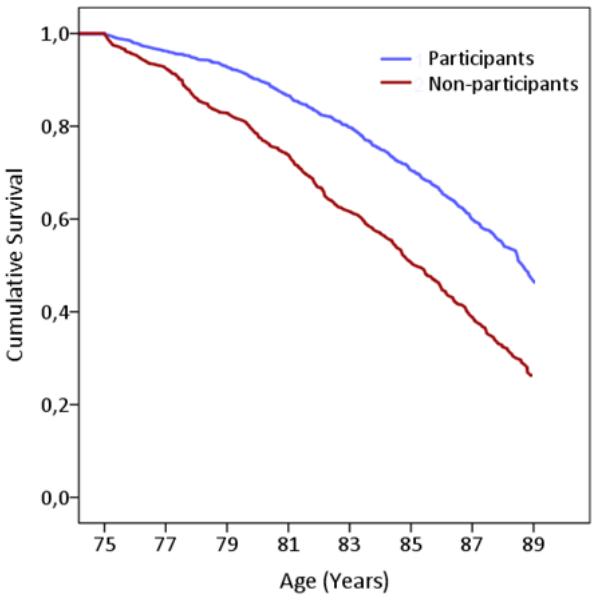


Figure 12. Cumulative survival in participants and non-participants in the OPRA cohort (Paper I)

5.2 PAPER II

A history of fall corresponded to an increased risk of distal forearm fracture (HR 1.60, CI 1.03-2.48) any fracture (HR 1.30, CI 1.03-1.65) and any osteoporosis-related fracture (HR 1.39, CI 1.08-1.79) in the 10-year follow-up after adjustment for possible confounding variables. Decreased gait speed (HR 1.37, CI 1.14-1.64) and a failed balance test (HR 1.98, CI 1.18-3.32) was associated with increased risk of hip fracture. Knee extension force was not associated with fracture risk in the 10-year follow-up.

In the first 5-year follow-up from the age of 75, a history of fall, decreased gait speed and failed balance test corresponded to an increased risk of hip fracture. In addition, a history of multiple falls was associated with increased risk of any osteoporosis-related fracture, whereas decreased gait speed and knee extension force was associated with increased clinical vertebral fracture risk.

In the second 5-year follow-up from the age of 80, a history of fall corresponded to increased risk of hip fracture, any fracture and any osteoporosis related fracture. Reduced gait speed was associated with increased risk of hip fracture.

5.3 PAPER III

When compared to non-smokers, current smokers had an increased risk of contracting any osteoporosis-related fracture (HR 1.47, CI 1.05-2.05) and vertebral fracture (HR 2.50 CI 1.58-3.95) after adjustment for possible confounding variables. Former smokers had an

increased risk of contracting proximal humerus fracture (HR 2.07, CI 1.19-3.57). Increased risk for other fracture types were not observed in current and former smokers after adjustment for possible confounding variables.

Time as a smoker was associated with increased risk of vertebral fracture (HR 2.66, CI 1.57-4.53) and decreased risk of distal forearm fracture (HR 0.38, CI 0.17-0.86), whereas amount smoked did not affect fracture rates. Time from cessation did not affect fracture rate in former smokers.

5.4 PAPER IV

In the DFF cohort, women with a distal forearm fracture had decreased central aBMD, increased time spent walking/day and more frequently reported a history of falls, compared to controls. These variables did not differ in the OPRA cohort.

Compared to controls, women with a distal forearm fracture in the DFF cohort had decreased forearm volumetric mean trabecular BMD (167.5 vs 125.1, $p < 0.001$), cortical BMD (1120.8 vs 1095.7, $p= 0.007$), total BMC at the 66% site (89.8 vs 81.4, $p < 0.001$) and BSIc (30.8 vs 22.1, $p < 0.001$) after adjustments for possible confounding variables. In addition, mean CSA (319.8 vs 336.4, $p= 0.033$) and endosteal circumference (27.8 vs 30.1, $p < 0.009$) was greater while cortical thickness was lower (1.996 vs 1.726, $p < 0.001$) in women with fracture. Periosteal circumference, Strength strain index and muscle parameters did not differ.

After additional adjustment for site specific aBMD by DXA, a difference in trabecular BMD, total BMC at the 66% site and cortical thickness remained.

In the OPRA cohort, differences in volumetric trabecular BMD, total BMC at the 66% site and BSIc was similar to those found in the DFF cohort. No difference in structural (geometric) parameters were observed in the OPRA cohort, between women with fracture and controls.

Mean PTH and 25OHD did not differ between women with and without a forearm fracture. In the DFF cohort, weak negative correlations were observed between PTH and site specific volumetric trabecular and cortical BMD as well as aBMD of the femoral neck. Weak positive correlations were observed between 25OHD and site specific volumetric trabecular BMD and aBMD of the lumbar spine. Significant correlations were absent in the OPRA cohort, except for a weak positive correlation between PTH and aBMD of the femoral neck.

5.5 PAPER V

Of the 161 women with a distal forearm fracture, 13 women (8%) were diagnosed with PHPT. Classical PHPT were observed in 7 women and normocalcemic PHPT in 6 women. In total, 6 women presented with symptomatic disease (3 with classical PHPT and 3 with normocalcemic PHPT). Osteoporosis (T-score < -2.5) were observed in 3 women. The observed PHPT prevalence in the 161 women with a distal forearm fracture was significantly greater than a population prevalence of 3.4% (p=0.004).

In addition, 32 women (20%) presented elevations of PTH for reasons other than PHPT.

6 DISCUSSION

External validity

When identifying risk factors in epidemiological research, it is important that the results are generalizable. In population based studies, external validity can be assessed by comparing the characteristics of participating individuals with non-participants, i.e. individuals that decline participation or for other reasons are unable to participate. If the non-participants differ substantially from the participants, the external validity is reduced which might induce non-response bias into the study. Thus, the external validity is not solely dependent on a high response rate. Knowledge of the non-participating individuals is essential when interpreting and correcting for non-response bias (80, 81). Most studies are dependent on voluntary, active participation and for understandable reasons prospective information on the characteristics of non-participants is poorly described.

The finding of increased mortality among non-participating women in the OPRA cohort is in line with findings from the EVOS study on vertebral fractures (82), as well as studies from other disease areas (81, 83-90). Indicating a poorer health status in non-participating women compared to participating women.

However, this did not seem to entail increased fracture rates in non-participants. On the contrary, multiple fractures, distal forearm fractures and vertebral fractures were slightly more frequent in participating women. Analysis of fracture incidence in participants and non-participants prior to inclusion have suggested that studies on osteoporosis and fracture might attract individuals prone to fractures (82, 91, 92), which in part might explain the seemingly increased fracture rates. A longer life-span and thus a longer time at risk for fracture might also contribute to some of the observed differences.

The rate of hip fracture and proximal humerus fracture did not differ between groups although an increased hip fracture rate among non-participants has been suggested previously (93). The finding of increased clinical vertebral fracture rate was somewhat in line with a previous study in which participants more frequently reported back pain, although, radiographic imaging were not used (92). Compared to others, our study had the advantage of being able to follow a large population for a long period. In terms of mortality and fracture rates, the OPRA cohort can be regarded as almost complete.

Physical performance tests and history of falls as clinical risk factors for fracture

In a clinical setting, establishing the future fracture risk is essential in determining the level of preventive interventions. It has been suggested that an impaired postural stability may be associated with an increased fracture risk, but measures of postural stability are not included in the most widely used tool for fracture risk assessment, FRAX. Postural stability can be evaluated by the occurrence of previous falls or through physical performance tests, e.g. balance, gait speed and lower extremity muscle strength.

The most commonly described measure of postural stability, *self-reported history of falls*, has been associated with increased risk of any fracture (94-98), hip fracture (99-101), distal forearm fracture (102-104) and proximal humerus fracture (105), although not all concur (102, 103, 106). *Reduced gait speed* have predominately been associated with increased risk of hip fracture (99, 107, 108) and a *failed standing balance tests* with any fracture (109), proximal humerus fracture (102, 105) and hip fracture (108), while *decreased lower extremity muscle strength* have shown some predictive value in terms of any fracture (109) and hip fracture (100). However, long-term prospective data is sparse and several studies have failed to demonstrate associations (95, 102, 103, 106, 107). The fracture predictive ability of these measures remains unclear.

A self-reported history of fall was during 10-years of follow up in the OPRA cohort associated with an increased risk of osteoporosis-related fracture, any fracture and distal forearm fracture. In general, history of multiple falls was associated with greater risks of fracture. However, we were not able to reproduce hip and proximal humerus fracture predictability as previously suggested (99-101, 105).

Reduced gait speed was a strong predictor for future hip fracture in our study. Associations were also observed with several other fracture types in the univariate analysis, but these associations were lost after adjustments for aBMD and other possible confounders.

A failed balance test at the age of 75 doubled the hip fracture risk which in the OPRA cohort was comparable to the fracture predictability of a standard deviation decrease in femoral neck aBMD. This finding was in line with other studies examining balance by postural sway (100) and tandem walk (107). Although not all concur (106), our findings have since been strengthened by the results by Lundin et al who demonstrated increased hip fracture risk with failing a one-leg standing balance test (108). A failed balance test did not increase the risk of other fractures in the OPRA cohort, even though this has been previously proposed (102, 105, 109).

Knee extension weakness did not seem to increase fracture risk after adjustments for femoral neck aBMD. However, some associations were seen in the univariate analysis, suggesting that some of the fracture predictability of knee extensor strength might be related to aBMD.

The balance test seemed to lose predictability with age as it was predictive of hip fracture at the age of 75, but not at the age of 80. On the other hand, gait speed and self-reported history of at least one fall were predictive of hip fractures in both 75-year old and 80-year old women.

These tests that evaluate the physical function are easy to implement and require no complex equipment, which are considerable benefits in a clinical environment.

Smoking, smoking cessation and fracture risk

Smoking has previously been associated with increased fracture risk and the dose as well as duration of exposure seem to affect the risk (110-112). However, reports showing a lack of association with hip fractures (99, 113, 114) and any osteoporosis-related fractures (115) have been presented in smokers. It has been indicated that the effect of smoking on bone mass and fracture risk is reversible (110, 112, 115), but a lack of reversible effect in terms of hip fractures have been shown (116).

Smokers in the OPRA cohort had an increased risk of fracture, and the risk of vertebral fracture was particularly increased. A biological effect of smoking on bone remodeling has been proposed (117), in line with our findings, suggesting a greater effect of smoking on trabecular bone with a higher remodeling rate, compared to cortical bone. Former smokers also had an increased risk of fracture, but not for vertebral fracture, suggesting reversible effects of smoking cessation primarily on skeletal sites comprised of trabecular bone.

The hip fracture risk was not increased in smokers in the OPRA cohort, although an association have been reported by several authors (112, 118, 119). Furthermore, a previous report on the OPRA cohort have shown decreased hip aBMD in smokers, which ought to increase the susceptibility for fracture (74). However, in a recent meta-analysis, associations between smoking and hip fracture risk were not found in most studies with elderly participants (112). The effect of smoking on hip fracture risk may be dependent on age; in younger populations, smoking seemed to increase hip fracture risk (112, 115). It is plausible that other risk factors for hip fracture are more prominent than smoking and aBMD in the elderly.

Smoking cessation did not affect the risk of any fracture which has previously been proposed

(112, 119). The effects of smoking on the risk of any and any osteoporosis-related fracture seemed to last for a long time, further emphasized by the lack of risk reduction with time from cessation. In addition, it appeared as if the risk of fracture was independent of the amount smoked.

Site-specific structural and material properties in elderly women with forearm fracture

Distal forearm fractures most often occur in younger postmenopausal women and the event of a distal forearm fracture may be the first sign of osteoporosis. Bone strength is dependent on the structural properties (geometric) and the material properties (mineralization and density) of the bone as well as the proportion of cortical and trabecular bone. Evaluation of these properties are important in understanding the pathogenesis of fracture.

Decrement of aBMD by DXA of the hip and forearm have shown an association with increased forearm fracture risk (54, 103, 120-122), but aBMD only partially evaluate aspects of bone strength (72). Using pQCT, geometric and material changes of the forearm have been proposed as causative factors in mechanical in vitro studies (72, 123, 124). However, clinical associations are sparse and with either a small sample size (121, 125-128) or retrospective approach (120, 129).

Findings in the DFF study suggests that both forearm trabecular and cortical bone decrements are present in postmenopausal women with a distal forearm fracture, even though the trabecular reductions seemed to be more prominent. Although not all concur (127), these findings are in line with previous reports (120, 121, 125, 126, 129).

Geometric changes were also observed. Women with fracture had decreased cortical thickness and increased endosteal circumference as well as cross sectional area. While a thinner cortex with increased diameter has been associated with ageing (72, 130-132), increasing size has not been demonstrated in previous studies in women with fracture (120, 121, 125-128). In normal ageing, it has been proposed that the cross-sectional area increases as a response to decrements of bone mass, providing a favorable structure in terms of bone strength (72, 130-132). The increased area in women with fracture might thus be an effect of the lower bone mass. However, these geometric alterations appeared insufficient in terms of maintaining bone strength in the women with a forearm fracture. Strength index, which accounts for both bone mass and geometry, has previously been associated with decreased bone resilience (130). However, the stress-strain index in our study was surprisingly not changed.

Site-specific aBMD by DXA was also lower among women with fracture and when adjusted for aBMD, some of the volumetric parameters lost significance. However, changes in volumetric trabecular BMD as well as cortical BMC and thickness persisted, indicating that volumetric changes not visualized by DXA were present.

Women with a forearm fracture in the DFF cohort had on average a higher activity level and more frequently reported a history of fall. Central aBMD of the femoral neck and aBMD of the lumbar spine were, in addition, lower among women with fracture compared to controls. These differences were not seen in a subset of women at the age of 85 in the OPRA cohort. In the OPRA cohort, volumetric changes were less apparent, apart from trabecular BMD. This suggests that other factors are of importance in the pathogenesis of forearm fracture in the elderly.

Parathyroid hormone disturbances in postmenopausal women with forearm fracture

Increased levels of PTH have been associated with decreased BMD in women with parathyroid disease and the reductions seems to be a result of both cortical and trabecular bone loss (133-137). In Paper IV, neither PTH nor 25OHD were associated with forearm fracture risk, but weak correlations of PTH and 25OHD with volumetric and areal BMD were observed.

Women with PHPT have been shown to be at risk of fracture (33, 40) and conversely, there have been suggestions of increased PHPT prevalence among women with fracture (78, 79). The high prevalence of PHPT in Paper V are in line with previous findings in women with a distal forearm fracture (78, 79), and other fractures (138-141). However, a lack of association has also been presented (142). Differences in study design and biochemical definition of PHPT complicates comparisons between studies and population prevalence. Nevertheless, by thoroughly examining 161 women with a distal forearm fracture we observed a substantial number of women with PHPT and other parathyroid hormone disturbances. Even though many of the women with PHPT in this study were mild cases, identifying these would be of value as the disease may progress and surveillance seems beneficial (32, 38, 143).

6.1 LIMITATIONS

Although performed according to a standardized protocol, personality traits of the physiotherapists who conducted the physical performance tests in Paper II might have influenced the results. Smoking habits were dependent on recall and personal declaration which may be a source of misclassification in Paper III. In the DFF study, the recruitment

rate was slow due to interruptions during the recruitment process, caused by personnel and workplace changes at the emergency ward. Although we did not consider the rate of refusal as high, this might be a source of bias. Comparisons between the DFF and OPRA cohort in Paper IV might be affected by differences in design and setup, and by the relatively small number of women with fracture and valid pQCT measurements in the OPRA cohort. In Paper V, the PHPT diagnosis was determined by repeated evaluations and a predetermined biochemical definition, confirmed by endocrinologists without affiliation to the study. However, this approach may be a source of selection bias. A major limitation is the lack of a control-group in this study. Validation of the findings in studies with a control group would be beneficial.

6.2 CONCLUSIONS

Several factors influence the risk of fracture in elderly women, and all factors need to be considered in order to perform an adequate fracture risk assessment. Mortality was increased among non-participants in the OPRA cohort when compared to those who participated, but it appeared that participants were representative in terms of fracture risk in general. Consideration of physical function and smoking habits appears to be advantageous in assessing fracture risk. Self-reported history of falls seems to be associated with increased risk of several fracture types, while reduced gait speed and a failed balance test appears to be associated with hip fracture. Smoking increases the risk of osteoporosis-related fractures, predominately vertebral fractures. Smoking cessation seems to decrease the risk of vertebral fractures but the risk of other fractures appears to be unaffected by smoking cessation. Trabecular and cortical bone reductions, as well as geometric alterations of the forearm, seems to be contributing factors in the pathogenesis of a distal forearm fracture. PTH and 25OHD show signs of being weakly correlated with trabecular and cortical bone in the forearm. The prevalence of PHPT among postmenopausal women with a forearm fracture might be elevated and further evaluation following a distal forearm fracture would appear to be beneficial.

7 ACKNOWLEDGEMENTS

I would like to acknowledge my principal supervisor Paul Gerdhem and co-supervisor Ingrid Bergström for all the hard work and support. I also would like to acknowledge the following: research nurse Ewa Steninger, DXA operators Ninni Qvist and Karin Björklund, statistician Per Näsman, co-writers Mats Holm Thorin, Karin Bergström and Kristina Åkesson for help with the studies and Mats Palmer for proofreading. I would like to acknowledge all colleagues and staff involved with recruitment and care of participants at the emergency ward, Department of Orthopedics and Department of Endocrinology. Thank you for all the hard work. Last but not the least I would like to acknowledge all the women in the studies.

This thesis was made possible by financial support by:
Medical Training and Research Agreement (ALF) funds.
Karolinska Institutet research funds.

8 REFERENCES

1. Clarke B. Normal Bone Anatomy and Physiology. Clinical Journal of the American Society of Nephrology. 2008;3:S131-S9.

2. Taichman RS. Blood and bone: two tissues whose fates are intertwined to create the hematopoietic stem-cell niche. Blood. 2005;105(7):2631-9.

3. Brodsky B, Persikov AV. Molecular structure of the collagen triple helix. In: Parry DAD, Squire JM, editors. Fibrous Proteins: Coiled-Coils, Collagen and Elastomers. Advances in Protein Chemistry. 70. San Diego: Elsevier Academic Press Inc; 2005. p. 301-+.

4. Rubin CT, Lanyon LE. Osteoregulatory nature of mechanical stimuli - function as a determinant for adaptive remodeling in bone. Journal of Orthopaedic Research. 1987;5(2):300-10.

5. Bonewald LF. Establishment and characterization of an osteocyte-like cell line, MLO-Y4. Journal of Bone and Mineral Metabolism. 1999;17(1):61-5.

6. Vaananen HK, Zhao H, Mulari M, Halleen JM. The cell biology of osteoclast function. Journal of Cell Science. 2000;113(3):377-81.

7. Parfitt AM. Targeted and nontargeted bone remodeling: relationship to basic multicellular unit origination and progression. Bone. 2002;30(1):5-7.

8. Carmeliet G, Van Cromphaut S, Daci E, Maes C, Bouillon R. Disorders of calcium homeostasis. Best Pract Res Clin Endocrinol Metab. 2003;17(4):529-46.

9. Brini M, Ottolini D, Cali T, Carafoli E. Calcium in health and disease. Met Ions Life Sci. 2013;13:81-137.

10. Potts JT. Parathyroid hormone: past and present. Journal of Endocrinology. 2005;187(3):311-25.

11. Mannstadt M, Juppner H, Gardella TJ. Receptors for PTH and PTHrP: their biological importance and functional properties. Am J Physiol. 1999;277(5 Pt 2):F665-75.

12. Usdin TB, Bonner TI, Hoare SRJ. The parathyroid hormone 2 (PTH2) receptor. Receptors & Channels. 2002;8(3-4):211-8.

13. Findlay DM, Sexton PM. Calcitonin. Growth Factors. 2004;22(4):217-24.

14. Nair R, Maseeh A. Vitamin D: The "sunshine" vitamin. J Pharmacol Pharmacother. 2012;3(2):118-26.

15. Basit S. Vitamin D in health and disease: a literature review. British Journal of Biomedical Science. 2013;70(4):161-72.

16. Holick MF. High prevalence of vitamin D inadequacy and implications for health. Mayo Clinic Proceedings. 2006;81(3):353-73.

17. Javaid MK, Cooper C. Prenatal and childhood influences on osteoporosis. Best Practice & Research Clinical Endocrinology & Metabolism. 2002;16(2):349-67.

18. Drake MT, Clarke BL, Lewiecki EM. The Pathophysiology and Treatment of Osteoporosis. Clinical Therapeutics. 2015;37(8):1837-50.

19. Seeman E. Pathogenesis of bone fragility in women and men. Lancet. 2002;359(9320):1841-50.

20. Sambrook P, Cooper C. Osteoporosis. Lancet. 2006;367(9527):2010-8.

21. Riggs BL, Melton LJ, Robb RA, Camp JJ, Atkinson EJ, Peterson JM, et al. Population-based study of age and sex differences in bone volumetric density, size, geometry, and structure at different skeletal sites. Journal of Bone and Mineral Research. 2004;19(12):1945-54.

22. Khosla S, Riggs BL. Pathophysiology of age-related bone loss and osteoporosis. Endocrinology and Metabolism Clinics of North America. 2005;34(4):1015-+.

23. NIH Consensus Development Panel on Osteoporosis Prevention D, and Therapy. Osteoporosis prevention, diagnosis, and therapy. Jama. 2001;285(6):785-95.

24. Kanis JA, Melton LJ, Christiansen C, Johnston CC, Khaltaev N. Perspective - the diagnosis of osteoporosis Journal of Bone and Mineral Research. 1994;9(8):1137-41.

25. Shepherd JA, Schousboe JT, Broy SB, Engelke K, Leslie WD. Executive Summary of the 2015 ISCD Position Development Conference on Advanced Measures From DXA and QCT: Fracture Prediction Beyond BMD. J Clin Densitom. 2015;18(3):274-86.

26. Holick MF, Binkley NC, Bischoff-Ferrari HA, Gordon CM, Hanley DA, Heaney RP, et al. Evaluation, Treatment, and Prevention of Vitamin D Deficiency: an Endocrine Society Clinical Practice Guideline. Journal of Clinical Endocrinology & Metabolism. 2011;96(7):1911-30.

27. Mosekilde L. Vitamin D and the elderly. Clinical Endocrinology. 2005;62(3):265-81.

28. Mosele M, Coin A, Manzato E, Sarti S, Berton L, Bolzetta F, et al. Association Between Serum 25-Hydroxyvitamin D Levels, Bone Geometry, and Bone Mineral Density in Healthy Older Adults. Journals of Gerontology Series a-Biological Sciences and Medical Sciences. 2013;68(8):992-8.

29. Fitzpatrick LA. Secondary causes of osteoporosis. Mayo Clinic Proceedings. 2002;77(5):453-68.

30. Canalis E, Mazziotti G, Giustina A, Bilezikian JP. Glucocorticoid-induced osteoporosis: pathophysiology and therapy. Osteoporos Int. 2007;18(10):1319-28.

31. van Driel M, van Leeuwen JP. Vitamin D endocrine system and osteoblasts. Bonekey Rep. 2014;3:493.

32. Macfarlane DP, Yu N, Leese GP. Subclinical and asymptomatic parathyroid disease: implications of emerging data. Lancet Diabetes & Endocrinology. 2013;1(4):329-40.

33. Mosekilde L. Primary hyperparathyroidism and the skeleton. Clinical Endocrinology. 2008;69(1):1-19.

34. Lundgren E, Rastad J, Thurfjell E, Akerstrom G, Ljunghall S. Population-based screening for primary hyperparathyroidism with serum calcium and parathyroid hormone values in menopausal women. Surgery. 1997;121(3):287-94.

35. Lundgren E, Hagstrom EG, Lundin J, Winnerback K, Roos J, Ljunghall S, et al. Primary hyperparathyroidism revisited in menopausal women with serum calcium in the upper normal range at population-based screening 8 years ago. World Journal of Surgery. 2002;26(8):931-6.

36. Jorde R, Bonaa KH, Sundsfjord J. Primary hyperparathyroidism detected in a health screening: The Tromso Study. Journal of Clinical Epidemiology. 2000;53(11):1164-9.

37. Hagstrom E, Lundgren E, Mallmin H, Rastad J, Hellman P. Positive effect of parathyroidectomy on bone mineral density in mild asymptomatic primary hyperparathyroidism. Journal of Internal Medicine. 2006;259(2):191-8.

38. Bilezikian JP, Brandi ML, Eastell R, Silverberg SJ, Udelsman R, Marcocci C, et al. Guidelines for the management of asymptomatic primary hyperparathyroidism: summary statement from the Fourth International Workshop. J Clin Endocrinol Metab. 2014;99(10):3561-9.

39. VanderWalde LH, Liu ILA, O'Connell TX, Haigh PI. The effect of parathyroidectomy on bone fracture risk in patients with primary hyperparathyroidism. Archives of Surgery. 2006;141(9):885-9.

40. Khosla S, Melton LJ, Wermers RA, Crowson CS, O'Fallon WM, Riggs BL. Primary hyperparathyroidism and the risk of fracture: A population-based study. Journal of Bone and Mineral Research. 1999;14(10):1700-7.

41. Caron NR, Pasieka JL. What Symptom Improvement Can Be Expected After Operation for Primary Hyperparathyroidism? World Journal of Surgery. 2009;33(11):2244-55.

42. Almqvist EG, Becker C, Bondeson AG, Bondeson L, Svensson J. Early parathyroidectomy increases bone mineral density in patients with mild primary hyperparathyroidism: A prospective and randomized study. Surgery. 2004;136(6):1281-7.

43. Cheng SP, Lee JJ, Liu TP, Yang PS, Liu SC, Hsu YC, et al. Quality of Life After Surgery or Surveillance for Asymptomatic Primary Hyperparathyroidism A Meta-Analysis of Randomized Controlled Trials. Medicine. 2015;94(23):14.

44. Cummings SR, Melton LJ. Epidemiology and outcomes of osteoporotic fractures. Lancet. 2002;359(9319):1761-7.

45. Ensrud KE. Epidemiology of Fracture Risk With Advancing Age. Journals of Gerontology Series a-Biological Sciences and Medical Sciences. 2013;68(10):1236-42.

46. Cooper C, Melton LJ. Epidemiology of osteoporosis. Trends in Endocrinology and Metabolism. 1992;3(6):224-9.

47. Gold DT. The nonskeletal consequences of osteoporotic fractures - Psychologic and social outcomes. Rheumatic Disease Clinics of North America. 2001;27(1):255-+.

48. Harrison RA, Siminoski K, Vethanayagam D, Majumdar SR. Osteoporosis-related kyphosis and impairments in pulmonary function: A systematic review. Journal of Bone and Mineral Research. 2007;22(3):447-57.

49. Burge R, Dawson-Hughes B, Solomon DH, Wong JB, King A, Tosteson A. Incidence and economic burden of osteoporosis-related fractures in the United States, 2005-2025. Journal of Bone and Mineral Research. 2007;22(3):465-75.

50. Melton LJ. Adverse outcomes of osteoporotic fractures in the general population. Journal of Bone and Mineral Research. 2003;18(6):1139-41.

51. Kanis JA, McCloskey EV, Johansson H, Cooper C, Rizzoli R, Reginster JY, et al. European guidance for the diagnosis and management of osteoporosis in postmenopausal women. Osteoporosis International. 2013;24(1):23-57.

52. Rizzoli R, Branco J, Brandi ML, Boonen S, Bruyere O, Cacoub P, et al. Management of osteoporosis of the oldest old. Osteoporosis International. 2014;25(11):2507-29.

53. Hernlund E, Svedbom A, Ivergard M, Compston J, Cooper C, Stenmark J, et al. Osteoporosis in the European Union: medical management, epidemiology and economic burden. Archives of Osteoporosis. 2013;8(1-2):115.

54. Marshall D, Johnell O, Wedel H. Meta-analysis of how well measures of bone mineral density predict occurrence of osteoporotic fractures. British Medical Journal. 1996;312(7041):1254-9.

55. Cranney A, Jamal SA, Tsang JF, Josse RG, Leslie WD. Low bone mineral density and fracture burden in postmenopausal women. Canadian Medical Association Journal. 2007;177(6):575-80.

56. Johnell O, Kanis JA, Oden A, Johansson H, De Laet C, Delmas P, et al. Predictive value of BMD for hip and other fractures. Journal of Bone and Mineral Research. 2005;20(7):1185-94.

57. Kanis JA, Borgstrom F, De Laet C, Johansson H, Johnell O, Jonsson B, et al. Assessment of fracture risk. Osteoporosis International. 2005;16(6):581-9.

58. Klibanski A, Adams-Campbell L, Bassford T, Blair SN, Boden SD, Dickersin K, et al. Osteoporosis prevention, diagnosis, and therapy. Jama-Journal of the American Medical Association. 2001;285(6):785-95.

59. Strom O, Borgstrom F, Kanis JA, Compston J, Cooper C, McCloskey EV, et al. Osteoporosis: burden, health care provision and opportunities in the EU: a report prepared in collaboration with the International Osteoporosis Foundation (IOF) and the European Federation of Pharmaceutical Industry Associations (EFPIA). Arch Osteoporos. 2011;6:59-155.

60. Kanis JA, Oden A, Johnell O, Johansson H, De Laet C, Brown J, et al. The use of clinical risk factors enhances the performance of BMD in the prediction of hip and osteoporotic fractures in men and women. Osteoporosis International. 2007;18(8):1033-46.

61. Kanis JA, Oden A, Johansson H, Borgstrom F, Strom O, McCloskey E. FRAX (R) and its applications to clinical practice. Bone. 2009;44(5):734-43.

62. Hans DB, Kanis JA, Baim S, Bilezikian JP, Binkley N, Cauley JA, et al. Joint Official Positions of the International Society for Clinical Densitometry and International Osteoporosis Foundation on FRAX (R). Journal of Clinical Densitometry. 2011;14(3):171-80.

63. Swedish National Board of Health and Welfare. The National guidelines for Musculoskeletal Diseases 2012 - Revised 2014 Retrieved from: http://www.socialstyrelsen.se/nationalguidelines/nationalguidelinesfororthopaedicconditions.

64. Blake GM, Fogelman I. Technical principles of dual energy x-ray absorptiometry. Seminars in Nuclear Medicine. 1997;27(3):210-28.

65. Damilakis J, Adams JE, Guglielmi G, Link TM. Radiation exposure in X-ray-based imaging techniques used in osteoporosis. European Radiology. 2010;20(11):2707-14.

66. Blake GM, Naeem M, Boutros M. Comparison of effective dose to children and adults from dual X-ray absorptiometry examinations. Bone. 2006;38(6):935-42.

67. Blake GM, Fogelman I. Role of dual-energy X-ray absorptiometry in the diagnosis and treatment of osteoporosis. Journal of Clinical Densitometry. 2007;10(1):102-10.

68. Genant HK, Cooper C, Poor G, Reid I, Ehrlich G, Kanis J, et al. Interim report and recommendations of the World Health Organization task-force for osteoporosis. Osteoporosis International. 1999;10(4):259-64.

69. Kanis JA, Delmas P, Burckhardt P, Cooper C, Torgerson D. Guidelines for diagnosis and management of osteoporosis. Osteoporosis International. 1997;7(4):390-406.

70. Watts NB. Fundamentals and pitfalls of bone densitometry using dual-energy X-ray absorptiometry (DXA). Osteoporosis International. 2004;15(11):847-54.

71. Adams JE. Quantitative computed tomography. European Journal of Radiology. 2009;71(3):415-24.

72. Engelke K, Adams JE, Armbrecht G, Augat P, Bogado CE, Bouxsein ML, et al. Clinical use of quantitative computed tomography and peripheral quantitative computed tomography in the management of osteoporosis in adults: The 2007 ISCD Official Positions. Journal of Clinical Densitometry. 2008;11(1):123-62.

73. Cointry GR, Ferretti JL, Reina PS, Nocciolino LM, Rittweger J, Capozza RF. The pQCT "Bone Strength Indices" (BSIs, SSI). Relative mechanical impact and diagnostic value of the indicators of bone tissue and design quality employed in their calculation in healthy men and pre- and post-menopausal women. Journal of Musculoskeletal & Neuronal Interactions. 2014;14(1):29-40.

74. Gerdhem P, Obrant KJ. Effects of cigarette-smoking on bone mass as assessed by dual-energy X-ray absorptiometry and ultrasound. Osteoporosis International. 2002;13(12):932-6.

75. Jonsson B. Life style and fracture risk. Thesis. Page 67: Lund University, Sweden; 1993.

76. Kontulainen SA, Johnston JD, Liu D, Leung C, Oxland TR, McKay HA. Strength indices from pQCT imaging predict up to 85% of variance in bone failure properties at tibial epiphysis and diaphysis. J Musculoskelet Neuronal Interact. 2008;8(4):401-9.

77. Tenne M, McGuigan F, Besjakov J, Gerdhem P, Akesson K. Degenerative changes at the lumbar spine-implications for bone mineral density measurement in elderly women. Osteoporosis International. 2013;24(4):1419-28.

78. Bergstrom I, Landgren BM, Freyschuss B. Primary hyperparathyroidism is common in postmenopausal women with forearm fracture and low bone mineral density. Acta Obstetricia Et Gynecologica Scandinavica. 2007;86(1):61-4.

79. Mallmin H, Ljunghall S, Larsson K, Lindh E. Screening for primary hyperparathyroidism in patients with fractures of the distal forearm Acta Chirurgica-the European Journal of Surgery. 1991;157(11-12):657-9.

80. Hartge P. Participation in population studies. Epidemiology. 2006;17(3):252-4.

81. Galea S, Tracy M. Participation rates in epidemiologic studies. Annals of Epidemiology. 2007;17(9):643-53.

82. Hasserius R, Karlsson MK, Nilsson BE, Redlund-Johnell I, Johnell O. Non-participants differ from participants as regards risk factors for vertebral deformities - A

source of misinterpretation in the European Vertebral Osteoporosis Study. Acta Orthopaedica Scandinavica. 2002;73(4):451-4.

83. Bisgard KM, Folsom AR, Hong CP, Sellers TA. Mortality and cancer rates in nonrespondents to a prospective study of older women- 5- year follow-up. American Journal of Epidemiology. 1994;139(10):990-1000.

84. Norton MC, Breitner JCS, Welsh KA, Wyse BW. Characteristics of nonresponders in a community survey of the elderly. Journal of the American Geriatrics Society. 1994;42(12):1252-6.

85. Hoeymans N, Feskens EJM, Van den Bos GAM, Kromhout D. Non-response bias in a study of cardiovascular diseases, functional status and self-rated health among elderly men. Age and Ageing. 1998;27(1):35-40.

86. Riedel-Heller SG, Busse A, Angermeyer MC. Are cognitively impaired individuals adequately represented in community surveys? Recruitment challenges and strategies to facilitate participation in community surveys of older adults. A review. European Journal of Epidemiology. 2000;16(9):827-35.

87. Manjer J, Carlsson S, Elmstahl S, Gullberg B, Janzon L, Lindstrom M, et al. The Malmo diet and cancer study: representativity, cancer incidence and mortality in participants and non-participants. European Journal of Cancer Prevention. 2001;10(6):489-99.

88. Hara M, Sasaki S, Sobue T, Yamamoto S, Tsugane S, Grp JS. Comparison of cause-specific mortality between respondents and nonrespondents in a population-based prospective study: Ten-year follow-up of JPHC Study Cohort I. Journal of Clinical Epidemiology. 2002;55(2):150-6.

89. Kauppi M, Sokka T, Hannonen P. Survey nonresponse is associated with increased mortality in patients with rheumatoid arthritis and in a community population. Journal of Rheumatology. 2005;32(5):807-10.

90. Suominen S, Koskenvuo K, Sillanmaki L, Vahtera J, Korkeila K, Kivimaki M, et al. Non-response in a nationwide follow-up postal survey in Finland: a register-based mortality analysis of respondents and non-respondents of the Health and Social Support (HeSSup) Study. BMJ open. 2012;2(2):e000657.

91. Gerdhem P, Akesson K. Rates of fracture in participants and non-participants in the osteoporosis prospective risk assessment study. Journal of Bone and Joint Surgery-British Volume. 2007;89B(12):1627-31.

92. Oneill TW, Marsden D, Silman AJ. Differences in the characteristics of responders and nonresponders in a prevalence survey of vertebral osteoporosis. Osteoporosis Int. 1995;5(5):327-34.

93. Buist DSM, LaCroix AZ, Brenneman SK, Abbott T. A population-based osteoporosis screening program: Who does not participate, and what are the consequences? Journal of the American Geriatrics Society. 2004;52(7):1130-7.

94. Edwards MH, Jameson K, Denison H, Harvey NC, Sayer AA, Dennison EM, et al. Clinical risk factors, bone density and fall history in the prediction of incident fracture among men and women. Bone. 2013;52(2):541-7.

95. Albrand G, Munoz F, Sornay-Rendu E, DuBoeuf F, Delmas PD. Independent predictors of all osteoporosis-related fractures in healthy postmenopausal women: The OFELY Study. Bone. 2003;32(1):78-85.

96. Cauley JA, Wu LL, Wampler NS, Barnhart JM, Allison M, Chen Z, et al. Clinical risk factors for fractures in multi-ethnic women: The women's health initiative. Journal of Bone and Mineral Research. 2007;22(11):1816-26.

97. Kung AWC, Lee KK, Ho AYY, Tang G, Luk KDK. Ten-year risk of osteoporotic fractures in postmenopausal Chinese women according to clinical risk factors and BMD T-scores: A prospective study. Journal of Bone and Mineral Research. 2007;22(7):1080-7.

98. Hippisley-Cox J, Coupland C. Predicting risk of osteoporotic fracture in men and women in England and Wales: prospective derivation and validation of QFractureScores. British Medical Journal. 2009;339.

99. Cummings SR, Nevitt MC, Browner WS, Stone K, Fox KM, Ensrud KE, et al. Risk-factors for hip fracture in white women. New England Journal of Medicine. 1995;332(12):767-73.

100. Nguyen ND, Pongchaiyakul C, Center JR, Eisman JA, Nguyen TV. Identification of high-risk individuals for hip fracture: A 14-year prospective study. Journal of Bone and Mineral Research. 2005;20(11):1921-8.

101. Hans D, Durosier C, Kanis JA, Johansson H, Schott-Pethelaz AM, Krieg MA. Assessment of the 10-year probability of osteoporotic hip fracture combining clinical risk factors and heel bone ultrasound: The EPISEM prospective cohort of 12,958 elderly women. Journal of Bone and Mineral Research. 2008;23(7):1045-51.

102. Kelsey JL, Browner WS, Seeley DG, Nevitt MC, Cummings SR. Risk-factors for fractures of the distal forearm and proximal humerus. American Journal of Epidemiology. 1992;135(5):477-89.

103. Nguyen TV, Center JR, Sambrook PN, Eisman JA. Risk factors for proximal humerus, forearm, and wrist fractures in elderly men and women - The Dubbo osteoporosis epidemiology study. American Journal of Epidemiology. 2001;153(6):587-95.

104. Vogt MT, Cauley JA, Tomaino MM, Stone K, Williams JR, Herndon JH. Distal radius fractures in older women: A 10-year follow-up study of descriptive characteristics and risk factors. The study of osteoporotic fractures. Journal of the American Geriatrics Society. 2002;50(1):97-103.

105. Lee SH, Dargent-Molina P, Breart G, Grp E. Risk factors for fractures of the proximal humerus: results from the EPIDOS prospective study. Journal of Bone and Mineral Research. 2002;17(5):817-25.

106. McGrother CW, Donaldson MMK, Clayton D, Abrams KR, Clarke M. Evaluation of a hip fracture risk score for assessing elderly women: The Melton Osteoporotic Fracture (MOF) study. Osteoporosis International. 2002;13(1):89-96.

107. DargentMolina P, Favier F, Grandjean H, Baudoin C, Schott AM, Hausherr E, et al. Fall-related factors and risk of hip fracture: The EPIDOS prospective study. Lancet. 1996;348(9021):145-9.

108. Lundin H, Saaf M, Strender LE, Nyren S, Johansson SE, Salminen H. Gait speed and one-leg standing time each add to the predictive ability of FRAX. Osteoporosis International. 2017;28(1):179-87.

109. Karkkainen M, Rikkonen T, Kroger H, Sirola J, Tuppurainen M, Salovaara K, et al. Association between functional capacity tests and fractures: An eight-year prospective population-based cohort study. Osteoporosis International. 2008;19(8):1203-10.

110. Yoon V, Maalouf NM, Sakhaee K. The effects of smoking on bone metabolism. Osteoporosis International. 2012;23(8):2081-92.

111. Ward KD, Klesges RC. A meta-analysis of the effects of cigarette smoking on bone mineral density. Calcified Tissue International. 2001;68(5):259-70.

112. Vestergaard P, Mosekilde L. Fracture risk associated with smoking: a meta-analysis. Journal of Internal Medicine. 2003;254(6):572-83.

113. Hemenway D, Colditz GA, Willett WC, Stampfer MJ, Speizer FE. Fractures and lifestyle - effect of cigarette-smoking, alcohol intake, and relative weight on the risk of hip and forearm fractures in middle aged women. American Journal of Public Health. 1988;78(12):1554-8.

114. Meyer HE, Tverdal A, Falch JA. Risk-factors for hip fracture in middle-aged Norwegian women and men. American Journal of Epidemiology. 1993;137(11):1203-11.

115. Kanis JA, Johnell O, Oden A, Johansson H, De Laet C, Eisman JA, et al. Smoking and fracture risk: a meta-analysis. Osteoporosis International. 2005;16(2):155-62.

116. Hoidrup S, Prescott E, Sorensen TI, Gottschau A, Lauritzen JB, Schroll M, et al. Tobacco smoking and risk of hip fracture in men and women. International Journal of Epidemiology. 2000;29(2):253-9.

117. Lappin DF, Sherrabeh S, Jenkins WMM, Macpherson LMD. Effect of smoking on serum RANKL and OPG in sex, age and clinically matched supportive-therapy periodontitis patients. Journal of Clinical Periodontology. 2007;34(4):271-7.

118. Forsen L, Bjorndal A, Bjartveit K, Edna TH, Holmen J, Jessen V, et al. Interaction between current smoking, leanness, and physical inactivity in the prediction of hip fracture Journal of Bone and Mineral Research. 1994;9(11):1671-8.

119. Cornuz J, Feskanich D, Willett WC, Colditz GA. Smoking, smoking cessation, and risk of hip fracture in women. American Journal of Medicine. 1999;106(3):311-4.

120. Melton LJ, Christen D, Riggs BL, Achenbach SJ, Muller R, van Lenthe GH, et al. Assessing forearm fracture risk in postmenopausal women. Osteoporosis International. 2010;21(7):1161-9.

121. Boutroy S, Van Rietbergen B, Sornay-Rendu E, Munoz F, Bouxsein ML, Delmas PD. Finite element analysis based on in vivo HR-pQCT images of the distal radius is associated with wrist fracture in postmenopausal women. Journal of Bone and Mineral Research. 2008;23(3):392-9.

122. Stone KL, Seeley DG, Lui LY, Cauley JA, Ensrud K, Browner W, et al. BMD at multiple sites and risk of fracture of multiple types: Long-term results from the study of osteoporotic fractures. Journal of Bone and Mineral Research. 2003;18(11):1947-54.

123. Augat P, Iida H, Jiang YB, Diao E, Genant HK. Distal radius fractures: Mechanisms of injury and strength prediction by bone mineral assessment. Journal of Orthopaedic Research. 1998;16(5):629-35.

124. Muller ME, Webber CE, Bouxsein ML. Predicting the failure load of the distal radius. Osteoporosis International. 2003;14(4):345-52.

125. Crockett K, Arnold CM, Farthing JP, Chilibeck PD, Johnston JD, Bath B, et al. Bone strength and muscle properties in postmenopausal women with and without a recent distal radius fracture. Osteoporosis International. 2015;26(10):2461-9.

126. Vico L, Zouch M, Amirouche A, Frere D, Laroche N, Koller B, et al. High-Resolution pQCT Analysis at the Distal Radius and Tibia Discriminates Patients With Recent Wrist and Femoral Neck Fractures. Journal of Bone and Mineral Research. 2008;23(11):1741-50.

127. MacIntyre NJ, Adachi JD, Webber CE. In vivo measurement of apparent trabecular bone structure of the radius in women with low bone density discriminates patients with recent wrist fracture from those without fracture. Journal of Clinical Densitometry. 2003;6(1):35-43.

128. Nielsen SP, Xie X, Barenholdt O. Geometric properties of distal radius and pathogenesis of colles fracture - A peripheral quantitative computed tomography study. Journal of Clinical Densitometry. 2001;4(3):209-19.

129. Schneider P, Reiners C, Cointry GR, Capozza RF, Ferretti JL. Bone quality parameters of the distal radius as assessed by pQCT in normal and fractured women. Osteoporosis International. 2001;12(8):639-46.

130. Ahlborg HG, Johnell O, Turner CH, Rannevik G, Karlsson MK. Bone loss and bone size after menopause. New England Journal of Medicine. 2003;349(4):327-34.

131. Wapniarz M, Lehmann R, Reincke M, Schonau E, Klein K, Allolio B. Determinants of radial bone density as measured by PQCT in pre- and postmenopausal women: The role of bone size. Journal of Bone and Mineral Research. 1997;12(2):248-54.

132. Nijs J, Westhovens R, Joly J, Cheng XG, Borghs H, Dequeker J. Diagnostic sensitivity of peripheral quantitative computed tomography measurements at ultradistal and proximal radius in postmenopausal women. Bone. 1998;22(6):659-64.

133. Stein EM, Silva BC, Boutroy S, Zhou B, Wang J, Udesky J, et al. Primary hyperparathyroidism is associated with abnormal cortical and trabecular microstructure and reduced bone stiffness in postmenopausal women. Journal of Bone and Mineral Research. 2013;28(5):1029-40.

134. Chen QX, Kaji H, Iu MF, Nomura R, Sowa H, Yamauchi M, et al. Effects of an excess and a deficiency of endogenous parathyroid hormone on volumetric bone mineral density and bone geometry determined by peripheral quantitative computed tomography in female subjects. Journal of Clinical Endocrinology & Metabolism. 2003;88(10):4655-8.

135. Hansen S, Jensen JEB, Rasmussen L, Hauge EM, Brixen K. Effects on Bone Geometry, Density, and Microarchitecture in the Distal Radius but not the Tibia in Women With Primary Hyperparathyroidism: A Case-Control Study Using HR-pQCT. Journal of Bone and Mineral Research. 2010;25(9):1941-7.

136. Negri AL, Del Valle EE, Zanchetta MB, Nobaru M, Silveira F, Puddu M, et al. Evaluation of bone microarchitecture by high-resolution peripheral quantitative computed tomography (HR-pQCT) in hemodialysis patients. Osteoporosis International. 2012;23(10):2543-50.

137. Kaji H, Yamauchi M, Chihara K, Sugimoto T. Relationship between endogenous parathyroid hormone and bone metabolism/geometry in female patients treated with glucocorticoid. Hormone and Metabolic Research. 2008;40(1):60-5.

138. Di Monaco M, Vallero F, Di Monaco R, Mautino F, Cavanna A. Primary hyperparathyroidism in elderly patients with hip fracture. Journal of Bone and Mineral Metabolism. 2004;22(5):491-5.

139. Malgo F, Appelman-Dijkstra NM, Termaat MF, van der Heide HJL, Schipper IB, Rabelink TJ, et al. High prevalence of secondary factors for bone fragility in patients with a recent fracture independently of BMD. Archives of Osteoporosis. 2016;11(1):8.

140. Scharla SH, Wolf S, Dull R, Lempert UG. Prevalence of low bone mass and endocrine disorders in hip fracture patients in Southern Germany. Experimental and Clinical Endocrinology & Diabetes. 1999;107(8):547-54.

141. Bours SPG, van Geel T, Geusens P, Janssen MJW, Janzing HMJ, Hoffland GA, et al. Contributors to Secondary Osteoporosis and Metabolic Bone Diseases in Patients Presenting with a Clinical Fracture. Journal of Clinical Endocrinology & Metabolism. 2011;96(5):1360-7.

142. Melton LJ, Achenbach SJ, O'Fallon WM, Khosla S. Secondary osteoporosis and the risk of distal forearm fractures in men and women. Bone. 2002;31(1):119-25.

143. Rubin MR, Bilezikian JP, McMahon DJ, Jacobs T, Shane E, Siris E, et al. The natural history of primary hyperparathyroidism with or without parathyroid surgery after 15 years. Journal of Clinical Endocrinology & Metabolism. 2008;93(9):3462-70.

9 APPENDIX

9.1 ATTRIBUTIONS

Paper I. Reprinted by permission from Creative Commons: Nordic Orthopaedic Federation. Acta Orthopaedica. External validity of a population-based study on osteoporosis and fracture. Wihlborg A, Åkesson K, Gerdhem P (2014).

Paper II. Reprinted by permission from RightsLinks: Springer Nature. Osteoporosis International. Fracture predictive ability of physical performance tests and history of falls in elderly women: a 10-year prospective study. Wihlborg A, Englund M, Åkesson K, Gerdhem P (2014).

Paper III. Reprinted by permission from RightsLinks: Springer Nature. Osteoporosis International. Smoking, smoking cessation, and fracture risk in elderly women followed for 10 years. Thorin M H, Wihlborg A, Åkesson K, Gerdhem P (2016).

Figure 1. Anatomy of human bone spongy structure vector illustration by Eranicle. Stockvektor-ID: 499224367. Image modified. Published with permission from www.shutterstock.com

Figure 2. Bone Detail Anatomy by Mmutlu. Stockvector-ID: 33044812. Image modified. Published with permission from www.shutterstock.com

Figure 3. The bone remodeling process involves the following steps: resorption, reversal, formation, mineralization and resting. In a healthy body, osteoclasts and osteoblasts work together by Designua. Stockvector-ID: 333409151. Image modified. Published with permission from www.shutterstock.com

Figure 4. Age and bone mass (By Anatomy & Physiology, Connexions Web site. https://upload.wikimedia.org/wikipedia/commons/9/91/615_Age_and_Bone_Mass.jpg Published with permission from Creative Commons)

Figure 5. Two slides comparing the vertebrae of a healthy 37 year old male with a 75 year old female suffering from osteoporosis (By Turner Biomechanics Laboratory, https://commons.wikimedia.org/wiki/File:Bone_Comparison_of_Healthy_and_Osteoporotic_Vertibrae.png Published with permission from Creative Commons)