CORTICOSTEROID-INDUCED OSTEOPOROSIS IN DAILY PRACTICE

Corticosteroid-Induced Osteoporosis in Daily Practice

Pathophysiology
Clinical Importance
Prevention and Intervention Strategies

A. Struijs

CORTICOSTEROID-INDUCED OSTEOPOROSIS IN DAILY PRACTICE

Pathophysiology
Clinical Importance
Prevention and Intervention Strategies

CORTICOSTEROID GEINDUCEERDE OSTEOPOROSE IN DE DAGELIJKSE PRAKTIJK

Pathofysiologie
Klinische betekenis
Preventie en Interventie Strategieën

PROEFSCHRIFT

Ter verkrijging van de graad van doctor
aan de Erasmus Universiteit Rotterdam
op gezag van de rector magnificus
Prof. dr. P.W.C. Akkermans M.A.
en volgens besluit van het College voor Promoties

De openbare verdediging zal plaatsvinden op
woensdag 19 januari 2000 om 9.45 uur
door
ADRIAAN STRUIJS
geboren op 8 augustus 1965 te Vlaardingen

Promotiecommissie

Promotores:
Prof. dr H.A.P. Pols
Prof. dr A.G.H. Smals

Overige leden:
Prof. dr S.W.J. Lamberts
Prof. dr H.J. Stam
Prof. dr J. Verhaar

Struijs, Ard
Corticosteroid-induced osteoporosis in daily practice
Thesis Erasmus University Rotterdam. –With ref.–With summary in Dutch.
ISBN 90-73235-28-6

Cover: Dick Ladage, Rotterdam
Lay-out: Ger Guijs, Rotterdam
Printing: Optima Grafische Communicatie, Rotterdam

Printing of this thesis was financially supported by:
Procter and Gamble Pharmaceuticals
Hoechst Marion Roussel
Fresenius Kabi
Eli-Lilly
Novartis Pharma B.V.

Voor mijn ouders

CONTENTS

PART 1

General Introduction and Scope of the Thesis

"The greatly compressed bodies of the vertebrae...
Were so soft they could easily be cut with a knife."
Harvey Cushing, 1932

GENERAL INTRODUCTION

Harvey Cushing already described in 1932 the skeletal phenotype of patients with endogenous hypercortisolism. He reported a tendency to become round-shouldered (kyphotic) even to the point of measurable height loss associated with lumbo-spinal pain [1]. His description is nowadays recognised as the typical clinical presentation of a patient with an osteoporotic stature and vertebral fractures. After the introduction of glucocorticoids in the treatment of a wide variety of diseases, also the association of osteoporosis with exogenous hypercortisolism was recognised [2]. It became the most frequent form of secondary osteoporosis [3]. Osteoporosis due to glucocorticoids seems to be dose dependent. It has to be emphasised that also the disorders for which these agents are given may themselves cause bone loss. The overall incidence of osteoporosis in patients taking glucocorticoids for more than six months approaches 50% [4, 5].

For over fifty years we now recognise the relation between glucocorticoid use and osteoporosis but only recently more data become available about the prevalence of glucocorticoid use and the relation between dosage and incidence of fracture. In the United Kingdom (UK) the prevalence of the long-term use of systemic glucocorticoids was studied recently by Walsh et al. He found that 0,5% of the Nottinghamshire population was taking long-term glucocorticosteroids [6]. Exact data on the chronic use of glucocorticoids in the Netherlands are not available, but it had been suggested that around 75000 patients are treated with corticosteroids [7].

If fracture occurs the patient will come under medical attention and only than the possibility of steroid-induced osteoporotic fracture will be taken into account. Thirty to thirty-five percent of patients receiving long-term glucocorticoid therapy have vertebral fractures, and the risk of hip fracture is 50% higher in these patients compared to that in age-matched controls [5]. Others reported 2- to 3-fold higher incidence of bone fractures in glucocorticoid treated patients with ranging from 8-18% [8-10]. From recent data published on the steroid dose and fracture risk in the UK, it appeared that already an average daily dose of 2,5 mg of prednisolone per day could increase the risk of non-vertebral

fracture risk with 17%. In doses between 2,5 mg and 7,5 mg this risk was 36% and in average daily dose over 7,5 mg it was increased with 64% [11].
The cost of treatment of osteoporotic fractures is high and the consequences for the patients are multiple. Therefore there is a clear need for strategies to stratify the risk for osteoporotic fractures. At present Bone Mineral Density (BMD) measurements with Dual X-ray Absorptiometry (DXA), provide an accurate and precise way, although it remains questionable whether the relation between BMD and fracture risk is similar in glucocorticoid treated patients compared to what is found in postmenopausal women. In postmenopausal women, a decrease of 1 Standard Deviation (SD) or about 12% in BMD, is associated with a doubling of fracture risk [12-15]. This relationship may be an underestimate in patients treated with glucocorticoids, who appear to suffer fractures at BMD values higher than in postmenopausal osteoporosis [16].
The identification of patients at risk, the development of prevention and treatment strategies, together with the creation of higher awareness of the medical profession on the risk of steroid induced osteoporosis are the challenges for the near future.

SCOPE OF THE THESIS

Glucocorticoid-induced osteoporosis is a serious disease that strikes millions of people world-wide every year. Nevertheless much debate is still going on about the preferred strategies to be followed and the magnitude of the problem. Therefore, the first aim of this thesis was to review the current strategies to identify the patient at risk and the options available for prevention and treatment of this serious skeletal disease. In addition we tried to obtain insight in the magnitude of the population at risk in The Netherlands. We also studied the effects of inhaled glucocorticoids on bone mineral density and markers of bone turnover.
The second aim was focused on the prevention and treatment of glucocorticoid-induced osteoporosis. First of all the acute effect of etidronate on markers of bone turnover was studied in glucocorticoid treated patients. Subsequently, in two separate studies the effects of etidronate in the prevention and treatment of glucocorticoid-induced osteoporosis was investigated.

PART 2

Update on the Pathogenesis and Treatment of Glucocorticoid-Induced Osteoporosis

CHAPTER 1

GLUCOCORTICOID-INDUCED OSTEOPOROSIS

1.1. DEFINITION

The currently accepted definition of osteoporosis is, a systemic skeletal disease characterised by low bone mass and micro-architectural deterioration of bone tissue, with a consequent increase in bone fragility and susceptibility to fractures [17]. BMD can be measured with acceptable accuracy and precision and forms the basis for an operational definition of osteoporosis with better clinical utility [18]. A working group of the World Health Organisation (WHO) has proposed guidelines for the interpretation of bone mass measurement in Caucasian woman. Four categories are discriminated, based on measurement of BMD and the presence or absence of osteoporotic fractures. The BMD of the individual is compared to the average mean of young healthy individuals and expressed in standard deviation (SD) below the mean [19].

Those categories are:

Normal bone mass
•BMD with a T-score > - 1 SD
Low bone mass or osteopenia
•BMD with a T-score < -1 SD and ≥ -2.5 SD
Osteoporosis
•BMD with a T-score < -2.5 SD
Established osteoporosis
•BMD with a T-score < -2.5 SD and the presence of fractures

A potential restriction of the WHO classification is, that it has been developed in an

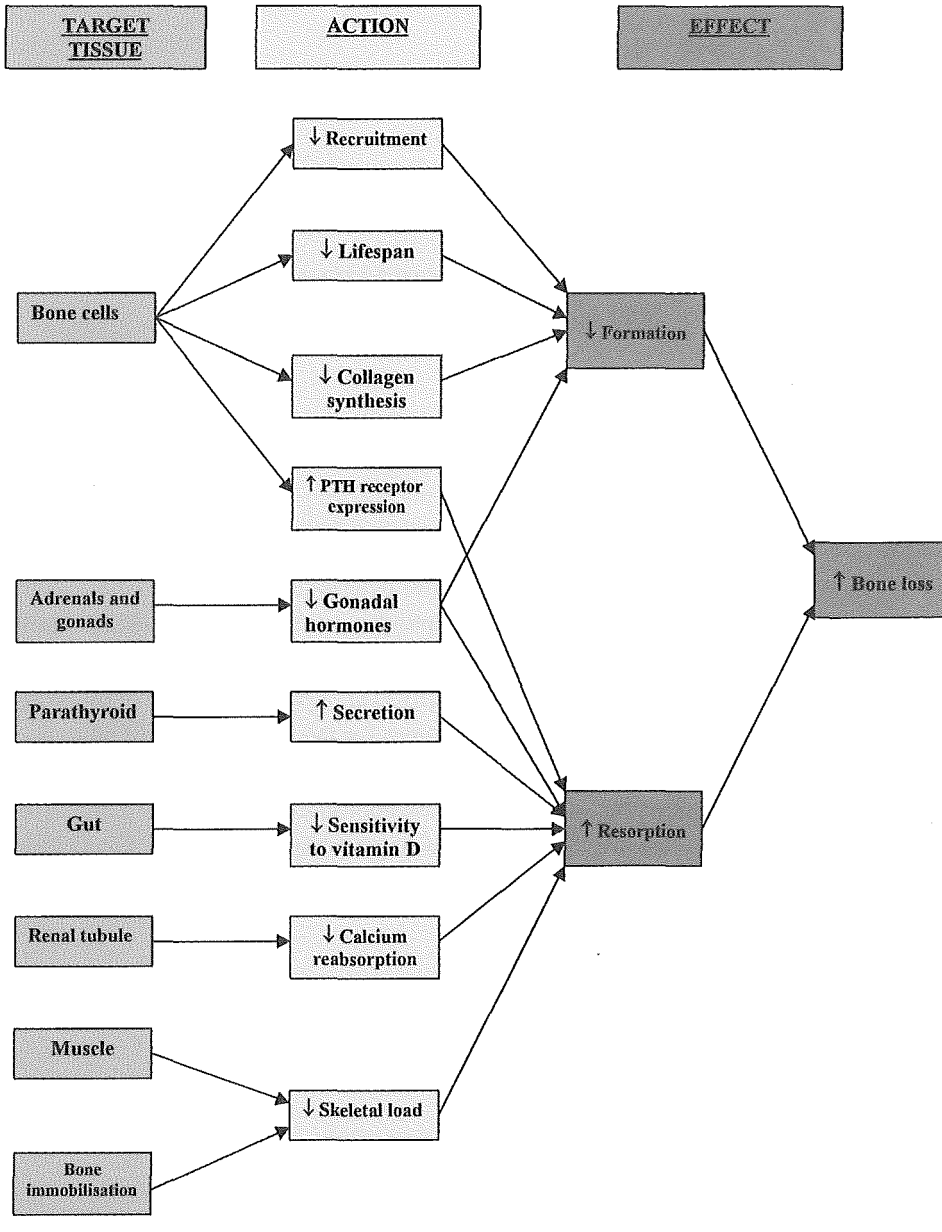


Figure 1.2.1 Effects of glucocorticoids on different target organs.

attempt to describe the severity of the disease for women with post-menopausal osteoporosis and may not be accurate for corticosteroid-induced osteoporosis [16,20]. However to establish an international standard, the WHO criteria are also used in corticosteroid-induced osteoporosis. Two groups have launched guidelines for corticosteroid induced osteoporosis [21,22]. Recently the UK consensus group updated their guideline [23]. We will discuss these guidelines in paragraph 4.2.

1.2. PATHOPHYSIOLOGY

Corticosteroids can affect bone directly, by altering calcium and bone metabolism, but also indirectly by altering different hormonal axes and protein metabolism [24-30]. (Figure 1.2.1)

1.2.1 Effects of glucocorticoids on the skeleton

1.2.1.1 Cellular level

Glucocorticoids exert their action on bone on many different levels. The precursors of osteoblasts are multipotent mesenchymal stem cells, while the precursors of osteoclasts are hematopoietic cells of the monocyte /macrophage lineage. The development of both osteoblasts and osteoclasts is controlled by growth factors and cytokines that are produced in the bone marrow microenvironment and is modulated by systemic hormones and probably by mechanical signals [31]. At the cellular level corticosteroids decrease the number of osteoblast precursor cells by about 86% and also shorten their lifetime [32]. Osteoblasts in different stages of differentiation have different responses to glucocorticoids, mediated through their glucocorticoid receptor [33]. The use of glucocorticoids results in increased differentiation of osteoblast precursors and thereby causes lower rates of proliferation and protein synthesis [34-40]. Glucocorticoids also affect several growth factors secreted by bone cells. The synthesis of insulin-like growth factors (IGFI) is inhibited and glucocorticoids increase the production of binding proteins that can trap IGF I. IGFs have opposite effects to glucocorticoids, since IGFs increase bone cell replication and bone formation. Therefore the inhibitory effects of glucocorticoids on the IGF axis are to an extent responsible for the inhibitory effects of these steroids on bone formation [41-43].
Decreased synthesis of matrix proteins like type I collagen [44] and osteocalcin [45] by the osteoblast, is a result of decreased gene transcription and decreased m-RNA stability

due to glucocorticoids. Not only type I collagen and osteocalcin but also osteopontin, fibronectin, B1 Integrin, bone sialoprotein and insulin like growth factor are modulated by glucocorticoids [46]. The inhibition of integrin expression in osteoblasts impairs their attachment to bone [47].
Another effect of glucocorticoids is the increased expression of PTH receptors on the osteoblast.
The response of bone cells and bone organ cultures to glucocorticoids depends on the timing and possibly the glucocorticoid dose. Gallagher demonstrated that low dose of glucocorticoids and short incubation periods stimulated both cell growth and collagen synthesis in human osteoblast cells [48]. In contrast a long-term culture with supraphysiological doses of glucocorticoids inhibited cell growth and collagen synthesis [49].
In contrast to osteoblasts, the osteoclastic response to glucocorticoids is indirectly mediated. Also reduction of osteoclast precursor cells and decrease of lifetime of the osteoclast has been reported [50].

1.2.1.2 Tissue level

In contrast to linear or appositional bone growth, skeletal turnover in adults occurs through quantitative changes in discrete bone remodelling sites called basic multi-cellular units (BMU's) or bone remodelling units (BRU's) [51-54]. This theory is now widely accepted, and the major events in the bone remodelling cycle have been defined as activation → resorption → reversal → formation → quiescence [51,55-57]. Normal bone turnover is characterised through the tight coupling of bone resorption and formation. However the use of glucocorticoids results in uncoupling of bone remodelling by acting in two discrete ways: 1) through direct action on the osteoblast, and 2) through increased bone resorption caused by indirect modulation of calciotropic hormones, cytokines, and prostaglandins [50].
Glucocorticoid-induced bone loss can be explained by the fact that the amount of new bone synthesised is less than that removed by osteoclast [58-62]. To this net negative effect contributes a decrease in the period of active bone formation [59]. Imbalance at the level of the BMU appears to be largely driven by the decrease in bone formation. Nevertheless there is also an increase in resorption although this seems to be more explicit in the early phase of glucocorticoid use [58-62]. Furthermore, the positive effect of glucocorticoids on the number of activated BMU's leads to an increased number of resorption areas in different stages of resorption and formation. This results in a progressive thinning of the trabeculae [58].
It has been shown, that later on the resorptive cavities no longer contain osteoclasts so

that it seems more a problem of the osteoblasts, which are too slow to fill the resorptive cavities. Thus, it is likely that the effects of glucocorticoids on bone formation are much more dramatic than their effects of glucocorticoids on bone resorption. This is consistent with the histological picture of glucocorticoid-induced osteoporosis, which consists of thinned rather than perforated trabeculae, whereas in idiopathic osteoporosis perforation appears to be more prominent [58].

1.2.2 Effects of glucocorticoids on calcium and phosphate homeostasis.

Normal calcium homeostasis and the effects of glucocorticoids are shown in figure 1.2.2. Two mechanisms of intestinal calcium absorption have been identified. The active transport is an energy dependent process and occurs against an intestinal calcium concentra-

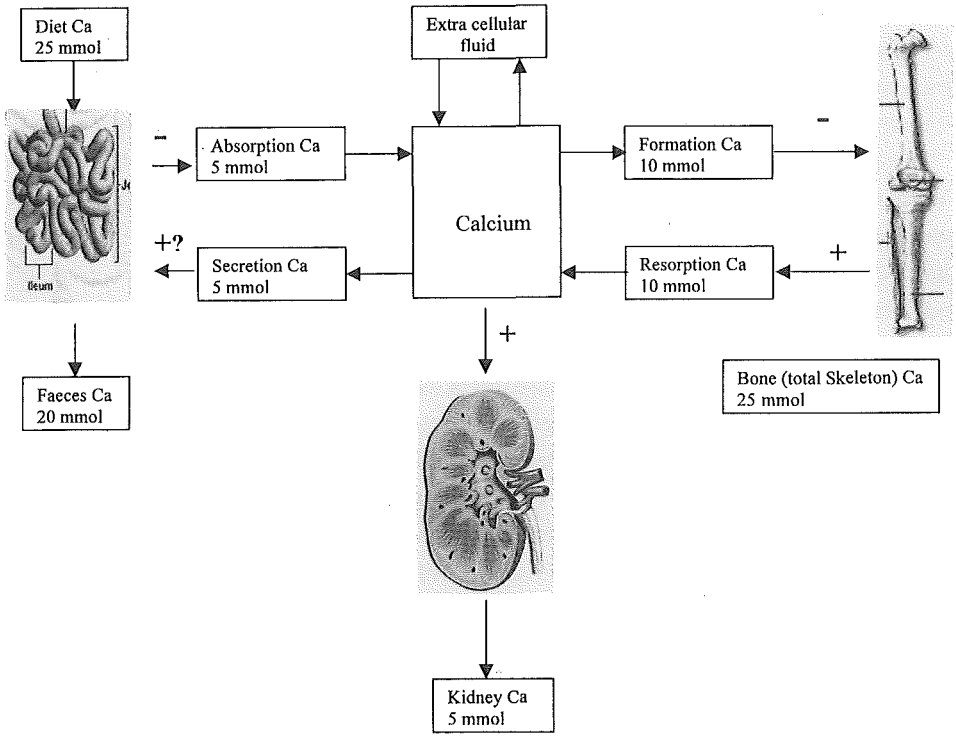


Figure 1.2.2 The interaction of glucocorticoids with the overall calcium homeostasis.

tion gradient. This system is already saturated by a low concentration of intestinal calcium and is vitamin D dependent [63]. The second mechanism is passive diffusion of calcium across the intestinal wall.

In most but not all studies, use of glucocorticoids is associated with a decreased intestinal absorption of calcium and phosphate [64-66]. Two studies reported normal calcium absorption in patients taking glucocorticoids, except in those with fractures [67,68] and another study even reported an increase in fractional calcium absorption in patients treated with corticosteroids compared to that in controls [69]. If intestinal calcium absorption decreases, it occurs early in the first weeks of glucocorticoid therapy and is partially dose dependent [70,71]. In glucocorticoid excess primarily the active intestinal calcium absorption is inhibited [72-75]. However, treatment with pharmacological doses of 1,25-dihydroxyvitamin D_3, can only partially reverse intestinal calcium loss [69]. Other studies showed that the impairment of intestinal calcium absorption appears also to be independent of vitamin D metabolism [64,71,76,77]. Therefore lowered fractional intestinal calcium absorption in corticosteroid treated patients can only be partially explained by lowered active calcium absorption. So other unknown effects of glucocorticoids on intestinal calcium absorption must be present. Inhibition of protein synthesis, especially decreased synthesis of vitamin D binding protein [8,25,78] and decreased calcium release from mitochondria because of depletion of mitochondria adenosine triphosphate [79] are suggested as potential mechanisms.

As passive diffusion of calcium across the intestinal wall is not altered in the presence of corticosteroid excess, it becomes the major pathway for calcium absorption. Therefore the overall reduction in fractional calcium absorption is even more severe when dietary calcium is restricted [80]. Thus high doses of calcium should be given to patients using glucocorticoids.

Another reason for calcium supplementation is the altered renal handling of calcium and phosphate in patients treated with glucocorticoids. Sustained glucocorticoid excess results in increased GFR, marked hypercalciuria, and a doubling of fasting urine calcium excretion [70,81-84]. This has been a consistent finding in different studies and is probably mediated by a direct inhibitory effect of glucocorticoids on renal tubular calcium reabsorption that occurs in spite of elevated levels of PTH and increased sodium excretion [76,82,84,85]. Phosphaturia and decreased tubular reabsorption of phosphate also have been reported [86-89].

Decreased intestinal calcium absorption and increased renal excretion of calcium will lead to a negative calcium balance and thereby to secondary hyperparathyroidism with increased bone turnover and subsequently increased bone resorption [66,76,82,90]. Furthermore increased bone resorption and decreased bone formation, caused by the

combination of glucocorticoids and secondary hyperparathyroidism, will lead to an increased filtered load of calcium and contributes to a greater renal calcium loss.

1.2.3 Effects of glucocorticoids on vitamin D and PTH homeostasis

The possible contribution of alterations in vitamin D metabolism with steroid-induced osteoporosis has been extensively studied but the results seem to be inconclusive. Low [64,91] and normal [76,92] levels of 25-$(OH)D_3$ have been found in corticosteroid treated patients. Circulating levels of the active metabolite 1,25-$(OH)_2$ D_3 have also been shown to be low [91], normal [77,92], or increased [71,75] in corticosteroid treated patients. This can be partly explained by short and long-term effects of glucocorticoids on vitamin D metabolism: on the short-term 1,25-$(OH)_2$ D_3 levels were increased, on the long-term 1,25-$(OH)_2$ D_3 and 25-$(OH)D_3$ levels were normal. Seeman et al [77] reported no effects on normal levels, production and clearance rates of 1,25-$(OH)_2$ D_3 in patients before and during treatment with high doses of glucocorticoids or before and after successful treatment of endogenous hypercortisolism. Low levels of 1,25-$(OH)_2$ D_3 and 25-$(OH)D_3$ could be explained by other effects like, the underlying disease for which glucocorticoids have been prescribed, sunlight exposure, dietary factors and seasonal variation of vitamin D metabolites. From these data an absolute deficiency of active vitamin D does not seem likely to be of predominant pathofysiologic importance in glucocorticoid induced osteoporosis.
The same problem returns when interpreting the studies on PTH in glucocorticoid treated patients. Elevated [66,76,93] and normal PTH values have been reported [35,77,86,94]. Elevated PTH levels are often found in patients on chronic glucocorticoid treatment. Increased secondary production of PTH, direct stimulation of parathyroid cell secretor activity, and increased osteoblast sensitivity to PTH are possible mechanisms by which glucocorticoids may modulate parathyroid activity and cause PTH mediated bone loss [84]. , When secondary hyperparathyroidism is present, administration of calcium supplements and 25-$(OH)D_3$ [76] can reverse it. It must be mentioned, that decreased calcium absorption, increased bone resorption and changes in circulating vitamin D and PTH are mainly observed in patients receiving the higher dosages of corticosteroids.

1.2.4 The effects of glucocorticoids on sex hormones

Glucocorticoids have a negative effect on the sex hormone status in men and women, which may in turn increase bone resorption (See figure 1.2.4). This is caused by decreased gonadal stimulation due to inhibition of pituitary gonadotrophin secretion. The

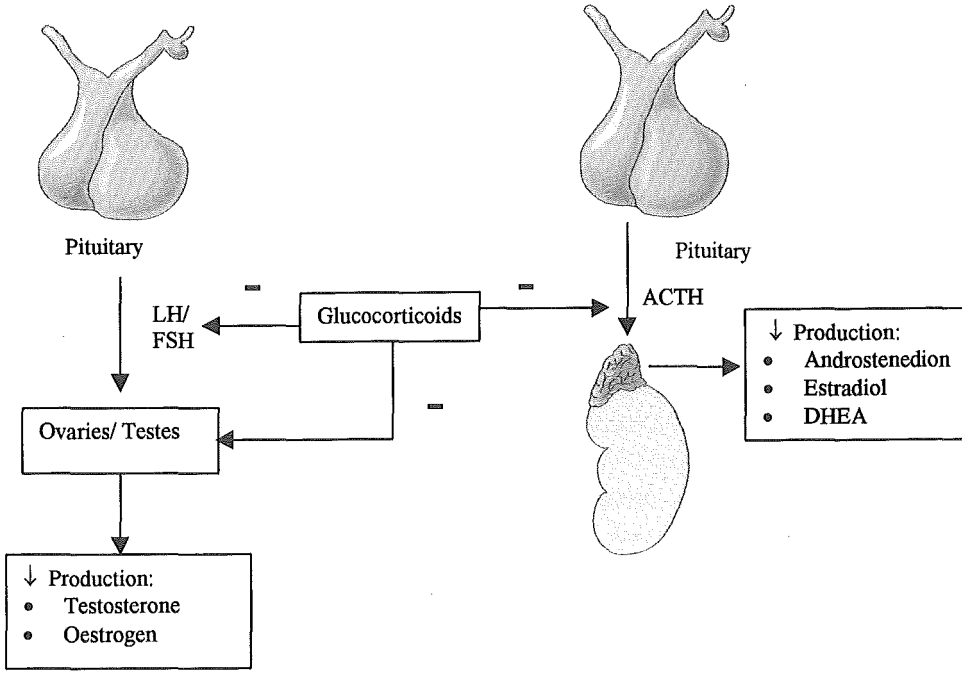


Figure 1.2.4 Effects glucocorticoids on pituitary and adrenal axes.

secretion of luteneizing hormone (LH) in response to luteneizing hormone-releasing hormone (LHRH) is reduced in both men and women [95,96]. Glucocorticoids also decrease follicle-stimulating hormone (FSH)-induced oestrogen production. Furthermore suppression of corticotrophin (ACTH) leads to adrenal atrophy and reduced production of adrenal androgens [97].

Also direct negative effects of glucocorticoids on ovaries and testes have been reported [95,96]. Glucocorticoids decrease testosterone production in the testis [98,99]. Men receiving glucocorticoid drugs have a dose-related reduction in circulating testosterone concentrations of nearly 50% in comparison with controls [29,30,100-102]. The combination of glucocorticoids and oestrogen deficiency has potentiating effects on the rate of bone loss [103]. So postmenopausal women receiving corticosteroids are particularly susceptible to bone loss [104].

1.2.5 Other factors that contribute to bone loss caused by glucocorticoids

Other factors that contribute to bone loss caused by glucocorticoids are summarised in table 1.2.5. The underlying disease for which corticosteroids are prescribed and the co-medication used may contribute to bone loss. Furthermore cytokines may play a modulatory role in this process, for example in rheumatoid arthritis. However, the contribution of these factors has only been marginally studied [104-109]. Chronic disease per se, with its decreased activity and mobility, also bears a risk for osteoporosis [110].

Miscellaneous factors in glucocorticoid bone loss
• Underlying disease • Modulation by cytokines • Co-medication • Myopathy • Immobilisation

Table 1.2.5

In post-organ-transplantation patients the concomitant administration of immunosupressants and glucocorticoids has negative effects on bone [108,109,111,112].
Glucocorticoids also have general catabolic effects. In muscle this is particularly important, as shown in patients with Cushing's syndrome [113-115]. A striking association between the presence of steroid myopathy and osteoporosis has been demonstrated. The muscle weakness is not only due to glucocorticoid use, but also determined by underlying conditions. In disease entities as rheumatoid arthritis or polymyositis, glucocorticoid -induced myopathy can hardly be distinguished from muscle weakness resulting from the disease process itself and subsequent inactivity [116]. Inactivity and muscle loss both lead to bone mineral loss [117]. All the factors involved in glucocorticoid induced bone loss are summarised in figure 1.2.5 (page 30).

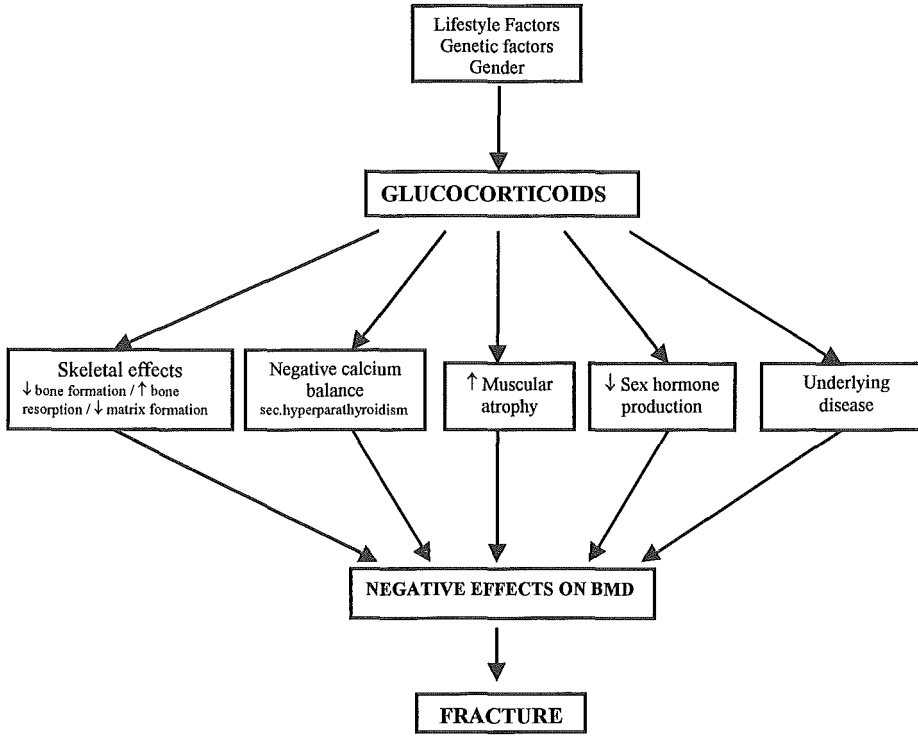


Figure 1.2.5 Direct and indirect effects of glucocorticoids on BMD and consequent fracture risk.

CHAPTER 2

SKELETAL AND NON-SKELETAL ASPECTS OF FRACTURE RISK

2.1. FRACTURE RISK IN PATIENTS TREATED WITH GLUCOCORTICOIDS

There are no reliable epidemiological data concerning the prevalence and incidence of corticosteroid use, the average dose used and its relation to fractures. These data are essential for developing preventive and therapeutic strategies. Furthermore, the factors involved in fracture risk are complex and involve of both skeletal and non-skeletal factors, as shown in figure 2.1.1 (page 32).

Observational data in patients with rheumatoid arthritis treated with glucocorticoids indicate that the risk of hip and other non-vertebral fractures is doubled [9,10], while vertebral fracture prevalence may be increased four- to five-fold [118,119]. In prospective studies in transplanted patients treated with glucocorticoids, fractures have been reported in one-third of the patients [120,121]. The determinants of low bone mass in glucocorticoid treated patients, like underlying disease, altered vitamin D metabolism, co-medication etc., have been described in chapter 1.

Obviously the risk of fracture is not only dependent on these factors, but also on the dose of glucocorticoids used. Higher doses of glucocorticoids result in higher fracture rates [11]. One cross-sectional and one retrospective study found a positive relation between cumulative dose of glucocorticoids and fracture risk [11,108]. Reid et al. suggested a positive correlation between cumulative glucocorticoid dose and low bone mineral density [122]. This was confirmed in other studies [108,109,123-126]. A cumulative dose of 10 grams of prednisone is associated with an increased risk of osteoporosis [127]. Only one study failed to demonstrate a relationship between bone density and the dose or

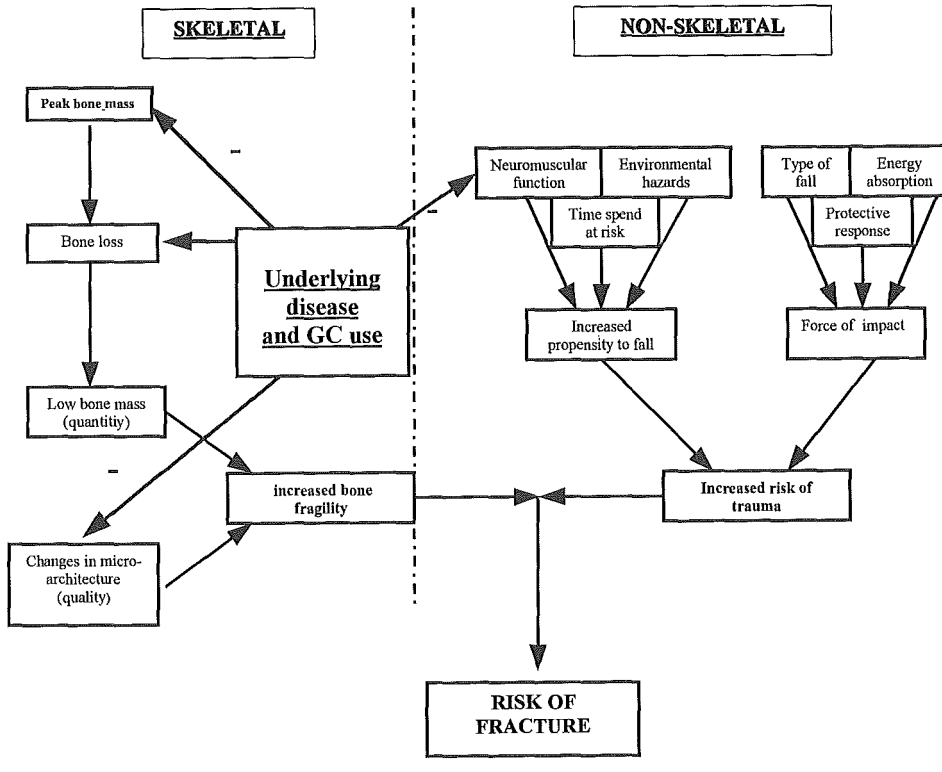


Figure 2.1.1 Determinants of fracture risk and the effect of glucocorticoids.

duration of glucocorticoid treatment [128].

The minimal dose of glucocorticoids associated with rapid bone loss is not well established. In general a daily dose of ≥ 7,5 mg/day is defined as a harmful dose for the skeleton, however individual variability of susceptibility can lead to bone loss at lower dosages, as shown very recently by van Staa [129]. One of the determinants of fracture risk which can be measured and modified is bone mass. The severity of low bone mass is characterised by the use of BMD measurement, often expressed as a T-score. In the T-score, both age and peak bone mass are weighted in a way that severity of (post-menopausal) osteoporosis could be expressed. With respect to glucocorticoid-induced osteoporosis it is still questionable whether the T-score is appropriate to express and

define glucocorticoid-induced osteoporosis. Studies to support the use of the T-score in glucocorticoid-induced osteoporosis are still lacking. Furthermore it is not known whether the correlation between BMD and fracture risk, as shown in postmenopausal osteoporosis, is similar in steroid treated patients. Luengo et al. suggested, in their cross sectional study of patients taking glucocorticoids for asthma, that the BMD fracture threshold compared to age-matched osteoporotics not using glucocorticoids, lies at a higher level [16]. Peel et al. found a 6.2 fold increase in vertebral fracture in 76 postmenopausal women with rheumatoid arthritis treated with glucocorticoids, although lumbar spine BMD was only decreased with < 1 SD [130]. Recently Banffer et al [20] presented also evidence for a higher susceptibility for fractures in case of glucocorticoid use. It appeared that bone loss is related to daily dose, duration of therapy and cumulative dose.

In our opinion, the cumulative corticosteroid dose appears to be a major risk factor for bone loss and subsequent fractures. Especially young people and postmenopausal women, who take excess amounts of corticosteroids for long periods of time, are at risk for accelerated bone loss [3,127]. In addition, prospective studies are urgently needed to define the relationship between BMD and fracture risk at different maintenance dose of glucocorticoids. Meanwhile we have to be pragmatic and treat our patients as described in chapter 4.

2.2 OTHER ASPECTS OF FRACTURE RISK

2.2.1. Biochemical markers of bone turnover

As described above, glucocorticoid induced bone loss results from a combination of inhibitory effects on bone formation and enhanced bone resorption, with the inhibitory effect on bone formation appearing to be the most important (See chapter 1).

The rate of formation or degradation of bone matrix can be assessed either by measuring enzymatic activity of bone forming or resorbing cells- such as alkaline and tartrate resistant acid phosphatase- or by measuring bone matrix components released into the circulation during formation and resorption. These markers have been separated into formation and resorption markers (see table 2.2.1, page 34). However, it should be kept in mind that bone markers could not discriminate between turnover changes in cortical or trabecular bone and not reflect changes at the level of the individual BMU.

Furthermore circulating levels of these markers can be influenced by factors other than bone turnover, such as their metabolic clearance (liver uptake, renal excretion, and/or

Table 2.2.1 Markers of bone metabolism.

FORMATION	RESORPTION
Serum	**Plasma**
• Osteocalcin • Total and bone specific alkaline phosphatase • Procollagen I carboxy-Terminal extension peptide (PITP)	• Tartrate-resistant acid phosphatase • Pyridinoline and pyridinoline containing peptides
	Urine
	• Urinary Pyridinoline and desoxypyridinoline (collagen cross-links) and containing peptides • Fasting urinary calcium and hydroxyproline • Urinary hydroxylysine glycosides

trapping on bone hydroxyapatite). Thus, each of these assays needs to be validated in a specific clinical situation before conclusions about their clinical utility can be drawn [131].

Advantages of these markers are the general availability and their ability to detect acute changes in bone metabolism. Also the discomfort of the patient by collection of blood or urine is minimal. The immunoassays of osteocalcin and bone specific alkaline phosphatase represent so far the most effective markers of bone formation in osteoporosis. Measurement of pyridinium cross-links, and some of its related peptides, is an improvement in the assessment of bone resorption. Well-characterised immunoassays are valuable alternatives to the HPLC technique of measuring crosslinks.

The use of biochemical markers in the prediction of osteoporotic fractures has only been studied in postmenopausal women. From a number of epidemiological studies it appears that especially markers of bone resorption predict future fractures independent of BMD [132,133]. Paragraph 3.4 describes the effects of glucocorticoids on markers of bone metabolism.

However additional studies are certainly needed to evaluate whether there markers are really helpful in a better stratification of the patient at risk Therefore the use of bone markers will improve assessing the risk of osteoporosis by BMD measurements.

2.2.2. Biomechanical aspects

The biomechanical competance of bone is for 75-80% explained by bone mineral density as can be measured by densitometry [134]. The remainder is explained by the composition of bone mineral or matrix [135]. Based on this observation one might expect that a rise in BMD will always be related to a reduction in fracture risk. That this is not always the case, is shown by the observations obtained during fluoride treatment [136-138]. Although treatment with this drug induces a considerable rise in BMD, no clear reduction in fracture incidence is observed. The potential explanation for this phenomenon is that fluoride probably increased the crystallinity of bone mineral with a subsequent decrease in elasticity [136]. As a consequence the fragility of bone can even increase. This observation clearly indicates that increases in bone mass can only results in fracture reduction in a case a normal bone matrix is formed. It is clear that such changes can only be assessed by invasive techniques and not by for instance DXA measurement. Although commonly available densitometric techniques can not assess these changes, bone mass measurements provide an accurate indication of vertebral compressive strength as assessed in vitro [139,140].

The amount of bone mineral can be measured in several ways. DXA is widely used nowadays. However, by using DXA only the areal density is measured (g/cm^2). With the use of quantitative computer tomography (QCT-scan) bone is measured in a three-dimensional way and consequently a volumetric density is obtained. Furthermore QCT offers the possibility to differentiate between trabecular and cortical bone. McBroom et al. found that removal of the vertebral cortex was associated with approximately 10% reduction in vertebral load to failure [141-146]. However QCT has the disadvantage of higher radiation exposure and the potential influence on the BMD measurement of changes in marrow fat, that for instance can occur during the use of glucocorticoids [147]. Recently Bolotin also demonstrated inaccuracies in DEXA measurements caused by body fat [148]. However, in general QCT predictions of local trabecular bone material properties appear not to be superior to those measured by DEXA [149].

Another factor which adds to the biochemical competance of bone is the spatial orientation of trabeculae, the so-called microarchitecture of bone. Currently experimental methods are available to obtain more insight in the microarchitecture of bone. It is possible to "measure" architecture by using the complete three-dimensional architectural structure of the trabecular bone measured by an advanced technology of three-dimensional micro computed tomography (micro-CT) scanning [150,151]. This can be used as a highly accurate and automated tool to measure precise changes in bone stereology, volume and projection, and micro-architecture in the evaluation of bone [152]. Unfortunately the

clinical implication of this technique is limited because micro-CT and micro-FEA can only be used in cases where bone biopsies are available.

The suggestion that microarchitecture is an important determinant of bone strength is also indirectly obtained from studies with inhibitors of bone resorption. The observed fracture reduction during treatment with this compounds appear not to be explained by the observed increase of BMD. Although definitive proof is lacking it has been suggested that the inhibition of bone resorption might decrease the chance that trabeculae are perforated, decreasing the loss of connectivity [153].

CHAPTER 3

INHALED GLUCOCORTICOIDS AND THEIR EFFECT ON BONE METABOLISM

3.1. INTRODUCTION

The introduction of inhaled glucocorticoids some 20 years ago was a significant breakthrough in the treatment of asthma limiting systemic side effects.
Inhaled beclomethasone is widely used in daily dosages ranging from 1000 to 2000 µg/d [154,155]. Data from the Prescription Pricing Authority in the UK show a 75% increase in prescriptions for inhaled and systemic glucocorticoids used in respiratory disease from 1991 till 1994. Respiratory diseases are the most common indication for the prescription of corticosteroids both in hospital and general practice. In the general practice setting, there are 40 prescriptions of inhaled steroids for every prescription of orally administered corticosteroids [21]. Threshold inhaled doses for critical adverse events are not known, but these appear to be rare with established effective doses (< 800 µg/day in adults and < 400 µg/day in children) [156]. Nevertheless, it is clear that inhaled glucocorticoids can result in suppression of adrenal function [157,158]. This effect definitely occurs with doses of beclomethasone dipropionate of 2000 µg/day [158], and doses of 3000 µg/day in case of budesonide [159,160]. Decreased circulating levels of osteocalcin [161-163] accompany suppression of cortisol production. High dose inhaled corticosteroids may have similar effects on bone density [164] and bone turnover as seen with orally administered corticosteroids [162,165]. Detrimental effects on bone metabolism, however, are assumed to be much lower with inhaled corticosteroids compared to oral corticosteroids, because of their high topical-to-systemic potencies and their metabolism by the liver [154].

3.2. EFFECTS OF INHALED GLUCOCORTICOIDS ON ADRENOCORTICAL FUNCTION

The occurrence/extent of adrenal suppression is the most extensively studied systemic effect of inhaled corticosteroids. However, even if moderate and high doses of exogenous corticosteroids affect the hypothalamic-pituitary-adrenal (HPA) axis, this rarely appears to be clinically important [166,167].

Nikolaizik showed a dose-dependent nocturnal cortisol suppression by inhaled budesonide of maximal 40%, given either as single evening dose of 400 μg or placebo or 400 μg budesonide twice daily for 2 weeks in healthy persons. Such suppression of the HPA axis was seen after a single inhalation, and there was no evidence of a cumulative effect. The single-dose suppression was completely reversed one day later. The speed of recovery after longer-term treatment could not be established in this study, as some suppression was still apparent 1 week after stopping therapy [168].

Apart from the HPA axis also significant suppression of adrenal androgens (androstenedione and dihydroepiandrosterone (DHEA)) has been described [169]. The clinical significance of such systemic effects, especially in women, is not yet clear and needs to be elucidated in long-term follow-up studies in asthmatic patients. In general, dosages of beclomethasone of budesonide up to 800 μg/day are not accompanied by clinical important suppression of the HPA axis. However, subtle adrenal suppression cannot be ruled out in dosages below 800 μg/day [170], because of different individual susceptibility and differences between inhaled corticosteroids and inhalation devices [171].

Furthermore most of the previous mentioned studies are biased by the use of the 250 μg corticotrophin-releasing test. Recent literature showed that the 1 μg corticotrophin test is more sensitive in discriminating suppression of the adrenal gland in users and non-users of inhaled glucocorticoids [172,173].

Finally, one has to be careful to extrapolate the effects of inhaled glucocorticoids on the HPA axis to their potential effects on other tissues like bone, skin and muscles [174].

3.3. EFFECTS OF INHALED GLUCOCORTICOIDS ON CALCIUM AND PHOSPHATE METABOLISM

Studies on the effects of inhaled corticosteroids on calcium and phosphate homeostasis revealed conflicting results. In a cross-sectional study, Reid et al demonstrated that total body calcium was reduced in subjects receiving inhaled glucocorticoids in a dose of 400 μg/day inhaled beclomethasone or 800 μg/day betamethasone [106]. Inhaled budesonide

3200 μg/day causes alterations in the tubular reabsorption of calcium and phosphate although these changes were considerably smaller than those seen with systemic administration of glucocorticoids [175]. However, another study found increased levels of serum total and ionised calcium in postmenopausal women on 200-2000 μg of inhaled beclomethasone daily [176].

In other studies no significant changes in intestinal calcium absorption, serum calcium, phosphate, and PTH have been documented with the use of high-dose inhaled steroids [67,161,169]. Toogood et al [169] showed that short term use of 600-2400 μg/day budesonide did not influence calcium metabolism even in doses that significantly suppressed the function of the hypothalamic-pituitary-adrenal axis. Luengo [67] also could not find evidence for calcium malabsorption or hyperparathyroidism.

Taken together, inhaled corticosteroids have minimal effects on the calcium and phosphate metabolism. The effects that have been described are dose related. The conflicting results in the different studies could be explained by possible confounding factors such as the different effects of several inhaled steroids, inhalation devices, previous systemic glucocorticoid use and the amount of medication absorbed in the oral-gastric-intestinal route.

3.4 EFFECTS OF INHALED GLUCOCORTICOIDS ON MARKERS OF BONE METABOLISM

Most authors stated that in contrast to oral steroids, inhaled steroids like beclomethasone and budesonide are considered to be free of adverse systemic side effects, if dosages below 1500 microgram are used [67,158,177]. Nevertheless, there is growing concern about the possible adverse effects of inhaled corticosteroids on bone. Studies about their effect on bone turnover, using biochemical indices, suggest that they may depress bone formation as evidenced by depressed serum osteocalcin [163,178] and increased bone resorption as shown by increased hydroxyproline levels [179].

When using bone markers to evaluate the effects of inhaled steroids, we have to keep in mind that not any isolated bone marker can be considered as a reliable guide to the extent of bone formation or resorption. Furthermore the relevance of some markers is not clear. Especially the older markers have problems regarding specificity. Therefore, it is important to have specific immunoassays, to avoid errors, which lead to doubts about the relevance of the used marker [165,177,180,181]. Correct handling of the serum samples is important because proteolytic enzymes in the serum will degrade some markers-like osteocalcin- to fragments.

Several studies have sought to evaluate the bone toxicity of inhaled glucocorticoids by using markers of bone metabolism (See table 3.4). All these studies showed a dose-dependent decrease of serum osteocalcin with inhaled beclomethasone [182]. In healthy volunteer's, serum osteocalcin levels returned to baseline within one week after of discontinuation of the drug [183]. No clear changes were observed in the markers of bone resorption [176].

Jennings et al. [161,162,184] were the only who evaluated the effect of inhaled budesonide, inhaled beclomethasone dipropionate and prednisolone on various indices of bone turnover in healthy volunteers. Serum osteocalcin decreased after the use of budesonide in a daily dose of < 1200 μg. This effect occurred to an even greater extent with equivalent doses of prednisolone and beclomethasone dipropionate. A dose of prednisolone of 20 mg/day may be associated with a 50% reduction in osteocalcin levels [185], which is similar to the effect of high dose inhaled budesonide [162]. Unlike beclomethasone (2,500 μg/day) and prednisolone, budesonide (3,200 μg/day) did not affect serum alkaline phosphatase [184].

Recent studies used the newer markers of bone formation and resorption, like P1CP (Procollagen type 1 C-terminal propeptide), type 1 carboxyterminal telopeptide (ICTP) and (deoxy)-pyridinoline crosslinks. A similar time-related suppression of bone formation, as in osteocalcin, was observed using P1CP in the early phase after starting inhaled glucocorticoids [186].

In conclusion, several short-term studies in healthy non-asthmatic volunteers and asthmatics taking high-dose inhaled steroids (> 1000 μg/day) have demonstrated changes in biochemical markers reflecting a decrease in bone formation (decreased osteocalcin and P1CP) and occasionally an increase in bone resorption (increased hydroxyproline) [161,163,164,178]. Whether these effects remain after long-term use is unclear. However, the only long term study (2,5 years) available at present did not report changes in any of the biochemical markers of bone turnover [187]. The early changes in markers of bone turnover have been directly correlated with the frequency of administering corticosteroids and the total dose used [163,176,178,179,183,188,189]. When inhaled glucocorticoids are used, changes in bone turnover markers were much less pronounced than in the case of oral steroids [169] and were more often seen with beclomethasone than with budesonide, when used at clinically comparable doses [163,189]. Similar dosages may vary in toxicity across individuals, since inter-individual variability has been demonstrated regarding the bronchial absorption of inhaled glucocorticoids [190]. Taken together, the early changes in biochemical markers of bone turnover appear to be related to the frequency and the dose of inhaled glucocorticoid used. Therefore the first principle of treatment remains the use of the lowest possible dose.

Author / Year / Ref	N	Disease	Drug used	Dose	Duration of use	Osteocalcin	Resorption markers	Comments
Teelucksing [183] 1991	8	No disease	Beclo	400-2000 µg/d		decreased	Not determined	After one week of discontinuation, returned to normal
Pouw 1991 [163]	8	No disease	Beclo	2000 µg/d	2 weeks	decreased	Not determined	
Jennings 1991 [161]	12	No disease	Budeso	0.8/1.6/3.2 mg/d		decreased	Not determined	Each dose 1 week
Hodsman 1991 [162]	20	No disease	Budeso	3.2 of 0.8mg/d	1 week	decreased	Not determined	10 received the lower and 10 the higher dose
Toogood 1991 [178]	40	No disease	Budes	1.2/2.4mg/d		decreased	Not determined	In both groups osteocalcin decreased
Ali 1991 [179]	8	No disease	Beclo	2000µg/d	4 weeks	Not determined	OH proline in urine elevated	No changes in the healthy control group, receiving 1800 µg/d. Alk phos decreased.
Puolijoki 1992 [176]	9	Asthma	Beclo	200/1000/2000 µg/d	Each dose three weeks	decreased	Not determined	All postmenopausal women
Kerstjens 1994 [187]	70	Asthma	Beclo	800µg/d	4 weeks and 2.5 years	osteocalcin and PICP decreased in the acute phase	Initial transient elevated ICTP and urinary OH-proline	No effects on bone markers after 2.5 years in this dose.
Puolijoki 1996 [191]	7	Asthma	Beclo	200,1000,2000 µg/d	Each dose 3 weeks	Not determined	PICP and ICTP no change	All postmenopausal women
Toogood 1997 [192]	30	Asthma	Budeso	0.4/0.8/1.6/2.4 mg/d	2 weeks	decreased	Not determined	Also testing two different devices

Beclo = Beclomethasone
Budeso = Budesonide
PICP = carboxy terminal propeptide of type I procollagen
ICTP = cross-linked carboxy terminal telopeptide of type I collagen
N = Number of participants

Table 3.4 Effects of inhaled glucocorticoids on the markers of bone metabolism.

Author / Yr / Ref	Method	N	Medication	Control group	BMD by	Site measured	Results (BMD)	Comments
Reid 1986 [106]	Cross-sectional	22	Beclomethasone 400 µg/d or betamethasone 800µg/d	Healthy	QCT	spine	Decreased.	
Wolff 1991 [201]	Cross-sectional Case control	5	Beclomethasone average dose 326 µg/d	Asthma on systemic GC treatment	SPA DPA	Radius, spine and hip	Decreased only in the control group not in inhaled group.	5 subjects in study and control group !
Packe 1992 [164]	Cross-sectional Case-control	57	Beclomethasone 1000-2000µg/d	Mild Asthma bronchus dilator alone	QCT	Spine	Decreased.	Also compared high dose inhaled with intermittent systemic GC, with high inhaled and continuous low dose systemic GC.
Ip 1994 [202]	Case-control	30	Beclomethasone and budesonide > 1000 µg/d	Healthy	DXA	Spine, femoral neck, trochanter, ward's	All sites decreased compared to control group in female not in male.	Also a correlation between BMI and higher average daily dose. No differences between women with or without previous systemic glucocorticoid use.
Herrala 1994 [203]	Prospective longitudinal clinical trail	19	Beclomethasone 1000 µg/d	Healthy	DXA	Spine, femoral neck, trochanter, ward's	No differences found.	Only postmenopausal women. Careful instructions to minimise GE absorption One year follow up.
Hanania 1995 [204]	Cross-sectional	36	Beclomethasone or budesonide 800 µg/d	18 Asthmatics broncho-dilator therapy alone	DXA	Femoral neck, spine, ward's	Z-score femoral neck decrease. No differences on other sites.	
Packe 1996 [196]	Cross-sectional	40	20 subjects on budesonide 800 µg/d 20 subjects on beclomethasone 1000 µg/d	17 Asthmatic never used inhaled or systemic steroids	QCT	spine	Decreased in both groups using inhaled steroids.	The mean BMD in 7 patients in the budesonide group, who never received oral GC, was approximately mid-way between control and budesonide with GC.
Luengo 1997 [205]	Case-control	48	Beclomethasone and budesonide Mean dose 662 +/- 250 µg/d	Healthy	DXA	Spine	No difference in decrease of BMD between groups.	No effect of systemic GC use during the study on BMD.
Struijs 1997 [195]	Prospective longitudual randomised	33	12 Beclomethasone 800 µg/d 11 budesonide 800 µg/d	10 Asthmatics broncho-dilator therapy alone	DXA	Spine, total hip, Femoral neck,ward	Decreased in all groups. Percent change from baseline in beclo group significant.	

GC = Glucocorticoid; GE = Gastro-Enteral
BMI= Body Mass Index; BMD = Bone Mineral Density
QCT = Quantative Computer Tomography
SPA = Single Photon Absorptiometry
DPA = Dual Photon Absorptiometry
DXA = Dual energy X-ray absoptiometry

Table 3.5 The efffects of inhaled glucocorticoids on BMD.

3.5 EFFECTS OF INHALED GLUCOCORTICOIDS ON BONE MINERAL DENSITY

There are limited data available on the effects of inhaled glucocorticoids on bone mass. As outlined in chapter 1, other risk factors contribute to osteopenia apart from the dose of steroids (see figure 1.2.1) [193,194]. These risk factors may even be of greater relevance when the total dose of steroids is not very high, as in the case of inhaled steroids. Moreover, severe asthma may in itself affect BMD through its effect on the lifestyle factors associated with BMD. Therefore, it is difficult to prove a direct link between the use of inhaled glucocorticoids and BMD. Most studies compare BMD of asthmatic patients treated with inhaled corticosteroids with BMD of healthy adults. This may lead to false conclusions about cause-effect relationships, since it is not known how BMD values would appear in non-steroid-treated asthmatic patients with similar disease severity. Another problem is that the assessment of the effects of inhaled corticosteroids on bone has often been complicated by the fact, that many patients have previously received short- or long-term treatment with oral corticosteroids, which are likely to have had effects on bone turnover.

The studies conducted are summarised in the table 3.5. All studies were done in patients with asthma. The results are difficult to interpret, because different methodologies were used to assess bone mass and differences in past and current use of oral corticosteroids (including mixed oral and inhaled glucocorticoid use). Furthermore other factors like, insufficient numbers of subjects, differences in gender and menopausal status, different delivery systems and the use of too low doses for a too short period of time to expect changes, are involved. In our own study we found a minor decrease of bone mass after one year [195] (See chapter 6). Long-term, controlled studies are needed to clarify the effects of inhaled corticosteroids on bone metabolism and to assess the effects of potential confounding factors such as age, gender, menopausal status, choice of drug, delivery system [171], dosing schedule and duration of therapy.
Also longitudinal studies of bone metabolism and BMD in patients with mild asthma starting inhaled budesonide or beclomethasone therapy are needed to show whether any initial changes already occur before systemic corticosteroid therapy is given [196].
It must be emphasised that the risk of osteoporosis should not defer clinicians from their prescription of inhaled glucocorticoids when indicated. However, recognition of their potential side effects on the skeleton is important. Therefore the lowest possible dose should be prescribed and BMD should be measured in patients at risk.

3.6 FUTURE EXPECTATIONS ON NEWER INHALED GLUCOCORTICOIDS AND THEIR EFFECT ON BONE MASS

Some studies suggested that the newer inhaled corticosteroid fluticasone propionate has a more beneficial ratio of clinical efficacy versus systemic effect [197]. Recent studies showed that in asthmatic patients treatment with 750 μg/day fluticasone was as efficacious as with 1500 μg/day of beclomethasone [198]. Serum markers of bone formation (osteocalcin and PICP) were affected by beclomethasone but not by fluticasone [199]. Recently Pauwels et al [200] suggested that changing existing therapy with inhaled corticosteroids (either beclomethasone or budesonide) to fluticasone in a halved dose, resulted in a significant recovery of bone density in the spine, femoral neck and Ward's triangle. Extensive data in this respect, however, have to be awaited.

CHAPTER 4

THERAPY FOR CORTICOSTEROID-INDUCED OSTEOPOROSIS

4.1 INTRODUCTION

Although glucocorticoids have a wide variety of clinical applications, their use is accompanied by multiple and serious side effects [2], including glucocorticoid-induced osteoporosis which contributes to significant morbidity [3,8,118,119,128]. Until recently, scarce data were available about prevention and treatment strategies in relation to glucocorticoid-induced osteoporotic fractures.

Its obvious that if a therapy is initiated with such a wide spectrum of side effects, as glucocorticoids, one should be aware of the correct indication and the lowest possible dose of such therapy. However, it appears that in a majority of hospitalised patients in the United States, glucocorticoids are not entirely prescribed in accordance with professional standards based on literature [206].

It is generally accepted that supra-physiologic doses of glucocorticoids, especially in the first 3-6 months, provoke an accelerated bone loss followed by a lower rate of loss in the subsequent years [207-211]. Cessation of glucocorticoid therapy after one year results in recovery of the BMD [209]. Therefore timing of initiating or cessation of therapy in glucocorticoid-induced osteoporosis remains difficult and needs further study. Data from van Staa showed that after two months of starting glucocorticoids, even in a dose as low as 2,5 mg/day, an increase in fracture occurs. These data has to be further analysed to find out the underlying mechanism, including non-glucocorticoid related factors [129].

The most prominent pathophysiologic mechanism in glucocorticoid-induced osteoporosis is the uncoupling of bone resorption and formation (Chapter 1). Unfortunately there is no safe and effective therapy yet, stimulating bone formation. There are only a few

prospective studies concerning bone loss and its prevention in patients starting glucocorticoid therapy [208,212-214]. Only three, large prospective, placebo controlled randomised clinical trails, has been performed focussing on the prevention and intervention of steroid-induced osteoporotic fractures. All used bisphosphonates, which are, classified as anti-resorptive agents [215-217]. The ideal treatment might be an anti-resorptive agent in combination with a stimulator of bone formation [218].

4.2. WHO AND WHEN TO TREAT

The ideal strategy in the prevention and treatment of glucocorticoid-induced osteoporosis would be intervention before bone loss and concomitant fractures occur. Such an approach might result in a better quality of life for the individual patient and a potential reduction of costs in medical and social care. The targets of different therapies are showed in figure 4.2.1 however the main problem is how to identify the patients at highest risk. Bone mass measurements figure prominently in guidelines about glucocorticoid-

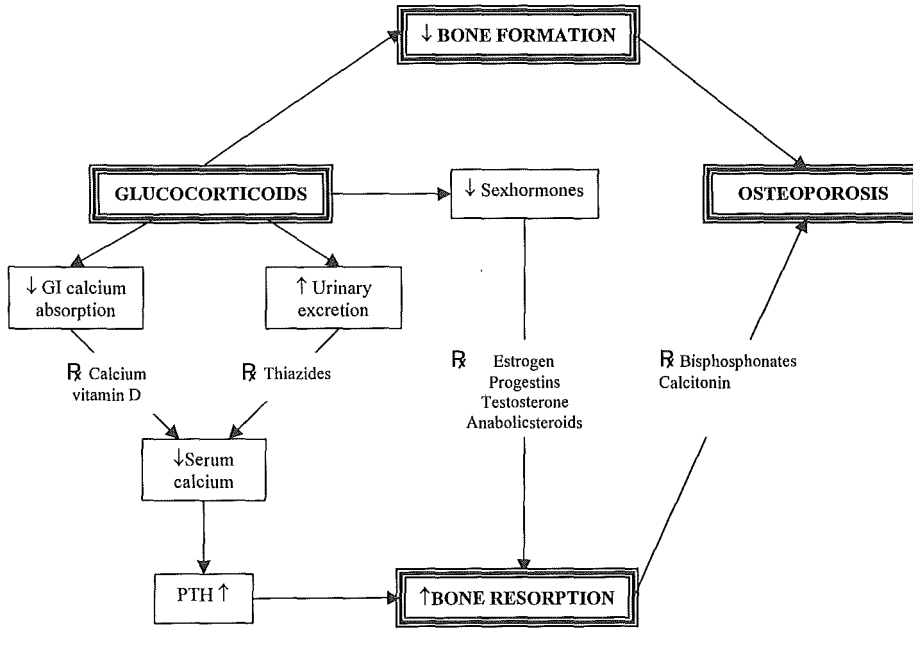


Figure 4.2.1 Treatment options

induced osteoporosis [22,23], however, no prospective data have been obtained indicating that they predict osteoporotic fractures in corticosteroid-induced osteoporosis. Also, the BMD threshold of increased risk of fracture is unclear (See chapter 2). There is some evidence that the BMD threshold is higher in patients with vertebral fractures receiving corticosteroids, compared to those with postmenopausal fractures [16,20,130].
Different cut-off points have been suggested with T scores from -1.0 to -1.5 SD [21-23,218,219]. Eastell's group [23] in their updated review suggests that drug intervention will be targeted at those patients, both men and women, whose BMD is $\geq$ 1 SD below the age-matched mean (Z-score) at spine or hip, or a T-score of -1.5 SD at these sites. They also suggested immediate intervention in those patients who received more than 15 mg prednisolone or equivalent daily. The American College of Rheumatologists suggests intervention when T-score is $\leq$ -1 SD. They also suggest calcium and vitamin D supplementation when initiating glucocorticoid treatment.
In non-steroid-treated patients, a previous history of low trauma fracture is a major risk for future fractures, irrespective of bone density. This is likely also to be the case in steroid treated patients, as previous fracture provides evidence that the skeleton has reached a point at which it is not able to withstand the stresses routinely placed upon it. Therefore patients with previous fractures should usually be offered preventive treatment against further bone loss, irrespective of their BMD. In my opinion the flow-chart from Eastell [23] with some modifications (Figure 4.2.2, page 48) is a useful tool in general practice.

4.3. ALTERNATIVES IN DOSING SCHEDULE AND THE USE OF NEWER GLUCOCORTICOIDS

Bone loss in corticosteroid treated patients is dose-related; therefore it is clear one has to prescribe the lowest effective dose to achieve minimum glucocorticoid exposure. Alternate day therapy preserves normal function of the pituitary-adrenal axis, however it does not prevent bone loss [220-222]. Best therapy is to stop the glucocorticoid treatment. What will happen to bone mass after stopping corticosteroid therapy? Data on this issue are scarce. Recovery of bone mass after cessation of corticosteroid treatment has been described by Rizzato et al and Laan et al [209,223]. However, Laan's study showed that more than 25% had to restart prednisone treatment. Physicians are to optimistic about the duration of treatment with glucocorticoids [224]. Hall et al. [225], performed a study in postmenopausal women with rheumatoid arthritis, 119 patients without previous prednisone treatment, 35 previously on prednisone therapy and 41 patients still on pred-

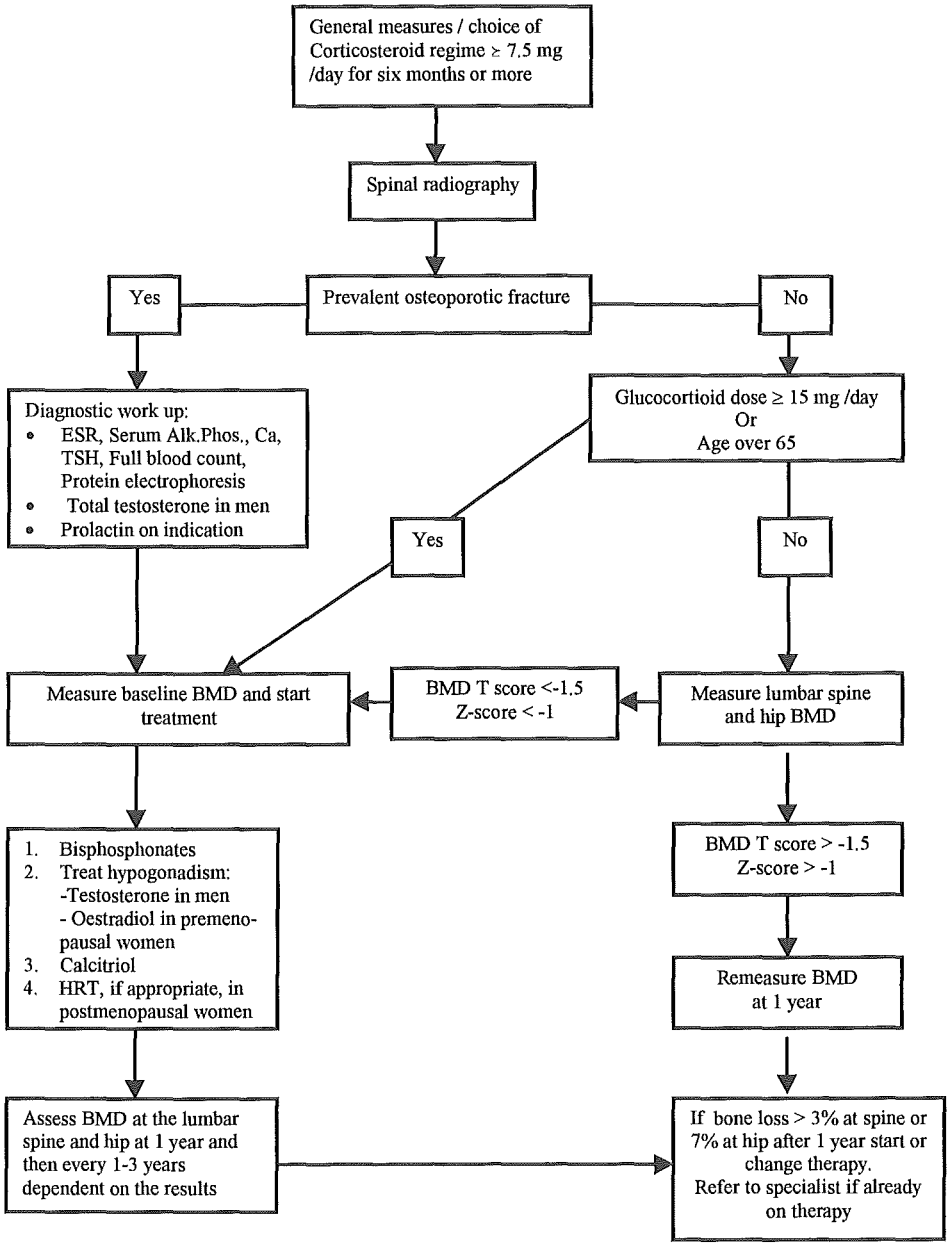


Figure 4.2.2 Prevention and management of glucocorticoid induced osteoporosis.

nisone. He compared them with healthy controls. It was remarkable that no differences in BMD were demonstrated between the groups except those still on glucocorticoids. However a bias is suggested because patients with RA are known to have a lower bone mass as healthy individuals.

The use of newer glucocorticoids, such as deflazacort, could have less toxic effects on bone. Studies showed that it has less inhibitory effect on calcium absorption and produces less bone loss compared to prednisone while maintaining its anti-inflammatory effects [93,226-228]. The crucial question, about which uncertainty remains, is the relative glucocorticoid potency of deflazacort [229-235]. It has been suggested that deflazacort has a significantly lower milligram potency than prednisone and is consequently being used in non-equivalent doses in the comparative studies.

4.4 CALCIUM AND VITAMIN D PREPARATIONS

There is an ongoing discussion about the supplementation of calcium and vitamin D in corticosteroid treated patients. From a pathophysiological point of view, calcium and to a lesser extent vitamin D supplementation are logical therapeutic strategies. Unfortunately the scarce clinical studies on the effect of calcium and/or vitamin D on bone mass in glucocorticoid treated patients are inconclusive [25,76,236-240]. Only calcitriol has been shown to be effective in the prevention of CIOP in patients starting glucocorticoid treatment [208].

It appeared that calcium and vitamin D are essential in bone metabolism and that deficiency of one of them leads to a decreased efficacy of other medication used in glucocorticoid-induced osteoporosis. In a clinical trial Mulder et al [241] showed that reduction in serum calcium and vitamin D result in a diminished effect of etidronate given to osteopenic postmenopausal women. Therefore basal serum calcium and vitamin D levels should be measured in patients using glucocorticoids, and eventually supplemented. Calcium intake of patients starting glucocorticoid treatment should be at least 1000-1500 mg a day.

400 IU/day of vitamin D is recommended for the average patient in whom there is no reason to suspect vitamin D deficiency. In those patients who may be at risk for vitamin D deficiency (malabsorption and/or low diet vitamin D), serum level of 25-hydroxyvitamin D should be measured.

The use of thiazide diuretics and sodium restriction has been shown to reduce calcium excretion [82,242,243]. However, these data must be interpreted with caution because no randomised controlled trial has been published in glucocorticoid treated patients. Fur-

thermore, thiazide diuretics may aggravate the loss of potassium in corticosteroid treated patients. Hypercalcemia may develop in patients treated with a combination of vitamin D and thiazides. Therefore serum calcium levels should be monitored [244].

4.5 CALCITONIN

Calcitonin is not registered for glucocorticoid-induced osteoporosis in the Netherlands. However, it is an inhibitor of bone resorption and some studies also suggest that it stimulate osteoblasts [245] and promotes enteral calcium absorption [246]. As its effect match the pathogenic factors involved in steroid induced osteoporosis, and suppression of endogenous calcitonin secretion has been described during glucocorticoid therapy [247, 248], on first site it seems a rational therapy. However, studies performed with calcitonin are conflicting. This is due to insufficient sample size, heterogeneity of study populations, retrospective designs [214], poor steroid dose control [249], different routes of administration and reliance on short term changes in surrogate measures of osteoporosis [250]. The mentioned positive effect on calcium absorption is an indirect effect, pertinent to all anti-resorptive drugs. They all decrease serum calcium by decreasing the efflux of calcium from the skeleton (see chapter 1). The results of the three randomised-controlled trials published [208,251,252], are conflicting. Healy et al. [252] and Sambrook et al. [208] found no beneficial effect, Adachi et al. [251] however, reported a significant increase in bone mass after one year of therapy. This may be due to differences in patient populations; timing and dose of steroids used, and differences in co-treatment with calcium and vitamin D in the study and control groups. Overall, in the first year, calcitonin treatment seems to have a positive effect on bone mass in prevention and treatment of osteoporosis. After one year the effects are similar to those in the control group treated with calcium and vitamin D. A useful property of calcitonin is its analgesic effect in the acute phase of vertebral fracture [253]. However, for this purpose cheaper alternatives are available. The lack of proven efficacy on BMD and fracture reduction is the major disadvantage and does not support the use of calcitonin in daily clinical practice.

4.6. SEX HORMONE REPLACEMENT

Unlike the effect on BMD in postmenopausal osteoporosis, data on the effect of hormonal replacement therapy on BMD in glucocorticoid-induced osteoporosis are very limited. Furthermore no fracture studies has been published. It is believed that in premenopausal

regularly menstruating women, HRT has no place. Nevertheless the use of oestrogen replacement in oestrogen-deficient women who are receiving high doses of corticosteroids has been recommended, as long as its use is compatible with other medical conditions of the patients [21-23].
Different mechanisms of actions by which HRT acts in CIOP have been described. First of all HRT results in inhibition of bone resorption. Furthermore, Greenberg et al. suggested that 17 beta-estradiol (E2) and progesterone stimulate calcitonin secretion by rapid, direct and specific effects on thyroid C cells in rats. Therefore, gonadal hormones might have an additional effect in inhibiting bone resorption through a direct effect on calcitonin secretion [254]. An overview of studies with HRT in glucocorticoid induced osteoporosis is provided in table 4.6.1 (page 52). In one of the few studies of HRT in female RA patients using steroids [255], a small subgroup of 4 steroid treated patients was taking estrogens alone (Estraderm® 50), because of hysterectomy. It appeared that these patients lost significantly more bone than the remaining patients who took combined HRT consisting of transdermal estradiol 50 µg daily with oral norethisterone. The recognised competition for osteoblast glucocorticoid receptors [33] and the positive results of a trial with progesterone in steroid-induced osteopenia [256,257], suggest that progesterone may be a more important component in HRT when it is used to treat patients who have received long-term glucocorticoid treatment. Further research on the effects of HRT on BMD and fracture risk, in glucocorticoid treated patients are necessarily, because the studies done were to small or not randomised.
Taken together clear evidence for a beneficial effect of HRT in the treatment of glucocorticoid-induced osteoporosis is lacking. The use of HRT in CIOP should be limited to proven hypogonadism in pre-menopausal woman, and can be considered in the first years after menopause.
In steroid treated men hypogonadism frequently occurs [29,102,258]. In the knowledge that hypogonadism per se accelerates bone loss [259-262], it seems reasonable to provide testosterone replacement to these individuals, even though data confirming this are very scarce. Reid reported, in a randomised crossover trial, that testosterone appeared to be effective as a therapy for low bone mass in 15 asthmatic men treated with glucocorticoids. Lumbar spine, but not hip BMD increased after one year of treatment. Also lean body mass increased. General guidelines, for the use of androgens in men, have been published recently [263].

Anabolic steroids have also been used in the treatment of glucocorticoid-induced osteoporosis. They have little place in the management of steroid induced osteoporosis in men, because they would reduce endogenous testosterone concentrations further. Their

Auth./ Yr/ref	Type study	N	Follow-up	Study medication	Results BMD	Comments
Hall 1994 [255]	Sub-analysis Of steroid treated subjects	21	24 months	Transdermal estradiol 50µg daily with oral nor-ethisterone 1 mg 12 days per month or Estraderm 50 after hysterectomy	Measured by DXA Lumbar spine +3.5 % Prox femur + 1.62 %	200 postmenopausal women with RA, on chronic GC use.
Studd 1989 [266]	Not mentioned, probably prospective	4	12 months	Oestradiol 75 mg and testosterone 100 mg percutaneous implant	Measured by DPA Spine + 13.4 % Hip +6.25%	Postmenopausal women with asthma. Longer than 5 years 5mg prednisone a day
Lukert 1992 [213]	Retrospective	15	12 months	8 patients receive 0.6256 mg Premarin daily for 25 days and 5 mg daily of medroxyprogesterone on days 15-25	One patient DXA And 7 patients DPA Spine + 4.1%	Postmenopausal or amenorrheic women treated with prednisone because of asthma
Sambrook 1992 [267]	Subanalysis of five patients	38	48 months	Different estrogen regimes	Measured by DPA Spine +0.7 % Hip non significant	38 postmenopausal women with RA, mean dose 6.2 mg / day
Grecu 1990 [256]	Prospective randomised controlled clinical trail	23	12 months	13 patients receive 200 mg medroxyprogesterone acetate at 6 weeks intervals and 1 gram of elemental calcium.	measured by QCT Spine +17 %	Asthmatic men, mean age 66, treated with 10-20 mg prednisone a day, for over 1 year
Reid 1996 [268]	Prospective randomised cross-over clinical trail	15	Two times 12 month	30 mg testosterone propionate, 60mg phenylprionate, 60 mg isocaproate, 100 mg decanoate, 250 mg/month IM depot injection	measured by DXA Lumbar spine +5.0 %	Asthmatic men, mean age 61, treated with 11.6 mg prednisone a day, during 8 years
Adami 1991 [264]	Prospective randomised controlled clinical trail	35	18 months	18 patients receive 50 mg nandrolone decanoate every 3 weeks IM and a diet with 1200 mg Ca and 600-800 IU vitamin D	measured by DPA Distal forearm + 5.1%	Women, 1 pre-, 17 postmenopausal treated with 10mg prednisone a day > 1 year because of rheumatic diseases

RA = Rheumatoid Arthritis; GC = Glucocorticoid; IM = Intra-muscular; BMD = Bone Mineral Density; QCT = Quantatative Computer Tomography; DPA = Dual Photon Absorptiometry
DXA = Dual energy X-ray absoptiometry

Table 4.6.1 Studies with sex-steroids in glucocorticoid-induced osteoporosis.

use in women is associated with beneficial effects on bone mass [264], but also with virilising side effects in 10-50% [264,265] of treated patients. The lack of controlled trials with adequate measurements of BMD at appropriate places, the virilising potential and the adverse effect on the lipid profile, make these drugs not advisable for use in steroid induced osteoporosis.

4.7 FLUORIDE

Presently, sodium fluoride is the only registered stimulator of bone formation [269,270]. There are several studies suggesting a positive effect on spinal BMD in both the prevention and treatment of steroid-induced osteoporosis [271-277]. Only the recent studies performed by Lems et al [271, 277] are randomised controlled prospective trials. However in post-menopausal osteoporosis fluoride has been associated with deterioration of bone quality and increased fracture risk [138,278]. This effect appears to be dose dependent as shown by others using lower dosages which resulted in a lower fracture incidence in postmenopausal osteoporosis [279,280]. So it appears that the therapeutic window for fluoride is small and its adverse effects are dependent on formulation and dosage. Slow release sodium fluoride appears to have no side effects when given intermittently [281].
In conclusion, the beneficial effects of fluoride on trabecular bone -the site of the greatest bone loss in corticosteroid treated patients- are counter-balanced by its interference with normal mineralisation when present in high concentrations (see also paragraph 2.2.1). Its therapeutic window and anti-fracture ability in both post-menopausal and corticosteroid-induced osteoporosis remain to be established. All together fluoride is not recommended as a first-line agent in steroid induced osteoporosis.

4.8. BISPHOSPHONATES

As earlier stated increased bone resorption induced by glucocorticoid use, could be counteracted by anti-resorptive agents such as calcitonin, hormone replacement therapy or bisphosphonates. In this paragraph the use of bisphosphonates will be discussed.

4.8.1 Possible mechanisms of action

Bisphosphonates adsorb to bone mineral and inhibit bone resorption. Physico-chemical, intracellular and extracellular mechanisms are involved. Bisphosphonates act by being

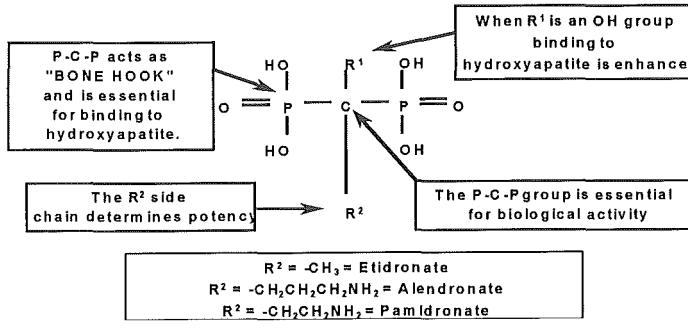


Figure 4.8.1 Biochemical structure and biological activity of bisphosphonates. (With kind permission from RGG Russell)

selectively adsorbed to mineral surfaces in bone and then taken up by osteoclasts. Thereafter they will interfere with recruitment, differentiation and resorptive activity of the osteoclast. The biochemical structure of a bisphosphonate comprises different parts with specific actions, see figure 4.8.1. [282]

It is likely that bisphosphonates are internalised by osteoclasts, during the process of bone resorption. They interfere with specific biochemical cell processes in- and induced apoptosis of- the osteoclasts. Recently Luckman et al [283,284] published the two possible mechanisms by which different bisphosphonates works. Bisphosphonates that closely resemble pyrophosphate, like clodronate and etidronate, can be metabolically incorparated into nonhydrolysable analogues of ATP that may inhibit ATP-dependent intracellular enzymes. In nitrogen-containing bisphosphonates, such as alendronate and risedronate, the effects on osteoclast function are mediated by inhibition of different enzymes of the mevalonate pathway. Subsequently prenylation of different key regulatory proteins is inhibited which causes loss of osteoclast activity and induction of apoptosis.
Bisphosphonates are considered as anti-resorptive compounds. In clinical studies it appeared that they are able to increase bone mass and reduce fracture rates. An explanation of these findings could be, that in addition to the previous described actions on the osteoclasts, the ability of bisphosphonates to reduce the activation frequency and birth rates of new BMU's and the enhancement of the osteon mineralisation all contribute to

the increase of bone mass and the reduction of fracture rate. Another interesting possibility is that bisphosphonates decrease resorption less than formation at the individual BMU level and, therefore, increase bone balance at this site [285-291].
Also the suggestion that alendronate to some extent increases bone formation at the level of the BMU in ovariectomised baboons, is an interesting finding [292]. A similar effect has been suggested in humans treated with clodronate [285,293]. Although certainly not conclusive, our own study described in chapter 10 could also support an anabolic effect on the skeleton.
Another possible mechanism of action of bisphosphonates is the immune modulating effects of bisphosphonates and the concomitant effect on the underlying disease. These are both factors that are especially involved in glucocorticoid-induced bone loss [294,295].
Another interesting observation is that the rather limited increase of BMD in postmenopausal patients with osteoporosis treated with bisphosphonates, substantially underestimates the fracture reduction demonstrated in clinical studies. It has been suggested that the observed reduction in fractures caused by bisphosphonates could only be explained for 30 to 50% by the increase of BMD. The additional effects of bisphosphonates on fracture reduction are not determined yet, but an additional effect of bisphosphonates on the quality of bone has been suggested [153,296].

4.8.2 Clinical experience with bisphosphonates

Compared to postmenopausal osteoporosis, the studies on prevention and treatment of glucocorticoid-induced osteoporosis with bisphosphonates are scarce. Homik and others recently did a database search, on controlled clinical trials concerning glucocorticoid-induced osteoporosis [297]. They found 23 controlled studies [210-212,215,216,298-315] assessing the treatment of CIOP. After selecting them for prospectiveness, mean dose of 7,5 mg prednisone / day or more, and adequate BMD measurements, 13 remained with 842 participants (See table 4.8.1 and 4.8.2, page 56 and 57). The efficacy of bisphosphonates, measured as percent change in BMD over one year ranged from -10% to +19% [212,303]. These results are heterogeneous; however, the studies dealt with heterogeneous patients with many different disease entities requiring glucocorticoids. The study of van Cleemput et al. [303], which is responsible for the 10% decrease in bone mass under bisphosphonate treatment was performed in a group of cardiac transplants with probably a vitamin D deficiency. As mentioned before bisphosphonates cannot act without sufficient supply of calcium and vitamin D [241]. Furthermore the methods of measuring bone mass were quite different in the different studies. The BMD data were collected by QCT, DPA and DXA. The results obtained with these methods can not be compared with each other.

Author Yr./ Ref	Study Design	Medication	Control Group Rx	Follow -up (year)	Diseases	N treat	N plac	Results BMD Treatment group vs. (control group)	Comments
Mulder 1994[210]	P.R.C.	Cyclic Etidronate	Nothing	1 year	Temporal Arteritis	10	10	Measured by DXA Spine + 1.4 % (- 5.0%)	
VanCleemput 1996[303]	P.R.C	Cyclic Etidronate	Calcium Vitamin D	1 year	Cardiac Transplan-tation	19	22	Measured by DXA Spine –10.3% (-7.0%) Fem Neck –8.9% (-5.6%)	Set up as a comparison between two treatments
Adachi 1997[216]	P.R.PI C.DB.	Cyclic Etidronate	Placebo and calcium	1 year	RA, PMR	54	62	Measured by DXA Spine +0.6% (- 3.7%) Fem Neck +0.2 (-1.7%)	Fracture reduction in subgroup of PMP women
Wolfhagen 1997[313]	P.R.C.	Cyclic Etidronate and calcium	Calcium	1 year	Primary Biliary Cirrhosis	6	6	Measured by DXA Spine +0.4% (-3.0%) Fem neck –0.1% (-1.5%)	Possible vitamin D disorder
Boutsen 1997[314]	P.R.C	Pamidronate and calcium	Calcium	1 year	RA,PMR	15	12	Measured by DXA Spine +3.6% (-5.3%) Fem Neck +2.2% (-5.3%)	No explanation for the drop outs
Roux 1998[211]	P.R.PI C.DB	Cyclic Etidronate and calcium	Placebo and calcium	1 year	RA, PMR, Vasculitis, SLE	51	56	85 patients measured by DXA and 32 by DPA Spine + 0.3% (-2.8%) Fem Neck –1.3% (-2.6%)	Two different methods of BMD measurements and all subjects were uncontrolled allowed to take vitamin D up to 1000 IU / day
Jenkins 1999[307]	P.R.PI C,DB	Cyclic Etidronate and calcium	Placebo and calcium	1 year	RA, PMR	15	13	Measured by DXA Spine +1.8% (-3.7%)	
Reid 1999[217]	P.R.PI C.DB	Risedronate pooled data 2.5 and 5 mg daily and calcium	Placebo and calcium	1 year	RA, PMR, SLE	228	170	Measured by DXA Spine + 4.2 % in men Premenopausal women + 2.5 %, postmenopausal women + 3.8 %	Preliminary data without data on the control group

RA = Rheumatoid Arthritis; GC = Glucocorticoid; IM = Intra-muscular; BMD = Bone Mineral Density; QCT = Quantatative Computer Tomography; DPA = Dual Photon Absorptiometry
DXA = Dual energy X-ray absoptiometry

Table 4.8.1 Clinical trials concerning primary prevention of glucocorticoid-induced osteoporosis with bisphosphonates.

In the meta-analysis by Homik et al [297], a statistically significant improvement in lumbar BMD with a weighted mean difference of approximately + 4% was found. Also a statistically significant effect on femoral BMD was observed, although to a lesser extent (+2.1%). Although there was a 24% reduction in spinal fractures, it did not reach statistical significance because most studies are not powered to draw conclusions about fracture risk and incidence. Recently Adachi et al [216] found a statistical significant reduction of

Author Yr./ Ref	Study Design	Medication	Control Group Rx	Follow -up (year)	Diseases	N treat	N plac	Results BMD Treatment group vs. (control group)	Comments
Reid 1988[212]	P.R.Pl C	Pamidronate	placebo	1 year	Asthma Collagen vascular diseases	16	19	Measured by QCT Spine +19.6 % (-8.8 %)	QCT is not comparable to DXA in percentages gain or loss
Skingle 1994[301]	P.R.C	Etidronate and calcium	Calcium	2 years	PMR, COPD Temporal arteritis	18	20	Measured by DXA Spine +4.1% (-0.8%) Fem Neck –1.0% (-1.0%)	A lot of dropouts because of too low dose of prednisolone
Worth 1994[299]	P.R.C.	Etidronate 7.5 mg/kg/day, 1000 IU vit D and calcium	Calcium	½ year	Asthma	14	19	Measured by DPA Spine +5.5% (-4.6%)	Different dosage regimen, only asthmatics BMD measured by DPA
Struijs 1995[300]	P.R.C.	Etidronate and calcium	Calcium	1 year	Asthma, COPD, temporal arteritis	19	20	Measured by DXA Spine +5.7% (-3.4%) Fem Neck +6.8% (-4.1 %)	Extremely osteoporotic patients, using Z-score
Eastell 1996[311]	P.R.Pl C.DB	Risedronate 2.5 mg/day	Placebo	2 years	RA	40	40	Measured by DXA Spine +1.4% (-1.6%) Fem Neck -1.0% (-3.6%)	Only published in abstract
Eastell 1996[311]	P.R.Pl C.DB	Cyclic Risedronate 15 mg / day	Placebo	2 years	RA	40	40	Measured by DXA Spine -0.1% (-1.6%) Fem Neck +0.9% (-3.6%)	Only published in abstract
Saag 1998[215]	P.R.Pl C.DB	Alendronate 5 or 10 mg/day	Placebo Calcium vitamin D	1 year	RA, PMR Asthma, SLE, pemphigus	137	134	Measured by DXA Spine +2.2%, Fem Neck – 0.03% in the 5 mg group Spine +2.78%, Fem Neck +0.22% in 10mg group. The control group spine (-0.04%), Fem Neck (-1.7%)	Both groups received 800-1000 mg of calcium/day and Vitamin D 250-500 IU/day.
Liberman 1999	P.R.Pl C.DB	Alendronate, 5mg or 10 mg Or 2,5mg in first year followed by 10 mg second year	Placebo calcium vitamin D	2 years	RA, PMR Asthma, SLE, pemphigus	134	51	Measured by DXA Spine +2.86%,Fem Neck +0.01 % in 5mg group Spine +3.86%, Fem Neck +0.62 % in 10 mg group Spine +3.85%, Fem Neck -0.63% in 2.5/10mg group Spine (–0.91%), Fem Neck (–2.98%) in placebo group	205 patients of the initial study entered the extension study and both groups received Vitamin D (250-500IU) and (800-1000) calcium daily. Only abstract published
Reid 1999[217]	P.R.Pl C.DB	Risedronate 5 and 2.5 mg daily and calcium	Placebo calcium and vitamin D	1 year	RA, PMR, SLE	290	?	Measured by DXA Spine +2.9% (+0.4%) Fem Neck +1.8% (-0.2%)	Statistical significant reduction of vertebral fractures. Preliminary data

Table 4.8.2 Clinical trials concerning secondary prevention of glucocorticoid-induced osteoporosis with bisphosphonates.

new fractures in postmenopausal women on corticosteroid use, treated with etidronate. Similar results have recently been reported regarding risedronate and alendronate [217,291].

REFERENCES

1. Cushing H, The basophil adenomas of the pituitary and their clinical manifestations. Bull. John Hopkins Hosp, 1932. 1: p. 137-195.
2. Boland EW and Hedley NE, Management of rheumatoid arthritis with smaller doses of cortisone acetate. JAMA, 1950. 144: p. 36-70.
3. Khosla S, Lufkin EG, Hodgson SF, Fitzpatrick LA, and Melton LJ, Epidemiology and clinical features of osteoporosis in young individuals. Bone, 1994. 15: p. 551-555.
4. Lukert B, Glucocortcoid-induced osteoporosis. Osteoporosis, ed. Marcus R, Feldman D, and Kelsey J. 1996, New York: Academic Press. 801-820.
5. Meunier PJ, Is steroid-induced osteoporosis preventable? N Eng J Med, 1993. 328: p. 1781-1782.
6. Walsh LJ, Wong CA, and Pringle M, A. Use of oral corticosteroids in the community and the prevention of secondary osteoporosis: a cross sectional study. Br Med J, 1996. 313: p. 344-346.
7. Heerdink ER, Clustering of drug use in the elderly, 1995: Utrecht.
8. Hahn TJ, Drug-induced disorders of vitamin D and mineral metabolism. Clin Endocrinol Metab, 1980. 9: p. 107-129.
9. Hooyman JR, Melton LJ, Nelson AM, O'Faloon WM, and Riggs BL, Fractures after rheumatoid arthritis. A population-based study. Arthritis Rheum, 1984. 27: p. 1353-1361.
10. Cooper C, Coupland C, and Mitchell M, Rheumatoid arthritis, corticosteroid therapy and the risk of hip fracture. Ann Rheum Dis, 1995. 54: p. 49-52.
11. van Staa TP, Cooper C, Abenhaim L, Begaud B, and Leufkens H, Individual susceptibility to steroid-induced fractures. Calcif Tiss Int, 1999. 64(Suppl 1): p. S38.
12. Wasnich RD, Ross PD, Heilbrun LK, and Vogel JM, Prediction of postmenopausal fracture risk with use of bone mineral measurements. Am J Obstet Gynecol, 1985. 153: p. 745-751.
13. Hui SL, Slemenda CW, and Johnston CC Jr, Baseline measurement of bone mass predicts fracture in white women. Ann Intern Med, 1989. 111: p. 355-361.
14. Black DM, Cummings SR, Genant HK, Nevitt MC, Palermo L, and Browner W, Axial and appendicular bone density predict fractures in older women. Bone Min Res, 1992. 7: p. 633-638.
15. Marshall D, Johnell O, and Wedel H, Meta-analysis of how well measures of bone mineral density predict occurrence of osteoporotic fractures. Br Med J, 1996. 312: p. 1254-1259.
16. Luengo M, Picado C, Rio LD, Guanabens N, Montserrat JM, and Setoin J, Vertebral fractures in steroid dependent asthma and involutional osteoporosis: a comparative study. Thorax, 1991. 46: p. 803-806.
17. Osteoporosis. in Fourth international symposium on osteoporosis and consensus development conference. 1993. Hong Kong.

18. Consensus Development Conference: Prophylaxis and Treatment of Osteoporosis. Am J Med, 1991.90: p. 107-110.
19. WHO Study Group, Assessment of fracture risk and its application to screening for osteoporosis. 1994, Geneva: World Health Organisation.
20. Banffer D, Zwinderman AH, Papapoulos SE, and Hamdy NAT, Bone mineral density measurements and the prevalence of vertebral fractures in long-term glucocorticoid users. Calcif Tiss Int, 1999. 64(Suppl 1): p. S73.
21. Eastell R, Management of corticosteroid-induced osteoporosis. J Int Med, 1995. 237: p. 439-447.
22. ACR Task Force on Osteoporosis Guidelines, Recommendations for the prevention and treatment of glucocorticoid-induced osteoporosis. Arthritis Rheum, 1996. 39(11): p. 1791-1801.
23. Eastell R, Reid DM, Compston J, Cooper C, et al., A UK consensus group on management of glucocorticoid-induced osteoporosis: an update. J Int Med, 1998. 244: p. 271-292.
24. Olbright T and Benker G, Glucocorticoid -induced osteoporosis: pathogenesis, prevention and treatment, with special regard to the rheumatic diseases. 1993. 234: p. 237-244.
25. Lukert BP and Raisz LG, Glucocorticoid-induced osteoporosis: pathogenesis and management. Ann Int Med, 1990. 112: p. 352-364.
26. Adachi JD, Bensen WG, and Hodsman AB, Corticosteroid-induced osteoporosis. Semin Arthritis Rheum, 1993. 22: p. 375-384.
27. Hosking DJ, Effects of corticosteroid on bone turnover. Respir Med, 1993. 87(suppl A): p. 15-20.
28. Healy JH, Glucocorticoid-induced osteoporosis. Curr Opinions Orthop, 1994. 5: p. 33-38.
29. Reid IR, Francis JT, Pybus J, and Ibbertson HK, Low plasma testosterone levels in glucocorticoid-treated male asthmatics. BMJ, 1985. 291: p. 574.
30. Reid IR, Veale AG, and France JT, Glucocorticoid osteoporosis. J Asth, 1994. 31: p. 7-18.
31. Manolagas SC and J. RL, Bone marrow, cytokines, and bone remodeling: Emerging insights into the pathophisiology of osteoporosis. N Engl J Med, 1995. 332: p. 305-311.
32. Dempster DW, Arlot MA, and Meunier PJ, Mean wall thickness and formation periods of trabecular bone packets in corticosteroid -induced osteoporosis. Calcif Tis Int, 1983. 35: p. 410-417.
33. Feldman D, Dziak R, Koehler R, and Stern P, Cytoplasmic glucocorticoÔid binding proteins in bone cells. Endocrinology, 1975. 96: p. 29-36.
34. Bressot C, Meunier PJ, Chapuy MC, Lejeune E, Edouard C, and Darby AJ, Histomorphic profile, pathophysiology and reversibility of corticosteroid-induced osteoporosis. Metab Bone Dis Rel Res, 1979. 1: p. 303-311.
35. Lund B, Storm TL, Melsen F, Mosekilde L, Andersen RB, Egmose C, and Sorensen OH, Bone mineral loss, bone histomorphometry and vitamin D metabolism in patients with rheumatoid arthritis on long-term glucocorticoid treatment. Clin Rheum, 1985. 4: p. 143-149.
36. Canalis E, Effect of cortisol on periosteal and nonperiosteal collagen and DNA synthesis in cultured rat calvariae. Calcif Tiss Int, 1984. 36: p. 158-166.
37. Chyun YS, Kream BE, and Raisz LG, Cortisol decreases bone formation by inhibiting periosteal cell proliferation. Endrocrinology, 1984. 114: p. 477-480.
38. Wong M-M, Rao LG, Ly H, Hamilton L, Tong J, Sturtridge W, McBroom R, Aubin JE, and Murray TM, Long-term effects of physiologic concentrations of dexamethasone on human bone derived cells. J Bone Miner Res, 1990. 5: p. 803-813.
39. McCulloch CAG and Tenebaum HC, Dexamethasone induces proliferation and terminal differentiation of osteogenic cells in tissue culture. Anatomical Record, 1986. 215: p. 397-402.

40. Hodge BO and Kream BE, Variable effects of dexamethasone on protein synthesis in clonal rat osteosarcoma cells. Endocrinology, 1988. 122: p. 2127-2133.
41. Delany AM, Dong Y, and Canalis E, Mechanisms of glucocorticoid action in bone cells. J Cell Biochem, 1994. 56: p. 295-302.
42. Chevalley T, Strong DD, Mohan S, Baylink DJ, and Linkhart TH, Evidence for a role for insuline-like growth factor binding proteins in glucocorticoid inhibition of normal human osteoblast-like cell proliferation. Eur J Endocrin, 1996. 134: p. 591-601.
43. Gabbitas B and Canalis E, Cortisol enhances the transcription of insulin-like growth factor-binding protein-6 in cultured osteoblasts. Endocrinol, 1996. 137: p. 1687-1692.
44. Lukert B, Mador A, Raisz LG, and Kream BE, The role of DNA synthesis in the response of fetal rat calvariae to cortisol. J Bone Min Res, 1991. 6: p. 453-460.
45. Morrison NA, Shine J, and Fragonas JC, 1.25-dihydroxyvitamin D-responsive element and glucocorticoid repression in the osteocalcin gene. Science, 1989. 146: p. 1158-1161.
46. Chen TL, Mallory JB, and Hintz RL, Dexamethasone and 1,25 dihydroxyvitamin D_3 modulate the synthesis of insulin-like growth factor-I in osteoblast-like cells. Calcif Tiss Int, 1991. 48: p. 278-282.
47. McCarthy MB, Doherty WJ, and Gronowicz GA, Glucocorticoids inhibit integrin expression in osteoblasts. J Bone Mineral Res, 1992. 7(suppl. 1): p. S97.
48. Gallagher JA, Beresford JN, MacDonald BR, and Russel RGG, Hormone target cell interactions in human bone, in Osteoporosis, C. C, Editor. 1984: Glostrup Hospital. p. 431-439.
49. Dietrich JW, Canalis EM, Maina DM, and Raisz G, Effects of glucocorticoids on fetal rat bone collagen synthesis in vitro. Endocrinology, 1979. 104: p. 715-721.
50. Adler RA and Clifford JR, Glucocorticoids and osteoporosis. Edocrinol Metab Clin North Am, 1994. 23: p. 641-654.
51. Frost HM, Bone remodeling dynamics, ed. Charles C Thomas. 1963: Springfield.
52. Frost HM, Bone remodeling and its relationship to metabolic bone disease, ed. C.C. Thomas. 1973: Springfield.
53. Jaworski ZFG, Physiology and pathology of bone remodeling. Orthop clinics North America, 1981. 12(3): p. 485-551.
54. Parfitt AM, The physiological and clinical significance of bone histomorphometric data, in Bone histomorphometry: techniques and interpretation, R. RR, Editor. 1983, CRC Press. p. 143-223.
55. Frost HM, Intermediary organization of the skeleton. 1986: CRC Press.
56. Baron R, Importance of the intermediate phases between resorption and formation in the measurement and understanding of the bone remodeling sequence, in Bone histomorphometry: proceedings of the second international workshop, M. PJ, Editor. 1977, Société de la Nouvelle Imprimerie Fournie: Toulouse. p. 179-183.
57. Eriksen EF, Normal and pathological remodeling of human trabecular bone: three dimensional reconstruction of the remodeling sequence in normal and in metabolic bone disease. Endocrine Rev, 1986. 7: p. 379-408.
58. Aaron JE, Francis RM, Peacock M, and Makins NB, Contrasting micro-anatomy of idiopathic and corticosteroid-induced osteoporosis. Clin Orthop and related research, 1989. 143: p. 294-305.
59. Dempster DW, Bone histomorphometry in glucocorticoid-induced osteoporosis. J Bone Min Res, 1989. 4: p. 137-141.
60. Hahn TJ and Halstead LR, Cortisol enhancement of PTH-stimulated cyclic AMP accumulation

in cultured fetal rat long bone rudiments. Calcif Tiss Int, 1979. 29: p. 173-175.
61. Jowsey J and Riggs BL, Bone formation in hypercortisolism. Acta Endocrinologica, 1970. 63:p. 21-28.
62. Meunier PJ, Dempster DW, Edouard C, Chapuy C, Arlot M, and Charhon S, Bone histomorphometry in corticosteroid-induced osteoporosis in Cushing's syndrome. Advances in Exp Med Biol, 1984. 171:p. 191-200.
63. Schachter D and Rosen SM, Active transport of 45 Ca by the small intestine and its dependence on vitamin D. Am J Physiol, 1959. 196: p. 357-362.
64. Klein RG, Arnaud SB, Gallagher JC, Deluca HF, and Riggs BL, Intestinal calcium absorption in exogenous hypercortisolism: role of 25-hydroxy vitamin D and corticosteroid dose. J Clin Invest, 1977. 64: p. 655-665.
65. Gennari C, Bernini M, and N. P, Glucocorticoids: radiocalcium and radiophosphate absorption in man, in Osteoporosis, a Multidisciplinary Problem, ASJ Dixon, Russel RGG, and Stamp TCB, Editors. 1983, Royal Society of Medicine: London.
66. Lukert BP and Adams JS, Calcium and phosphorus homeostasis in man: Effect of corticosteroids. Arch Int Med, 1976. 136: p. 1249-1253.
67. Luengo M, Picado C, Piera C, Guanabens N, Montserrat J, Rivera J, and Setoin J, Intestinal calcium absorption and parathyroid hormone secretion in asthmatic patients on oral or inhaled steroid treatment. Eur Resp J, 1991. 4: p. 441-444.
68. Morris HA, Need AG, O'Loughlin PD, Horowitz M, Bridges A, and Nordin BEC, Malabsorption of calcium in corticosteroid-induced osteoporosis. Calcif Tiss Int, 1990. 46: p. 305-308.
69. Colette C, Monnier L, Herbute NP, Blotman F, and Miruoze J, Calcium absorption in corticoid treated subjects: effects of single oral dose of calcitriol. Horm Metab Res, 1987. 19: p. 335-338.
70. Gallagher JC, Aaron J, Horsman A, Wilkinson R, and Nordin BE, Corticosteroid osteoporosis. Clin Endocrinol Metab, 1973. 2: p. 355.
71. Hahn TJ, Halstead LR, and Baran DT, Effects of short-term glucocorticoid administration on intestinal calcium absorption and circulating vitamin D metabolite concentrations in man. J Clin Endocrinol Metab, 1981. 52: p. 111-115.
72. Kimberg DV, Baerg RD, Gershon E, and Graudusius RT, Effect of cortisone treatment on the active transport of calcium by the small intestine. J Clin Invest, 1971. 50: p. 1309-1321.
73. Favus MJ, Walling MW, and Kimberg DV, Effects of 1,25-dihydroxycholecalciferol on intestinal calcium transport in cortisone-treated rats. J Clin Invest, 1973. 52: p. 1680-1685.
74. Aloia JF, Semla HM, and Yeh JK, Discordant effects of glucocorticoids on active and passive transport of calcium in the rat duodenum. Calcif Tiss Int, 1973. 36: p. 327-331.
75. Lukert BP, Stanbury SW, and Mawer EB, Vitamin D and intestinal transport of calcium: effects of prednisolone. Endocrinology, 1973. 93: p. 718-722.
76. Hahn TJ, Halstead LR, Teitelbaum SL, and Hahn BH, Altered mineral metabolism in glucocorticoid-induced osteopenia: effect of 25-hydroxyvitamin D administration. J Clin Invest, 1979. 64: p.655-665.
77. Seeman E, Kumar R, Hunder GG, Scott M, Heath HIII, and Riggs BL, Production, degradation, and circulating levels of 1,25 - dihydroxyvitamin D in health and in chronic glucocorticoid excess. J Clin Invest, 1982. 69: p. 1302-1309.
78. Feher JJ and Wasserman RH, Intestinal calcium-binding protein and calcium absorption in cortisol treated chicks: effect of Vitamin D_3 and 1,25- dihydroxyvitamin D_3. Endocrinology, 1979. 104: p. 547-551.

79. Kimura S and Rasmussen H, Adrenal glucocorticoids, adenine nucleotide translocation, and mitochondrial calcium accumulation. J Biol Chem, 1977. 252: p. 1217-1225.
80. Wajchenberg BL, Pereira VG, Kieffer J, and Ursic S, Effect of dexamethasone on calcium metabolism and 47 Ca kinetics in normal subjects. Acta Endocrinol, 1969. 61: p. 173-192.
81. Nielsen HK, Thomsen K, Eriksen EF, Charles P, and Mosekilde L, The effects of high dose glucocorticoid administration on serum bone gamma carboxyglutamic acid-containing protein, serum alkaline phosphatase and vitamin D metabolites in normal subjects. Bone Miner, 1988. 4: p. 105-113.
82. Suzuki Y, Ichikawa Y, Saito E, and Homma M, Importance of increased urinary calcium excretion in the development of secondary hyperparathyroidism of patients under glucocorticoid therapy. Metabolism, 1983. 32: p. 151-156.
83. Reid IR and Ibberston HK, Evidence for decreased tubular reabsorption of calcium in glucocorticoid-treated asthmatics. Hormone Research, 1987. 27: p. 200-204.
84. Hodgson SF, Corticosteroid-induced osteoporosis. Endocrinol Metab Clin North Am, 1990. 19: p. 95-111.
85. Tohmon M, Fukase M, Kishihara M, Kadowaki S, and Fujita T, Effect of glucocorticoid administration on intestinal, renal, and cerebellar calbindin-D28K in chicks. J Bone Min Res, 1988. 3: p. 325-331.
86. Findling JW, Adams NS, Lemann J, Gray RW, Thomas CJ, and Tyrrell JB, Vitamin D metabolites and parathyroid hormone in Cushing's syndrome: relationship to calcium and phosphorus homeostasis. J Clin Endocrinol Metab, 1982. 54: p. 1039-1044.
87. Frieberg JM, Kinsella J, and Sacktor B, Glucocorticoids increase the Na+-H+ exchange and decrease the Na + gradient-dependent phosphate-uptake systems in renal brush border membrane vesicles. Proc Natl Acad Sci USA, 1982. 79: p. 4932-4936.
88. Cosman F, Nieves J, Herbert J, Shen V, and Lindsay R, High-dose glucocorticoids in multiple sclerosis patients exert direct effects on the kidney and skeleton. J Bone Miner Res, 1994. 9: p. 1097-1105.
89. Au WY, Cortisol stimulation of parathyroid hormone secretion by rat parathyroid glands in organ culture. Science, 1976. 193: p. 1015-1017.
90. Fucik RF, Kukreja SC, Hargis GK, Bowser EN, Henderson WJ, and Williams GA, Effects of glucocorticoids on the function of the parathyroid glands in man. J Clin Endocrinol Metab, 1975. 40: p. 152-155.
91. Chesney RW, Mazess RB, Hamstra AJ, DeLuca HF, and O'Reagan S, Reduction of serum 1,25-dihydroxyvitamin D_3 in children receiving glucocorticoids. Lancet, 1978. 2: p. 1123-1125.
92. Zerwekh JE, Emkey RD, and Harris ED jr, Low-dose prednisone therapy in rheumatoid arthritis: effect on vitamin D metabolism. Arthritis Rheum, 1984: p. 1050-1052.
93. Gennari C, Imbimbo B, Montagnani M, Bernini M, and Avioli LV, Effects of prednisone and deflazacort on mineral metabolism and parathyroid hormone activity in humans. Calcif Tiss Int, 1984. 36: p. 245.
94. Slovik DM, Neer RM, Ohman JL, Lowell FC, Clark MB, Segre GV, and Potts JT jr, Parathyroid hormone and 25-hydroxyvitamin D levels in glucocorticoid treated patients. Clin Endocrinol, 1980. 12: p. 243.
95. Sakakura M, Takebe K, and Nakagawa S, Inhibition of luteinizing hormone secretion induced by synthetic LRH by long-term treatment with glucocorticoids in human subjects. J Clin Endocrinol

Metab, 1975. 40: p. 774-779.
96. Luton JP, Thieblot P, Valcke JC, Mahoudeau JA, and Bricaire H, Reversible gonadotrophin deficiency in male Cushing's disease. J Clin Endocrinol Metab, 1977. 45: p. 488-495.
97. Crilly RG, Marshall DH, and N. BE, Metabolic effects of corticosteroid therapy in post-menopausal women. J Steroid Biochem, 1979. 11: p. 429-433.
98. Hsueh AJ and Erickson GF, Glucocorticoid inhibition of FSH-induced estrogen production in cultured rat granulosa cells. Steroids, 1978. 32: p. 639-648.
99. Schaison G, Durand F, and Mowszowicz I, Effect of glucocorticoids on plasma testosterone in man. Acta Endocrinol, 1987. 89: p. 126-131.
100. Reid IR, Steroid osteoporosis. Calcif Tiss Int, 1989. 45: p. 63-67.
101. Doerr P and Pirke KM, Cortisol-induced suppression of plasma testosterone in normal adult males. J Clin Endocrinol Metab, 1976. 43: p. 622-629.
102. MacAdams MR, White RH, and Chipps BE, Reduction of serum testosterone levels during chronic glucocorticoid therapy. Ann Intern Med, 1986. 104: p. 648-651.
103. Goulding A and Gold E, Effects of chronic prednisolone treatment on bone resorption and bone composition in intact and ovarectomized rats and ovarectomized rats receiving B-estradiol. Endocrinology, 1988. 122: p. 482-487.
104. Als OS, Gotfredsen A, and Christiansen C, The effect of glucocorticoids on bone mass in rheumatoid arthritis patients: Influence of postmenopausal state. Arthritis Rheum, 1985. 28: p. 369-375.
105. Reid IR, Glucocorticoid osteoporosis- mechanisms and management. Eur J Endocrinol, 1997. 137: p. 209-217.
106. Reid DM, Nicoll JJ, Smith MA, Higgins B, Tothill P, and Nuki G, Corticosteroids and bone mass in Asthma: Comparisons with rheumatoid arthritis and polymyalgia rheumatica. Br Med J [Clin Res], 1986. 293: p. 1463-1466.
107. Reid DM, Kennedy NS, Smith MA, Tothill P, and Nuki G, Total body calcium in rheumatoid arthritis: Effects of disease activity and corticosteroids treatment. Br Med J [Clin Res], 1982. 285: p. 330-332.
108. Rizzato G, Tosi G, Mella C, Zanni D, Sisti S, and Loglisci T, Researching osteoporosis in prednisone treated sarcoid patients. Sarcoidosis, 1987. 4: p. 45-48.
109. Varanos S, Ansell BM, and Reeve J, Vertebral collapse in juvenile chronic arthritis: Its relationship with glucocorticoid therapy. Calcif Tiss Int, 1987. 41: p. 75-78.
110. Seeman E, Melton LJ III, O'Fallon WM, and Riggs BL, Risk factors for spinal osteoporosis in men. Am J Med, 1983. 75: p. 977-983.
111. Leidig-Bruckner G, Eberwein S, Czeczatka D, Dodidou P, et al., Incidence of osteoporotic fractures after liver or heart transplantation during long-term follow-up. J Bone Miner Res, 1997. 12(Suppl. 1): p. 145.
112. Cvetkovic M, Mann GN, Romero DF, Liang WG, Ma Y, and Epstein, The deleterious effects of long-term cyclosporin A, cyclosporin C, and FK506 on bone mineral metabolism *in vivo*. Transplantation, 1994. 57: p. 1231-1237.
113. Afifi AK, Bergman RA, and Harvey JC, Steroid myopathy. Clinical histologic and cytologic observations. Johns Hopkins Med J, 1968. 123: p. 158-173.
114. William RS, Triamcinolone myopathy. Lancet, 1959. 1: p. 887.
115. Harman JB, Muscular waisting and corticosteroids therapy. Lancet, 1959. 1: p. 887.
116. Askari A, Vignos PJ Jr, and Moskowitz RW, Steroid myopathy in connective tissue disease.

Am J Med, 1976. 61: p. 485-492.
117. Lips P, van Ginkel FC, and Netelenbos JC, Lower mobility and markers of bone resorption in the elderly. Bone Min Res, 1990. 9: p. 49-57.
118. Verstraeten A and Dequeker J, Vertebral and peripheral bone mineral content and fracture incidence in postmenopausal patients with rheumatoid arthritis: effect of low dose corticosteroids. Ann Rheum Dis, 1986. 45: p. 852-857.
119. Lems WF, Jahangier ZN, Jacobs JWG, and Bijlsma JWJ, Vertebral fractures in patients with rheumatoid arthritis treated with corticosteroids. Clin Exp Rheum, 1995. 13: p. 293-297.
120. Porayko MK, Weisner RH, Hay JE, Krom RA, Dickson ER, Beaver S, and Schwerman L, Bone disease in liver transplant recipients: incidence, timing and risk factors. Transplant Proc, 1991. 23: p. 1462-1465.
121. Shane E, Rivas MDC, Silverberg SJ, Kim TS, Staron RB, and Bilezekian JP, Osteoporosis after cardiac transplantation. Am J Med, 1993. 94: p. 257-264.
122. Reid I and Heap S, Determinants of vertebral mineral density in patients receiving longterm glucocorticoid therapy. Arch. Intern Med, 1990. 150: p. 2545.
123. LoCascio V, Bonnuci E, Imbimbo B, Ballantini P, and Milani S, Bone loss in response to long-term glucocorticoid therapy. Bone and Mineral, 1990. 8: p. 39-51.
124. Als OS, Christiansen C, and Hellesen C, Prevalence of decreased bone mass in rheumatoid arthritis.Relation to anti-inflammatory treatment. Clin Rheumatol, 1984. 3: p. 201.
125. D'Angelo A, Fabris A, Sartori L, Malvasi L, Travaglia P, Gambari PF, and Todesco S, Mineral metabolism and bone mineral content in rheumatoid arthritis. Effect of corticosteroids. Clin Exp Rheumatol, 1985. 3: p. 143.
126. Deding A, Tougaard L, Jensen MK, and Rodbro P, Bone changes during prednisone treatment. Acta Med Scand, 1977. 202: p. 253.
127. Dykman TR, Gluck OS, Murphy WA, Hahn TJ, and Hahn BH, Evaluation of factors associated with glucocorticoid-induced osteopenia in patients with rheumatic diseases. Arthritis Rheum, 1985. 28: p. 361.
128. Adinoff AD and Hollister JR, Steroid-induced fractures and bone loss in patients with asthma. N Engl J Med, 1983. 309: p. 265-268.
129. Staa van TP, Pharmacoepidemiologic risk evaluation in bone diseases, in Department of pharmacoepidemiology and pharmacotherapy. 1999, Utrecht: Utrecht.
130. Peel NFA, Moore DJ, Barrington NA, Bax DE, and Eastell R, Risk of vertebral fracture and relationship to bone mineral density in steroid treated rheumatoid arthritis. Ann Rheum Dis, 1995. 54: p. 801-806.
131. Delmas PD and G. P, Utility of biochemical markers of bone turnover in osteoporosis, in Osteoporosis, Marcus R, Feldman D, and K. J, Editors. 1996, Academic Press: London. p.1075 1088.
132. Garnero P, Hauser E, Chapuy MC, Marcelli C, Grandjean H, and M. C, Can markers of bone turnover predict hip fractures in elderly women?The epidos study. J.Bone Miner Res, 1995. 10: p. 6.
133. Van Daele PLA, Seibel MJ, Burger H, Grobbee DE, Leeuwen JPTM, Birkenhager JC, and P. HAP, Case-control analysis of bone resorption markers, disability and fracture risk: the Rotterdam study. BMJ, 1996. 312(482-483).
134. Smith CB and Smith DA, Relations between age, mineral density and mechanical properties of human femoral compacta. Acta Orthop Scand, 1976. 47: p. 496-502.

135. Parfitt AM and Duncan H, Metabolic bone disease effecting the spine, in The spine. Vol 2, Rothman RH and Simeone FA, Editors. 1982, WB Saunders: Philadelphia. p. 775-905.
136. Wolinsky I, Simkin A, and Guggenheim K, Effects of fluoride on metabolism and mechanical properties of rat bone. Am J Physiol, 1972. 223: p. 46-50.
137. Rich C and Feist E, The action of fluoride on bone, in Fluoride in Medicine, V. TL, Editor. 1970, Hans Huber: Bern. p. 70-87.
138. Riggs BL, Hodgson SF, and O'Fallon WM, Effects of Fluor treatment on the fracture rate in postmenopausal women with osteoporosis. New Engl J Med, 1990. 322: p. 802-809.
139. Ruegssegger P, Bone density measurement, in Osteoporosis; a guide to diagnosis and treatment, B.a. Dambacher, Editor. 1996, Karger: Basel. p. 103-116.
140. Melton LJ, Kan SH, Frye MA, Wahner HW, O'Fallon WM, and Riggs BL, Epidemiology of vertebral fractures in woman. Am J Epidemiol, 1989. 129: p. 1000-1012.
141. Brinckmann P, Biggerman M, and Hilweg D, Prediction of the compressive strength of human lumbar vertebrae. Spine, 1989. 14: p. 606-610.
142. Cody DD, Goldstein SA, Flynn MJ, and Brown EB, Correlations between vertebral regional bone mineral density and whole bone fracture load. Spine, 1991. 16: p. 146-154.
143. Granhed H, Jonson R, and Hansson T, Mineral content and strength of lumbar vertebrae: a cadaver study. Acta Orthop Scand, 1989. 60: p. 105-109.
144. Hansson T, Roos B, and Nachemson A, The bone mineral content and ultimate compressive strenght of lumbar vertebrae. Spine, 1980. 5: p. 46-55.
145. McBroom RJ, Hayes WC, Edwards WT, Goldberg RP, and White AA III, Prediction of vertebral body compressive fracture using quantitative computed tomography. J Bone Joint Surg, 1985. 67-A: p. 1206-1214.
146. Mosekilde L, Bentzen SM, Ortoft G, and Jorgensen J, The predictive value of quantative computed tomography for vertebral density and ash density. Bone, 1989. 10: p. 465-470.
147. Mazess RB, Errors in measuring trabecular bone by computed tomography due to marrow and bone composition. Calcif Tissue Int, 1983. 35: p. 148-152.
148. Bolotin HH, New perspective on the causal influence of soft tissue composition on DXA-measured *in vivo* bone mineral density . J Bone Miner Res, 1998. 13: p. 1739-1746.
149. Cody DD, McCubbrey DA, Divine GW, Gross GJ, and G. SA, Predictive value of proximal femur bone densitometry in determing local orthogonal material properties. Biomech, 1996. 29: p. 753-761.
150. Hodgskinson R and Currey JD, Effects of structural variation in Young's modulus of non-human cancellous bone. Proc Inst Mech Engrs, 1990. 204: p. 43-52.
151. Odgaard A, Kabel J, van Rietbergen B, Dalstra M, and Huiskes R, Fabric and elastic principal directions of cancellous bone are closely related. J Biomech, 1997. 30: p. 487-495.
152. Buchman SR, Sherick DG, Goulet RW, and Goldstein SA, Use of micro-computed tomography scanning as a new technique for the evaluation of membranous bone. J Craniofac Surg, 1998. 9: p. 48-54.
153. Pols HAP and Lockefeer JHM, Behandeling met bisphosphonaten: Alleen een effect op de botmassa? NTOB, 1997. 1: p. 22.
154. Hanania NA, Chapman KR, and Kesten S, Adverse effects of inhaled corticosteroids. Am JMed, 1995. 98: p. 196-208.
155. Stead RJ and Cooke NJ, Adverse effects of inhaled corticosteroids. Br Med J, 1986.

293: p.1463-1466.
156. Kamada AK, M.R. Szefler SJ, Boushey HA, Chinchilli JM, Drazen JE, Isreal E, Lazarus SC, and Lemanske RF, Issues in the use of inhaled glucocorticoids. Am J Respir Crit Care Med, 1996. 153: p. 1739-1748.
157. Frost HM, The pathomechanics of osteoporosis. Clin Orthop, 1985. 200: p. 198-225.
158. Smith MJ and Hodson ME, Effects of long-term inhaled high-dose beclomethasone dipropionate on adrenal function. Thorax, 1983. 38: p. 676-681.
159. Johansson SA, Andersson KE, Brattsand R, Gruvstad E, and Hedner P, Topical and systemic glucocorticoid potencies of budesonide, beclomethasone dipropionate and prednisolone in man. Eur J Respir, 1982. 63(Suppl 122): p. 74-82.
160. Pedersen S and Fuglsang G, Urine cortisol excretion in children treated with high doses of inhaled corticosteroids. A comparision of budesonide and beclomethasone. Eur Resp J, 1988. 1: p. 433-435.
161. Jennings BH, Andersson KE, and Johansson SA, Assessment of systemic effects of inhaled glucocorticoids: comparison of the effects of inhaled budesonide and oral prednisolone on adrenal function and markers of bone turnover. Eur. J Clin Pharmacol, 1991. 40: p. 77-82.
162. Hodsman AB, Toogood JH, Jennings B, Fraher L J, and Baskerville JC, Differential effects of inhaled budesonide and oral prednisolone on serum osteocalcin. J Clin Endocrinol Metab, 1991. 72: p. 530-540.
163. Pouw EM, Prummel MF, Oosting H, Roos CM, and Endert E, Beclomethasone inhalation decreases serum osteocalcin concentrations. BMJ, 1991. 302: p. 627-628.
164. Packe GE, Douglas JG, McDonald AF, Robins SP, and Reid DM, Bone density in asthmatic patients taking high dose inhaled beclomethasone and intermittent systemic corticosteroids. Thorax, 1992. 47: p. 414-417.
165. Beresford JN, Gallagher JA, Poser JW, and Russel RGG, Production of osteocalcin by human bone cells in vitro. Effects of $1,25(OH)_2D_3$, $24,25(OH)_2D_3$, parathyroid hormone and glucocorticoids. Metab Bone Dis Rel Res, 1984. 5: p. 229-234.
166. Wong J and Black P, Acute adrenal insufficiency associated with high dose inhaled steroids. Br Med J, 1992. 304: p. 1415.
167. Van Heest JA, Bokma JA, van Oudheusden LJ, and Stricker BH, Adrenal cortex suppression attributed to the use of budesonide. Ned Tijdschr. Geneesk, 1994. 138: p. 2204-2207.
168. Nikolaizik WH, Marchant JL, Preece MA, and W. JO, Noctural cortisol secretion in healthy adults before and after inhalation of budesonide. Am J Respir Crit Care Med, 1996. 153: p. 97-101.
169. Toogood JH, Crilly RG, Jones G, Nadeau J, and W. GA, Effect of high dose inhaled budesonide on calcium and phosphate metabolism and the risk of osteoporosis. Am Rev Respir Dis, 1988. 138: p. 57-61.
170. Dekhuizen PNR, Bootsma GP, and Herwaarden CLA van, Klinische effectiviteit en bijwerkingen van inhalatiecorticosteroiden bij asthma. Ned Tijdschr Geneeskd, 1994. 138: p. 1408-1413.
171. Toogood JH, B.J., Jennings B, Lefcoe NM, Johansson SA, Use of spacers to facilitate inhaled corticosteroid treatment of asthma. Am Rev Respir Dis, 1984. 129: p. 723-729.
172. Broide J, Soferman R, Kivity S, Golander A, Dickstein G, Spirer Z, and Weisman Y, Low dose adrenocorticotropin test reveals impaired adrenal function in patient taking inhaled corticosteroids. Endocrinol Metab, 1995. 80: p. 1243-1246.
173. Dickstein G, Arad E, and Shechner C, Low dose ACTH stimulation test. The Endocrinologist,

1997. 7: p. 285-293.
174. Monson JP, Systemic effect of inhaled corticosteroids. Thorax, 1993. 48: p. 955-956.
175. Jennings BH, Andersson KE, and Johansson SA, The assesment of the systemic effects of inhaled glucocorticosteroids. The effects of inhaled budesonide vs oral prednisolone on calcium metabolism. Eur J Clin Pharmacol, 1991. 41(1): p. 11-16.
176. Puolijoki H, Lippo K, Herrala J, Salmi J, and Tala E, Inhaled beclomethasone decreases serum osteocalcin in postmenopausal asthmatic women. Bone, 1992. 13: p. 285-288.
177. Civitelli R, Biochemical markers of bone turnover, in Primer on the metabolic bone diseases and disorders of mineral metabolism. 2nd ed, L. Avioli, Editor. 1993, Raven Press: New York. p. 39-61.
178. Toogood JH, J.B., Hodsman AB, Baskerville J, Fraher LJ, Effects of dose and dosing schedule of inhaled budesonide on bone turnover. J Allergy Clin Immun, 1991. 88: p. 572-580.
179. Ali NJ, Capewell S, and Ward MJ, Bone turnover during high dose inhaled corticosteriod treatment. Thorax, 1991. 46: p. 160-164.
180. Delmas PD, Markers of bone formation and resorption, in Primer on the metabolic bone diseases and disorders of mineral metabolism. 2nd ed, M. Favus, Editor. 1993, Raven Press: New York. p. 108-112.
181. Reid DM, Methods of measurement of bone turnover and clinical evaluation of osteoporosis : relevance to asthma and corticosteroid therapy. Respir Med, 1993. 87(Suppl A): p. 9-14.
182. Bootsma GP, Dekhuizen PNR, Festen J, and van Herwaarden CLA, Effects of inhaled corticosteroids on bone. Neth J Med, 1997. 50: p. 254-260.
183. Teelucksingh S, Padfield PL, Tibi L, Gough KJ, and Holt PR, Inhaled corticosteroids, bone formation and osteocalcin. Lancet, 1991. 338: p. 60-61.
184. Jennings B, The assessment of systemic effects of inhaled glucocorticoids, . 1990: Lund.
185. Kotowicz MA, H.S., Hunder GG, Cedel SL, Mann KG, Riggs BL, Relationship of glucocorticoid dosage to serum bone gla-protein concentration in patients with rheumatologic disorders. Arthrit Rheum, 1990. 33: p. 1487-1491.
186. Oikarinen A, Autio P, Vuori J, Vaananen K, Ristelli L, Kiistala U, and Ristelli J, Systemic glucocorticoid treatment decreases serum concentrations of carboxyterminal propeptide of type I procollagen and aminoterminal propeptide of type III procollagen. Br J Dermatol, 1992. 126: p. 172-178.
187. Kerstjens HAM, Postma DS, van Doormaal JJ, van Zanten AK, Brand PL, Dekhuijzen PN, and Koeter GH, Effects of short-term and long-term treatment with inhaled corticosteroids on bone metabolism. Thorax, 1994. 49: p. 652-656.
188. Sorva R, Tunpeinen M, Juntunen-Backman K, Kanonen SL, and Sorva A, Effects of inhaled budesonide on serum markers of bone metabolism in children with asthma. J Allergy Clin Immunol, 1992. 90: p. 808-815.
189. Leech JA, Hodder RV, Ooi DS, and Gay J, Effects of short-term inhaled budesonide and beclomethasone dipropionate on serum osteocalcin in premenopausal women. Am Rev Respir Dis, 1993. 148: p. 113-115.
190. Laroche M, Porteau L, Caron P, Moulinier L, Didier A, Amigues JM, and Cantagrel Mazieres B, Osteoporotic vertebral fractures in a man under high-dose inhaled glucocorticoid therapy. A case report with a review of the literature. Rev Rhum, 1997. 64: p. 267-270.
191. Puolijoki H, Risteli J, Herrala J, Risteli L, and L. K, Effect of inhaled beclomethasone on serum markers of collagen metabolism in postmenopausal asthmatic women. Respir Med, 1996. 90(6):

p. 339-342.
192. Toogood JH, White FA, Baskerville JC, Fraher LJ, and J. B, Comparision of the antiasthmatic, oropharyngeal, and systemic glucocorticoid effects of budesonide administered through a pressurised aerosol plus spacer or the Turbuhaler dry powder inhaler. J Allergy Clin Immunol, 1997. 99(2): p. 186-193.
193. Riggs BL and Melton LJ III, Involutional osteoporosis. N Engl J Med, 1986. 314: p. 1676-1686.
194. Hahn TJ, Steroid and drug induced osteopenia, in Primer on metabolic bone disease and disorders of mineral metabolism, F. MJ, Editor. 1990, William bird press: Richmond. p. 158-162.
195. Struijs A and M. H, The effects of inhaled glucocorticoids on bone mass and biochemical markers of bone homeostasis: a 1 year study of beclomethasone versus budesonide. Neth J Med, 1997. 50: p. 233-237.
196. Packe GE, Robb O, Robins SP, Reid DM, and Douglas JG, Bone density in asthmatic patients taking inhaled corticosteroids: comparision of budesonide and beclomethasone dipropionate. J Royal Coll Phys London, 1996. 30: p. 128-132.
197. Holliday SM, Faulds D, and Sorkin EM, Inhaled fluticasone propionate. A review of its pharmacodynamic and pharmacokinetic properties, and therapeutic use in asthma. Drugs, 1994. 47: p. 318-331.
198. Bootsma GP, Dekhuijzen PNR, Festen J, Mulder P, and van Herwaarden CLA, Comparison of fluticasone propionate and beclomethasone dipropionate on direct and indirect measurements of bronchial hyperresponsiveness in patients with stable asthma. Thorax, 1995. 50: p. 1044-1050.
199. Bootsma GP, Dekhuizen PNR, Festen J, Mulder PGH, Swinkels LMJW, and van Herwaarden CLA, Fluticasone propionate does not influence bone metabolism, in contrast to beclomethasone dipropionate. Am J Respir Crit Care Med, 1996. 153: p. 924-930.
200. Pauwels RA, Yernault JC, Demedts MG, and G. P, Safety and efficacy of fluticasone and beclomethasone in moderate to severe asthma. Belgian Multicenter Study Group. Am J Respir Crit Care Med, 1998. 157: p. 827-832.
201. Wolff AH, Adelsberg B, Aloia J, and Zitt M, Effect of inhaled corticosteroid on bone density in asthmatic patients: a pilot study. Ann Allergy, 1991. 67: p. 117-121.
202. Ip M, Lam K, Yam L, Kung A, and Ngy M, Decreased bone mineral density in premenopausal asthma patients receiving long term inhaled steroids. Chest, 1994. 105: p. 1722-1727.
203. Herrala J, Puolijoki H, Impivaara O, Liippo K, and Tala E, Bone mineral density in asthmatic woman on high dose inhaled beclomethasone dipropionate. Bone, 1994. 15: p. 621-623.
204. Hanania NA, Chapman KR, Sturtridge WC, Szalai IP, and Kesten S, Dose-related decrease in bone density among asthmatic patients treated with inhaled corticosteroids. J Allergy Clin Immunol, 1995. 96: p. 571-579.
205. Luengo M, del Rio L, Pons F, and P. C, Bone mineral density in asthmatic patients treated with inhaled corticosteroids: a case-control study. Eur Resp J, 1997. 10(9): p. 2110-2113.
206. Pensabeni-Jasper T and Panush RS, Review: Corticosteroid usage: observations at a community hospital. Am J Med Sci, 1996. 311: p. 234-239.
207. Sambrook P, Birminghan J, Kempler S, Kelly P, Eberl S, Pockock N, Yeates M, and Eisman J, Corticosteroid effects on proximal femur bone loss. J Bone Miner Res, 1990. 5: p. 1211-1216.
208. Sambrook P, Birmingham J, Kelly P, Kempler S, Ngnyen T, Pocock N, and Eisman J, Prevention of corticosteroid osteoporosis. Prevention of corticosteroid osteoporosis, 1993. 328: p. 1747-1752.
209. Laan RFJM, van Riel PLCM, van de Putte LBA, van Erning LJThO, van 't Hof MA, and Lemmens.

JAM, Low-dose prednisone induces rapid reversible axial bone loss in patients with rheumatoid arthritis. Ann Intern Med, 1993. 119: p. 963-968.
210. Mulder H and Struijs A, Intermittent cyclical etidronate in the prevention of corticosteroid-induced bone loss. Br J Rheumatol, 1994. 33: p. 348-350.
211. Roux C, Oriente P, Laan R, Huhges RA, et al., Randomized trail of effect of cyclical etidronate in the prevention of corticosteroid-induced bone loss. Clin Endocrinol Metab, 1998. 83: p. 1128-1133.
212. Reid IR, King AR, Alexander CJ, and Ibbertson HK, Prevention of steroid induced osteoporosis with (3-amino-1-hydroxypropylidene)-1, 1-bisphosphonate (APD). Lancet, 1988. 1: p. 143-146.
213. Lukert BP, Johnson BE, and Robinson RG, Estrogen and progesterone replacement therapy reduces glucocorticoid-induced bone loss. J Bone Miner Res, 1992. 7: p. 1063-1069.
214. Montemurro L, Schiraldi G, Fraioli P, Tosi G, Riboldi A, and Rizzato G, Prevention of corticosteroid-induced osteoporosis with salmon calcitonin in sarcoid patients. Calcif Tissue Int, 1991. 49: p. 71-76.
215. Saag KG, Emkey R, Schnitzer TJ, Brown JP, et al., Alendronate for the prevention and treatment of glucocorticoid-induced osteoporosis. N Engl J Med, 1998. 339: p. 292-299.
216. Adachi JD, Bensen WG, Brown J, Hanley D, et al., Intermittent etidronate therapy to prevent corticosteroid-induced osteoporosis. N Eng J Med, 1997. 337: p. 382-387.
217. Reid DM, Cohen S, Pack S, Chines A, and E. D, The effects of risedronate on bone in the prevention and treatent of corticosteroid-induced osteoporosis. Calcif Tiss Int, 1999. 64(suppl 1): p. S119.
218. Kanis JA, Melton LJ, Christiansen C, Johnston CC, and Kahltaev N, The diagnosis of osteoporosis. J Bone Miner Res, 1994. 9: p. 1137-1141.
219. Compston JE, Audran M, Avouac B, Bouvenot G, et al., Recommendations for the registration of agents used in the prevention and treatment of glucocorticoid-induced osteoporosis. Calcif Tissue Int, 1996. 59: p. 323-327.
220. Ruegsegger P, Medici TC, and Anliker M, Corticosteroid-induced bone loss. A longitudinal study of alternate day therapy in patients with bronchial asthma using quantitative computed tomography. Eur J Clin Pharmacol, 1983. 25: p. 615.
221. Gluck OS, Murphy WA, Hahn TJ, and Hahn B, Bone loss in adults receiving alternate day glucocorticoid therapy. A comparision with daily therapy. Arthritis Rheum, 1981. 24: p. 892-898.
222. Sheagren JN, Jowsey J, and Kelly PJ, Effect on bone growth of daily versus alternate day corticosteroid administration: an experimental study. J Lab Clin Med, 1977. 89: p. 120-130.
223. Rizzato G and Montemurro L, Reversibility of exogenous corticosteroid-induced bone loss. Eur Respir J, 1993. 6: p. 116-119.
224. Peat ID, Healy S, Reid DM, and Ralston SH, Steroid induced osteoporosis: an opportunity for prevention? Ann Rheum Dis, 1995. 54: p. 66-68.
225. Hall GM, Spector TD, Griffin AJ, Jawad ASM, Hall ML, and Doyle DV, The effect of rheumatoid arthritis and steroid therapy on bone density in postmenopausal women. Arthritis Rheum, 1993. 36: p. 1510-1516.
226. Devogelaer JP, Huaux JP, and Dufour W, Bone-sparing action of deflazacort versus equipotent doses of prednisone: a double blind study in males with rheumatoid arthritis, in Osteoporosis 1987, Christiansen C, Johansen JS, and Riis BJ , Editors. 1987, Osteopress: Copenhagen. p. 1014.
227. Gray RES, Doherty SM, Galloway J, Coulton L, Broe de M, and Kanis JA, A double-blind

study of deflazacort and prednisone in patients with chronic inflammatory disorders. Arthritis Rheumatism, 1991. 34: p. 287-295.

228. LoCascio V, Ballanti P, Milani S, Bertoldo F, LoCascio C, Zanolin EM, and Bonucci E, A histomorphometric long-term longitudinal study of trabecular bone loss in glucocorticoid treated patients: prednisone versus deflazacort. Calcif Tissue Int, 1998. 62: p. 199-204.
229. Balsan S, Steru D, Bourdeau A, Grimberg R, and Lenoir G, Effect of long-term maintenance therapy with a new glucocorticoid, deflazacort, on mineral metabolism and statural growth. Calcif Tissue Int, 1987. 40: p. 303-309.
230. Imbimbo B, Tuzi T, Porzio F, and Schiavetti L, Clinical equivalence of a new glucocorticoid, deflazacort and prednisone in rheumatoid arthritis and SLE patients. Adv Exp Med Biol, 1984. 171: p. 241-256.
231. Bruno G, Paroli M, Floeridia M, Franceschini PM, and Errigo E, Comparative study of prednisone and deflazacort in the treatment of acute bronchial asthma, in Glucocorticoid effects and their biological consequences: advances in glucocorticoid therapy, Avioli VL, Genarri C, and Imbimbo B, Editors, Exerpta Medica: Amsterdam. p. 79-86.
232. Lund B, Egsmose C, Jorgensen S, and K. MR, Establishment of the relative antiinflammatory potency of deflazacort and prednisone in polymyalgia rheumatica. Calcif Tissue Int, 1987. 41: p. 316-320.
233. DiMunno O, Imbimbo B, Mazzantini M, Milani S, Occhipinti G, and Pasero G, Deflazacort versus methylprednisolone in polymyalgia rheumatica: clinical equivalence and relative antiinflammatory potency of different treatment regimens. J Rheumatol, 1995. 22: p. 1492-1498.
234. Krogsgaard MR, Thamsborg G, and Lund B, Changes in bone mass during low dose corticosteroid treatment in patients with polymyalgia rheumatica - a double blind, prospective comparision between prednisolone and deflazacort. Ann Rheum Dis, 1996. 55: p. 143-146.
235. Weisman MH, Dose equivalency of deflazacort and prednisone in the treatment of steroid dependent rheumatoid arthritis, in In Proceedings of the 4th International Symposium on Osteoporosis, Christiansen C and Riis B, Editors. 1993: Copenhagen. p. 515.
236. Braun JJ, B.-F.D., Rietveld AH, Juttmann JR, Visser TJ, Birkenhager JC, Influence of 1a (OH)D_3 administration on bone and bone mineral metabolism in patients on chronic glucocorticoid treatment; a double blind controlled study. Clin Endocrinol, 1983. 19: p. 265.
237. Dykman TR, Haralson KM, and Gluck OS, Effect of oral 1,25 dihydroxyvitamin D and calcium on glucocorticoid-induced osteopenia in patients with rheumatic diseases. Arthritis Rheum, 1984. 27: p. 1336-1343.
238. Reid IR and Ibbertson HK, Calcium supplements in the prevention of steroid-induced osteoporosis. Am J Clin Nutr, 1986. 44: p. 287-290.
239. Buckley LM, Leib ES, Cartularo KS, Vacek PM, and Cooper SM, Calcium and vitamin D-3 supplementation prevents bone loss in the spinje secondary to low-dose corticosteroids in patients with rheumatoid arthritis -a randomized, double-blind, placebo controlled trail. Ann Int Med, 1996. 125: p. 961-968.
240. Adachi JD, Bensen WG, Bianchi F, Cividino A, et al., Vitamin D and calcium in the prevention of corticosteroid-induced osteoporosis- a 3 year follow-up. J Rheumatol, 1996. 23: p. 995-1000.
241. Koster JC, Hackeng WHL, and Mulder H, Diminished effect of etidronate in vitamin D deficient osteopenic postmenopausal women. Eur J Clin Pharmacol, 1996. 51(2): p. 145-147.
242. Adams JS, Wahl TO, and Lukert BP, Effects of hydrochlororthiazide and dietary sodium

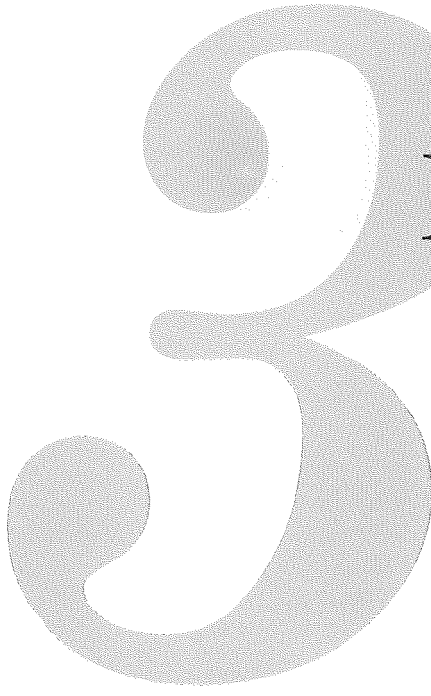
restriction on calcium metabolism in corticosteroid treated patients. Metabolism, 1981. 30: p. 217-221.
243. Yamada H, Long-term effect of 1 alpha-hydroxyvitamin D, calcium and thiazide administration on glucocorticoid-induced osteoporosis. Nippon Naibunpi Gakkai Zasshi, 1989. 65: p. 603-614.
244. Schwartzman MS and Franck WA, Vitamin D toxicity complicating the treatment of senile, postmenopausal, and glucocorticoid -induced osteoporosis. Four case reports and a critical commentary on the use of vitamin D in these disorders. Am J Med, 1987. 82: p. 224-230.
245. Azria M, Endogeneous calcitonin, in The calcitonins: Physiology and pharmacology, A. M, Editor. 1989, Karger: Basel. p. 48.
246. Chesnut CH, Baylink DJ, Gruber HE, Ivey JL, et al., Synthethic salmon calcitonin in post-menopausal osteoporosis: current concepts, in Calcitonin: Das therapeutische potential bei osteoporose, Avioli LV and Dambacher MA, Editors. 1984, Scharrauer: Stuttgart.
247. Locascio VL, Adami S, Avioli LV, Comicacini L, Galvanini G, Genarri C, Imbimbo B, and Scuro LA, Suppressive effect of chronic corticoid treatment on circulating calcitonin in man. Calcif Tissue Int, 1982. 34: p. 309-311.
248. Mulder H, Hackeng WH, vd Lely AJ, and Siersema-Smallegange M, Change of serum calcitonin in patients receiving glucocorticoids: an acute phase study. Neth J Med, 1990. 36: p. 288-290.
249. Luengo M, Picado C, Del Rio L, Guanabens N, Montserrat JM, and Setoain J, Treatment of steroid induced osteopenia with calcitonin in corticosteroid dependent asthma. Am Rev Resp Dis, 1990. 142: p. 104-107.
250. Ringe JD and Welzel D, Salmon calcitonin in the therapy of corticoid-induced osteoporosis. Eur J Clin Pharmacol, 1987. 33: p. 35-39.
251. Adachi JD, Bensen WG, Bell MJ, Bianchi FA, et al., Salmon calcitonin nasal spray in the prevention of corticosteroid-induced osteoporosis. Br J Rheumatol, 1997. 36: p. 255-259.
252. Healy JH, Paget SA, Williams-Russo P, S.R. Szatrowski TP, Spiera H, Mitnick H, Ales K, and Schwartzberg, A randomized controlled trail of salmon calcitonin to prevent bone loss in corticosteroid treated temporal arteritis and polymyalgia rheumatica. Calcif Tissue Int, 1996. 58: p. 73-80.
253. Lyritis GP, Paspati I, Karachalios T, Ioakimidis D, Skarantavos G, and Lyritis PG, Pain relief from nasal salmon calcitonin in osteoporotic vertebral crush fractures. A double blind placebo-controlled clinical study. Acta Orhtop Scand, 1997. 275(suppl): p. 112-114.
254. Greenberg C, Kukreja SC, Bowser EN, Hargis GK, Henderson WJ, and Williams GA, Effects of estradiol and progesterone on calcitonin secretion. Endocrinology, 1986. 118: p. 2594-2598.
255. Hall GM, Daniels M, Doyle D, and Spector TD, Effect of hormone replacement therapy on bone mass in rheumatoid arthritis patients treated with and without steroids. Arthritis Rheumatism, 1994. 37: p. 1499-1505.
256. Grecu EO, Weinshelbaum A, and Simmons R, Effective therapy of glucocorticoid-induced osteoporosis with medroxyprogesterone acetate. Calcif Tissue Int, 1990. 46: p. 294-299.
257. Grecu EO, Simmons R, and Baylik DJ, Effects of medroxyprogesterone acetate on some parameters of calcium metabolism in patients with glucocorticoid-induced osteoporosis. Bone and Mineral, 1990. 13: p. 153-161.
258. Fitzgerald RC, Skingle SJ, and Crisp AJ, Testosterone concentrations in men on chronic glucocorticosteroid therapy. J R Coll Physicians Lond, 1997. 31: p. 168-170.
259. Murphy S, Khaw KT, Cassidy A, and Compston JE, Sex hormones and bone mineral density in

elderly men. Bone Miner, 1993. 20: p. 133-140.
260. Kubler A, Schulz G, Cordes U, Beyer J, and Krause U, The influence of testosterone substitution on bone mineral density in patients with Klinefelter's syndrome. Exp Clin Endocrinol, 1992. 100: p. 129-132.
261. Devogelaer JP, Decooman S, and Dedeuxchaisnes CN, Low bone mass in hypogonadal males-effect of testosterone substitution therapy: a densitometric study. Maturitas, 1992. 15: p. 17-23.
262. Isaia G, M. M, Pecchio F, Sciolla A, Distefano M, and Molinatti GM, Effect of testosterone on bone in hypogonadal males. Maturitas, 1992. 15: p. 47-51.
263. Bagatell CJ and Bremner WJ, Androgens in men- uses and abuses. N Engl J Med, 1996. 334: p. 707-714.
264. Adami S, Fossaluzza V, Rossini M, Bertoldo F, Gatti D, Zamberlan N, and LoCascio V, The prevention of corticosteroid-induced osteoporosis with nandrolone decanoate. Bone Mineral, 1991. 15: p. 72-81.
265. Need AG, Corticosteroids and osteoporosis. Austr NZ J Med, 1987. 17: p. 267-272.
266. Studd JWW, Savvas M, and Johnson M, Correction of corticosteroid-induced osteoporosis by percutaneous hormone implants. Lancet, 1989. 1: p. 339.
267. Sambrook P, Birmingham J, Champion D, Kelly P, Kempler S, Freund J, and Eisman J, Postmenopausal bone loss in rheumatoid arthritis: effects of estrogens and androgens. J Rheumatol, 1992. 19: p. 357-361.
268. Reid IR, Wattie DJ, Evans MC, and S. JP, Testosterone therapy in glucocorticoid-treated man. Arch Intern Med, 1996. 156: p. 1173-1177.
269. Lau KHW, Farley JR, and Baylink DJ, Phosphotyrosyl protein phosphatases. Biochem, 1989. 257: p. 23-36.
270. Fakkeldij TMV, Damen CA, Duursma SA, and Scheven BAA, Comparision of fluoride and vanadate effects on proliferation of human osteoblast-like cells in vitro. Calcif Tiss Int, 1993. 52: p. 226.
271. Lems WF, Jacobs JWG, Bijlsma JWJ, van Veen GJM, Houben HHML, Haanen HCM, Gerrits MI, and van Rijn HJM, Is addition of sodium fluoride to cyclic etidronate beneficial in the treatment of corticosteroid-induced osteoporosis? Ann Rheum Dis, 1997. 56: p. 357-363.
272. Rickers H, Deding A, Christiansen C, and Rodbro P, Mineral loss in cortical and trabecular bone during high dose prednisone treatment. Calcif Tiss Int, 1984. 36: p. 269-273.
273. Meunier PJ, Birancon D, and Chavassieux P , Treatment with fluoride: bone histomorphometric findings, in Osteoporosis 1987, Christiansen C, Johansen JS, and Riis BJ, Editors. 1987, Osteopress: Copenhagen. p. 824-828.
274. Greenwald M, Brandli D, Spector S, Silverman S, and Golde G, Corticosteroid-induced osteoporosis: effect of a treatment with slow release sodium fluoride. Osteoporosis Int, 1992. 2: p. 303-304.
275. Bayley TA, Muller C, Harrison J, Basualdo J, Sturtridge W, and Josse R, The long-term treatment of steroid-osteoporosis with fluoride. J Bone Miner Res, 1990. 5: p. 157-161.
276. Rizzoli R, Chevalley T, Slosman DO, and Bonjour JP, Sodium monofluorophosphate increases vertebral bone mineral density in patients with corticosteroid-induced osteoporosis. Osteoporosis Int, 1995. 5: p. 39-46.
277. Lems WF, Jacobs WG, Bijlsma JW, Croone A, Haanen HC, Houben HH, Gerrits MI, and van Rijn HJ, Effect of sodium fluoride on the prevention of corticosteroid-induced osteoporosis. Osteoporosis Int, 1997. 7: p. 575-582.

278. Kleerekoper M, Peterson EL, Nelson DA, Phillips E, Schork MA, Tilley BC, and Parfitt AM, A randomised trail of sodium fluoride treatment for postmenopausal osteoporosis. Osteoporosis Int, 1991. 1: p. 155-161.
279. Mamelle N, Meunier PJ, Dusan R, Guillaume M, Martin JL, Gaucher A, Prost A, Ziegler G, and Netter P, Risk benefit ratio of sodium fluoride treatment in primary vertebral osteoporosis. Lancet, 188. 2: p. 361-363.
280. Pal CYC, Sakhaee K, Adams-Huet B, Piziak V, Peterson RD, and Poindextere JR, Treatment of postmenopausal osteoporosis with slow release sodium fluoride. Ann Int Med, 1995. 123: p. 401-408.
281. Murray TM and Ste-Marie LG, Prevention and management of osteoporosis: consensus statements from the scietific advisory board of the osteoporosis society of Canada.7.Fluoride therapy for osteoporosis. CMAJ, 1996. 155: p. 949-954.
282. Russell RGG and R. MJ, Bisphosphonates: From laboratory to the clinic and back again. Bone, 1999. 25: p. 97-106.
283. Luckman SP, Hughes DE, Coxon FP, Russel RGG, and Rogers MJ, Nitrogen-containing bisphosphonates inhibit the mevalonate pathway and prevent post-translational prenylation of GTP-binding proteins, including ras. J Bone Miner Res, 1998. 13: p. 581-589.
284. Luckman SP, Coxon FP, Ebetino FH, Russel RGG, and Rogers MJ, Heterocycle-containing bisphosphonates cause apoptosis and inhibit bone resorption by preventing proteinprenylation: Evidence from structure-activity relationship in J774 macrophages. J Bone Miner Res, 1998. 13: p. 1668-1678.
285. Giannini S, D'Angelo A, Malvasi L, Castrignanao R, Pati T, Tronca R, Liberto L, Nobile M, and Crepaldi G, Effects of one-year cyclical treatment with clodronate on postmenopausal bone loss. Bone, 1993. 14: p. 137-141.
286. Reid IR, Wattie DJ, Evans MC, Gamble GD, Stapleton JP, and Cornish J, Continious therapy with pamidronate, a potent bisphosphonate, in postmenopausal osteoporosis. J Clin Endocrinol Metab, 1994. 79: p. 1595-1559.
287. Reginster JY, Lecart MP, Deroisy R, Sarlet N, Denis D, Ethgen D, Collette J, and Franchimont P, Prevention of postmenopausal bone loss by tiludronate. Lancet, 1989. 2: p. 1469-1471.
288. Storm T, Thamsborg G, Steiniche T, Genant HK, and Sorensen OH, Effect of intermittent cyclical etidronate therapy on bone mass and fracture rate in woman with postmenopausal osteoporosis. N Engl J Med, 1990. 322: p. 1265-1271.
289. Storm T, Steiniche T, Thamsborg G, and Melsen F, Changes in bone histomorphometry after long-term treatment with intermittent, cyclic etidronate for postmenopausal osteoporosis. J Bone Miner Res, 1993. 8: p. 199-208.
290. Watts NB, Harris ST, Genant HK, Wasnich RD, et al., Intermittent cyclical etidronate treatment of postmenopausal osteoporosis. N Engl J Med, 1990. 323: p. 73-79.
291. Liberman UA, Weiss SR, Broll J, Minne HW, Quan H, and Bell NH, Effect of oral alendronate on bone mineral density and the incidence of fractures in postmenopausal osteoporosis. N Engl J Med, 1995. 333: p. 1437-1443.
292. Balena R, S. Toolan BC, hea M, Markatos A, et al., The effects of 2 year treatment with the aminobisphosphonate alendronate on bone metabolism, bone histomorphometry, and bone strength in ovariectomized non human primates. J Clin Invest, 1993. 92: p. 2577-2586.
293. Fleisch H, Bisphosphonates in bone disease: From the laboratory to the patients.2nd ed. 1995,

London: Parthenon publishing group. 128.
294. Eggelmeijer F, Papapoulos SE, van Paassen HC, Dijkmans BA, and B. FC, Clinical and biochemical response to single infusion of pamidronate in patients with active rheumatoid arthritis: a double blind placebo controlled study. J Rheumatol, 1994. 21(11): p. 2016-2020.
295. Bijvoet OLM, Frijlink WB, Jie K, van der Linden H, et al., APD in Paget's disease of bone. Role of the mononuclear phagocyte system. Arthritis and Rheumatism, 1980. 23(10): p. 1193-1203.
296. Cummings SR, Black DM, and Vogt TM, For the FIT research group. Changes in BMD substantially underestimate the antifracture effects of alendronate and other antiresorptive drugs. J Bone Min Res, 1996. 11: p. S102.
297. Homik JE, Cranney A, T. Shea B, Tugwell P, Wells G, Adachi JD, and Suarez-Almazor ME, A meta-analysis on the use of bisphosphonates in corticosteroid-induced osteoporosis. J Rheumatol, 1999. 26(5): p. 1148-1157.
298. Adachi JD, Cranney A, Goldsmith CH, Bensen WG, et al., Intermittent cyclic therapy with etidronate in the prevention of corticosteroid-induced bone loss. J Rheumatol, 1994. 21: p. 1922-1926.
299. Worth H, Stammen D, and Keck E, Therapy of steroid-induced bone loss in adult asthmatics with calcium, vitamin D, and a diphosphonate. Am J Respir Crit Care Med, 1994. 150: p. 394-397.
300. Struijs A, Snelder AA, and Mulder H, Cyclical etidronate reverses bone loss of the spine and proximal femur in patients with established corticosteroid-induced osteoporosis. Am J Med, 1995. 99: p. 235-242.
301. Skingle SJ and Crisp AJ, Increased bone density in patients on steroids with etidronate. Lancet, 1994. 344: p. 543-544.
302. Gallacher SJ, Fenner JA, Anderson K, Bryden FM, Banham SW, Logue FC, Cowan RA, and Boyle IT, Intravenous pamidronate in the treatment of osteoporosis associated with corticosteroid dependent lung disease: An open pilot study. Thorax, 1992. 47: p. 932-936.
303. Van Cleemput J, Daenen W, Geusens P, Dequeker P, Van De Werf F, and VanHaecke J, Prevention of bone loss in cardiac transplant recipients. A comparision of bisphosphonates and vitamin D. Transplantation, 1996. 61: p. 1495-1499.
304. Reid IR, Heap SW, King AR, and Ibbertson HK, Two-year follow-up of bisphosphonate (APD) treatment in steroid osteoporosis. Lancet, 1988. 2: p. 1144.
305. Sebaldt RJ, Adachi JD, Bensen WG, Bianchi F, et al., Intermittent cyclic therapy with etidronate prevents corticosteroid-induced bone loss: two years of follow-up. Scand J Rheumatol 1996, 1996. 103(suppl): p. 91-93.
306. Pitt P, Li F, Bloom B, Todd P, Pack S, Hughes G, and Moniz C, A double-blind placebo controlled study to determine the effects of intermittent cyclical etidronate on bone mineral density in patients on long term corticosteroid treatment. Bone, 1997. 20(supll 4): p. 100S.
307. Jenkins EA, Walker-Bone KE, Wood A, McCrae FC, Cooper C, and Cawley MI, The prevention of corticosteroid-induced osteoporosis with intermittent cyclical etidronate. Scand J Rheumatol, 1999. 28(3): p. 152-156.
308. Geusens P, Vanhoof J, Stalmans R, Joly J, Dequeker J, Nijs J, and Raus J, Cyclic etidronate increases bone density in the spine and hip in postmenopausal women on chronic corticosteroid treatment. A double-blind controlled study. Bone, 1997. 20(suppl 4): p. 9S.
309. Lane N, Genant H, and Engleman E, Effect of intermittent cyclic etidronate (ICT) therapy for glucocorticoid-induced osteoporosis in rheumatoid arthritis (RA): interim analysis. Arthritis Rheum, 1993. 36: p. S51.

310. Diamond T, McGuigan L, Barbagallo S, and Bryant C, Cyclic etidronate plus ergocalciferol prevents glucocorticoid-induced bone loss in postmenopausal women. Am J Med, 1995. 98: p. 459-463.
311. Eastell R, Devogelaer JP, Peel NFA, Gill C, Bax DE, Nagant de Deuxchaisnes C, and Russel RGG, A double-blind placebo-controlled study to determine the effects of risedronate on bone loss in glucocorticoid-treated rheumatoid arthritis patients. J Bone Miner Res, 1996. 11: p. 1812.
312. Krieg MA, Thiebaud D, Gillard-Berguer D, Goy JJ, and Burckhardt P, Intermittent intravenous pamidronate prevents the dramatic bone loss after heart transplantation. J Bone Miner Res, 1996. 11.
313. Wolfhagen F, van Buuren H, den Ouden J, Hop W, van Leeuwen J, Schalm S, and Pols HAP, Cyclical etidronate in the prevention of bone loss in corticosteroid-treated biliary cirrhosis. J Hepatol, 1997. 26: p. 325-330.
314. Boutsen Y, Jamart J, Esselinckx W, Stoffel M, and Devogelaer JP, Primary prevention of gluc corticoid-induced osteoporosis with intermittent intravenous pamidronate: a randomized trail. Calcif Tissue Int, 1997. 61: p. 266-271.
315. Gonnelli S, Rottoli P, Cepollara C, Pondrelli C, Cappiello V, Vagliasindi M, and Genarri C, Prevention of corticosteroid-induced osteoporosis with alendronate in sarcoid patients. Calcif Tissue Int, 1997. 61: p. 382-385.



PART

Oral and Inhaled Glucocorticoids

CHAPTER 5

THE PREVALENCE OF PATIENTS AT RISK FOR GLUCOCORTICOID-INDUCED OSTEOPOROSIS IN THE NETHERLANDS

A. Struijs[1], S. A. de Witte[1], H. Mulder[2], H. A. P. Pols[1]
[1]Erasmus Medical Centre, Rotterdam, [2]Site Management Organisation "Good Clinical Practice", Rotterdam

(submitted)

ABSTRACT

Patients receiving long-term prednisone (7.5 mg prednisone daily for ≥ 3 months), thereby at risk for developing steroid induced osteoporosis, were identified by using a specially designed computer program which screened 22 large population based databases from general pharmacists, randomly chosen throughout the Netherlands. Concomitant data about prescription of drugs used for prevention and treatment of osteoporosis were obtained.
The investigated population consisted of 248.169 people, 157.461 subjects with a public insurance and 90.708 subjects with a private insurance. Nine hundred sixty-four subjects (532 women and 432 men, aged 4-98 years) were taking long-term prednisone (mean dose 12.5 mg) for ≥ 3 months. Of these 964 patients, 351 (36%) were taking some form of anti-osteoporotic medication.
The age-specific prevalence of long-term prednisone use, was calculated in the population with a public insurance. An age-related increase of prednisone use was observed from 0.015% at the age of 0-10 up to the highest prevalence (1.4%) in the age group 80-89 (overall prevalence of 0.27%). Our computer program can create an effective signalling system in identifying patients at risk for steroid induced osteoporosis.

KEYWORDS

Steroid induced osteoporosis; glucocorticosteroids; prevalence; anti-osteoporotic therapy; population based.

INTRODUCTION

Systemic glucocorticoids are used in the treatment of a wide variety of sometimes life-threatening diseases. The adverse effects of long-term use of these agents are multiple and often serious, including steroid-induced osteoporosis which contributes to significant morbidity [1,2].

The use of a supra-physiological dose of glucocorticosteroids (i.e. ≥ 7.5 mg prednisone daily), provokes a decline in bone mass, with an accelerated bone loss in the first three to six months of treatment [3-5]. It has been reported that, thirty to thirty-five percent of patients receiving long-term glucocorticoid therapy suffer from one or more vertebral fractures.The overall reported risk of hip and vertebral fractures in these patients increases from 50% up to 100% [6-8]. A steep increase in fracture risk at doses over 7.5 mg prednisone daily, is observed [9, 10].

Therefore, determining the prevalence of patients at highest risk for steroid-induced osteoporosis implies the selection of patients with a daily use of more than 7.5 mg prednisone for at least 3 months. It has been suggested that 75,000 patients are currently being treated with glucocorticosteroids in the Netherlands [11]. However, a more precise estimate of the prevalence of patients using potentially osteoporosis inducing dosages of glucocorticosteroids has not been calculated.

The objective of this study was to determine the prevalence of subjects using the potentially harmful dosage of > 7.5 mg of prednisone and to estimate the use of medication for prevention and treatment of osteoporosis in the Netherlands.

METHODS

Out of 650 pharmacists in the Netherlands, with a specific "Euroned" database system, 25 were randomly selected, covering the whole Dutch geographic, including both urban and rural areas. The pharmacists were first approached by mail and then personally invited to take part in the survey, resulting in the participation of 22 of them. Three resigned by lack of motivation.

A specially designed computer program, extracting patients with long-term use of prednisone subsequently examined the databases. The most commonly prescribed steroid was prednisone, in 98% of the cases. Therefore, the further analysis was restricted to this glucocorticoid. From the selected patients we recorded: age, sex, steroid daily dose, duration of therapy, public or private insurance and co-prescription of anti-osteoporotic drug as used in the prevention and treatment of corticosteroid-induced osteoporosis in the Netherlands. (See table 1.) The legend duration (prescription length) was calculated by dividing the number of filled tablets or capsules by the described daily number. Concommitant medication was defined as any drug of which the legend duration overlapped with the legend duration of the index drug. The index drug was defined as all orally given prednisone, ATC code H02AB07.

Anti-osteoporotic drugs (ATC-code)
• Calcium (A12AA) • Bisphosphonates (M05BA,M05BB) • Vitamin D (A11CC) • HRT (Estradiol, conjugated estrogens and combintions with progestagens) (G03CA,G03DC, G03FA, G03FB)

Table 1

Health care in the Netherlands is divided through a system of public and private insurance schemes. Almost all subjects with a public insurance are registered at the pharmacy, even those not on any medication. Therefore, we were able to determine the exact prevalence, in the year 1998, for the use of long-term prednisone in subjects with a public insurance in the Netherlands. The study was approved by the Medical Ethical Committee.

RESULTS

The total examined population comprised 248.169 subjects of which 157.461 with a public and 90.708 with a private insurance. Out of the total population, 964 patients used ≥ 7.5 mg of prednisone for longer than 3 months, with a mean dose of 12.5 mg per day (fig 1). Age in the patient group ranged from 4-98 (median 69) years, 532 were women (fig 2).
Of this 964 patients 351 (36%) received some form of medication used in prevention or treatment of osteoporosis. Of the prednisone treated patients an avarage of 36% used any

Fig 1- Dose Prednisone

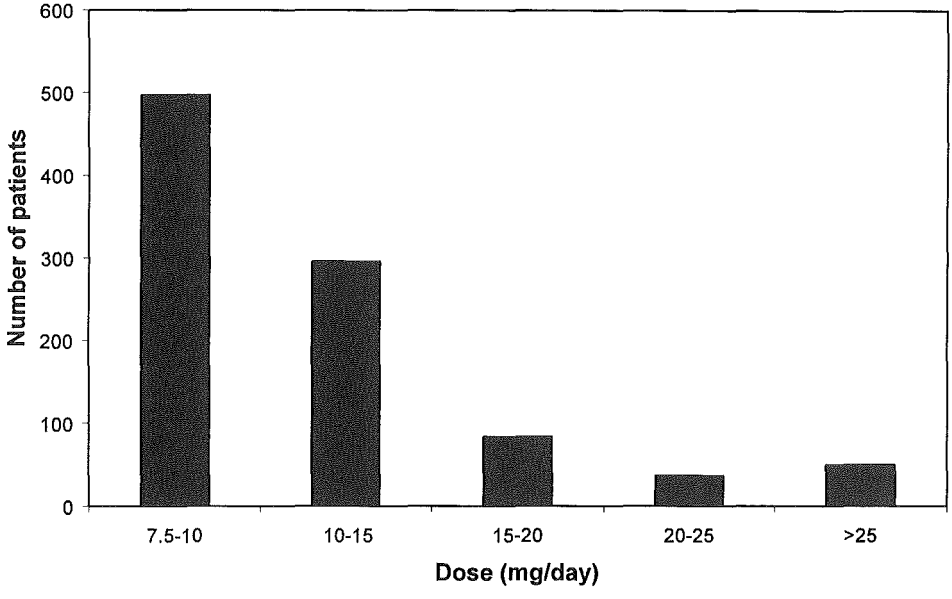


Fig 2- Age distribution of men and women, taking more than 7.5 mg prednisone for at least 3 months

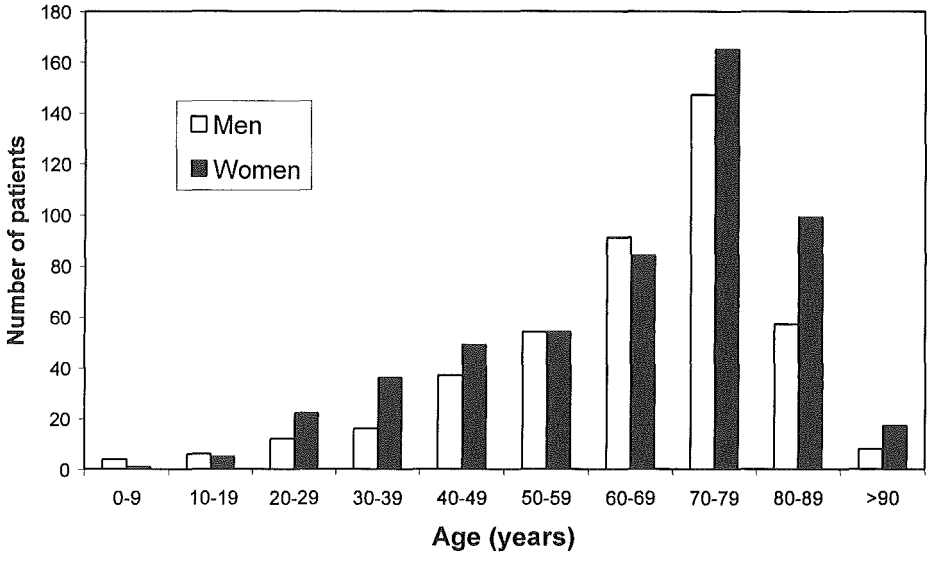
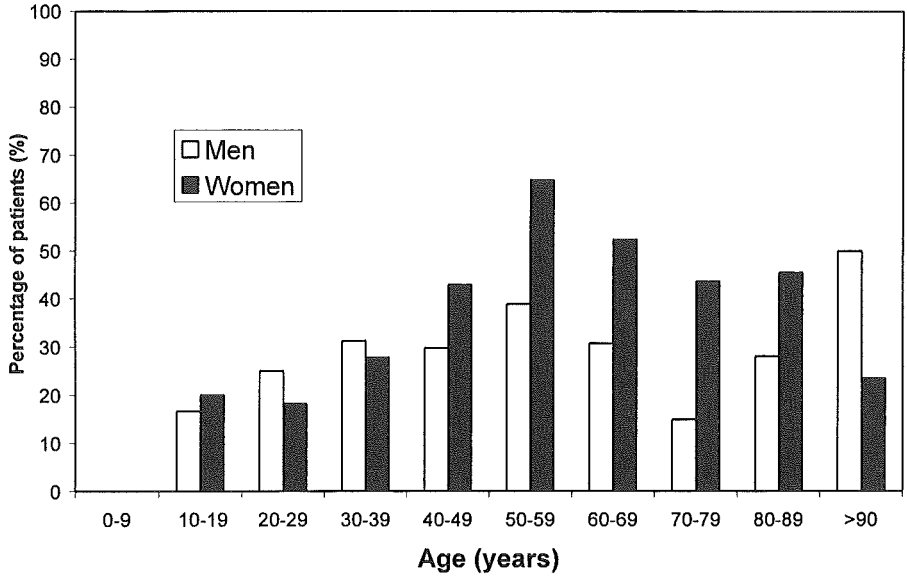


Fig 3- Percentage of patients using some form of anti-osteoporotic medication



form of anti-osteoporotic treatment. In those below 40 years of age this percentage declined to around 25% (Fig. 3) The co-medication comprised calcium (26%), bisphosphonates (40%), vitamin D (6%), vitamin D with calcium (16%), vitamin D with cyclic calcium and bisphosphonates (4%) and hormone replacement therapy (8%) (Fig 4).

From the overall population, we separated the group of 157.461 people with a public insurance of whom we had the exact data about users and non-users of medication. This group had an age-sex distribution similar to that of the general population in the Netherlands as compared to population figures of 1998 provided by the Dutch Central Bureau of Statistics. In this population the overall prevalence of prednisone use $\geq$ 7.5 mg for more than 3 months was 0.27%. A steep increase was observed in people over 55 years of age, with a mean prevalence of 0.77%, with a subsequent decline in the oldest group. In those in the age group 80-89 over 80 years old the prevalence was 1.4% (fig 5).

DISCUSSION

Very few studies have estimated the prevalence of long-term use of systemic corticosteroids in the general population [12,13]. In 1996, Walsh et al. reported that, irrespective of dose,

0.5% of the Nottinghamshire population in the United Kingdom was taking long-term glucocorticosteroids with a median dose of 6.8 mg daily.

We are the first to determine the prevalence of patients at risk for steroid-induced osteoporosis using a daily dose of at least 7.5 mg prednisone for longer than 3 months. We found a prevalence of 0.27% in the total population and 0.77% of people over 55 years. The latter comprises women who are already at risk for osteoporosis given their postmenopausal status [14].

The steep increase in prevalence in the elderly is impressive, reaching a prevalence of 1.4% in patients in their eighties. In people over 90 years of age there was a gradual decline in prevalence of glucocorticoid use, which is probably due to a lower incidence of diseases for which prednisone is prescribed. Another explanation is that the very elderly reached this old age due to their healthy constitution and therefore not using glucocorticoids.

Unlike Walsh et al [12] we observed no statistically significant difference in the age specific prevalence between men and women (fig 3). Probably because we focussed on the higher dosages of prednisone taken for at least three months. Also the social class of the selected population could be a factor. Due to the different registration of private and public insured subjects in the pharmacists' database, we were only able to calculate the prevalence in the public insured subjects which represents a group with a lower income. Nevertheless, the age distribution in the population examined was similar to the Dutch population at large. If we

Fig 4- Co-prescription

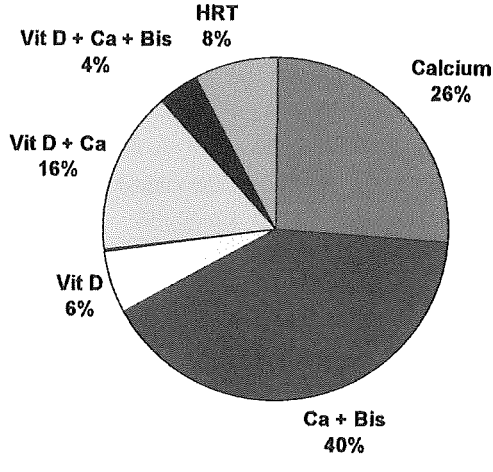
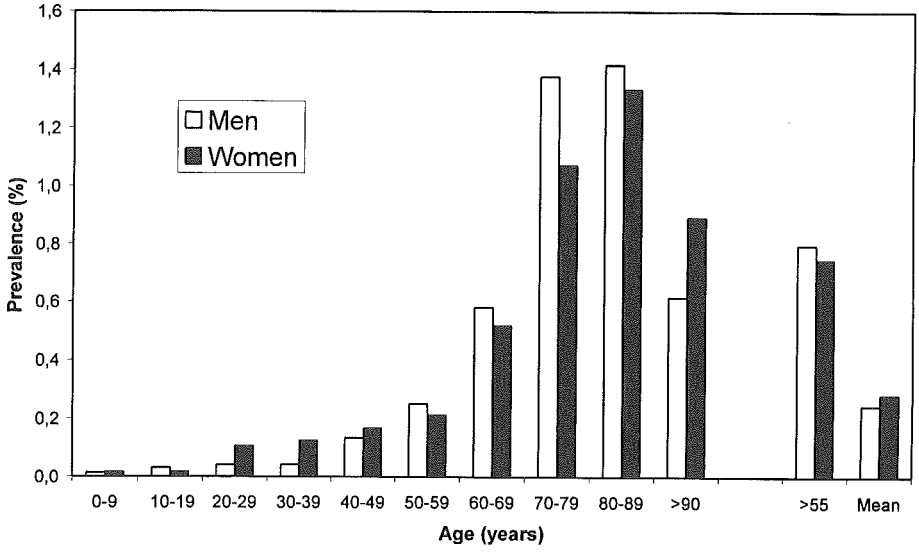


Fig 5- Prevalence of use of more than 7.5 mg prednisolone for at least 3 months



extrapolate our prevalence figures to the total Dutch population (15.6 million in 1998), at least 40.000 people are at high risk for steroid induced osteoporosis. This figure corresponds with estimations of 75.000 people using glucocortico-steroids overall [11]. Our determination of prevalence is certainly an underestimation since our study was limited to the outpatient population and missed patients using repeated short-term prednisone courses. Furthermore, we excluded other glucocorticoids, such as dexamethasone, since their use in the general population is very low [11,12].

Prevention therapy for steroid induced osteoporosis is only given in a minority of patients receiving glucocorticosteroids [15]. This is probably due to the low awareness of the long-term adverse effects of glucocorticoids on the skeleton, even when it is used in a relatively high dose [16][17].

In our study, 36% of the patients at risk received a prescription of any form of an anti-osteoporotic prescription. Adult patients younger than 40 years used even less prophylactics, only around 25% (fig 4). Yet this percentage is higher than found by Walsh et al in the UK, who reported use of anti-osteoporotic medication in only 14% of patients using oral glucocorticosteroids. We probably overestimated the use of co-prescription, since estrogen and progestagen or combined use will be also prescribed for other reasons than osteoporosis. Even

without hormone prescription the co-prescription figure is still higher than the 14% determined by Walsh et al probably due to recent growth in awareness of the adverse effects of glucocorticoids on bone and implementation of guidelines, such as proposed by The American College of Rheumatology [18] and recently the United Kingdom Consensus Group [19,20].

From this study, it appeared that a substantial number of subjects are at risk for steroid induced osteoporosis, while only in a minority of cases preventive measurements have been taken. Most countries focussed on case finding concerning glucocorticoid-induced osteoporosis. Our method of extracting patients at risk with a computer program could provide a tool that is easy to use in the identification of long-term glucocorticoid users in general practice. For example, extracting patients at risk from a database of 10,000 takes about 15 minutes, and provides detailed information about dose and duration of steroid therapy, with personal data available only to the pharmacist and general practitioner. Provided that pharmacists have automated records of prescription, its use in daily practice may create a signalling system, ultimately resulting in a better identification of patients at risk for glucocorticoid-induced osteoporosis.

ACKNOWLEDGEMENTS

We thank Prof. A.G.H. Smals for helpful comments on the manuscript and L.P. Rutten for his creativity in computer programming.

REFERENCES

1. Khosl S, Lufkin EG, Hodgson SF, Fitzpatrick LA, and Melton LJ, Epidemiological and clinical features of osteoporosis in young adults. Bone, 1994. 15: p. 551-555.
2. Lems WF, Jahangier ZN, Jacobs JWG, and B. JWJ, Vertebral fractures in patients with rheumatoid arthritis treated with corticosteroids. Clin Exp Rheum, 1995. 13: p. 293-297.
3. Sambrook P, Birmingham J, Kelly P, Kempler S, Ngyen T, Pocock N, and Eisman J, Prevention of steroid osteoporosis. A comparison of calcium, calcitriol, an calcitonin. N Engl J Med, 1993. 328: p. 1747-1752.
4. Laan RFJM, van Riel PLCM, van de Putte LBA, van Erning LJThO, van 't Hof MA, and Lemmens JAM, Low-dose prednisone induces rapid reversible axial bone loss in patiens with rheumatoid arthritis. Ann Intern Med, 1993. 119: p. 963-968.
5. Mulder H and Struijs A, Intermittent cyclical etidronate in the prevention of corticosteroid-induced bone loss. Br J Rheumatol, 1994. 33: p. 348-350.
6. Meunier PJ, Is steroid-induced osteoporosis preventable? N Engl J Med, 1993. 328: p. 1781-1782.
7. Cooper C, Coupland C, and Mitchell M, Rheumatoid arthritis, corticosteroid therapy and hip fracture. Ann Rheum Dis, 1995. 54: p. 49-52.
8. Hooyman JR, Melton LJ, Nelson AM, O'Fallon WM, and Riggs BL, Fractures after rheumatoid arthritis. A population-based study. Arthritis Rheum, 1984. 27: p. 1353-1561.
9. van Staa TP, Pharmacoepidemiologic risk evaluation in diseases, in Thesis. 1999, Utrecht University, The Netherlands.
10. van Staa TP, Cooper C, Abenheim L, Begaud B, and Leufkens H, Individual susceptibility to steroid-induced fractures. Calc Tissue Int, 1999. 64: p. S38.
11. Heerdink ER, Clustering of drug use in the elderly., in Thesis. 1995, Utrecht University, The Netherlands.
12. Walsh LJ, Wong CA, Pringle M, and Tattersfield AE, Use of oral corticosteroids in the community: a cross sectional study. BMJ, 1996. 313: p. 344-346.
13. Thompson P, Bhalla A, Palferman T, Smith J, Woodland J, Tobias J, and Stone M, Corticosteroid use in primary care. Calc Tissue Int, 1999. 64: p. S96.
14. Adachi JD, Bensen WG, Brown J, Hantley D, et al., Intermittent etidronate therapy to prevent corticosteroid-induced osteoporosis. N Engl J Med, 1997. 337: p. 382-387.
15. Peat ID, Healy S, Reid DM, and Ralston SH, Prevention of steroid induced osteoporosis. An opportunity for prevention? Ann Rheum Dis, 1995. 54: p. 66-68.
16. Hudson JQ, Small RE, and Bukley L, Perception of pharmacists about adverse effects of corticosteroid therapy: focus on osteoporosis. J Am Pharm Assoc, 1998. 38(6): p. 710-716.

17. Papapoulos SE, Glucocorticoid-induced osteoporosis, in Osteoporosis, Papapoulos SE, et al., Editors. 1996, Elsevier: Amsterdam. p. 359-367.
18. American College of Rheumatology Task Force on Osteoporosis Guidelines. Recommendations for the prevention and treatment of glucocorticoid-induced osteoporosis. Arthritis Rheum, 1996. 39: p. 1791-1801.
19. Eastell R, Reid DM, Compston J, Cooper C, et al., An UK Consensus Group on management of glucocorticoid-induced osteoporosis: an update. J Int Med, 1998. 244: p. 271-292.
20. Lips P, Prevention of corticosteroid-induced osteoporosis. BMJ, 1999. 318: p. 1366-1367.

CHAPTER 6

THE EFFECTS OF INHALED GLUCOCORTICOIDS ON BONE MASS AND BIOCHEMICAL MARKERS OF BONE HOMEOSTASIS

- A ONE YEAR STUDY OF BECLOMETHASONE VERSUS BUDESONIDE -

A. Struijs MD and H. Mulder MD, PhD

(Published as "The effects of inhaled glucocorticoids on bone mass and biochemical markers of bone homeostasis: a 1 year study of beclomethasone versus budesonide". Neth J Med 1997;50:233-237)

SUMMARY

Bone mass and biochemical bone markers were prospectively studied in 33 patients with chronic obstructive pulmonary disease treated during one year with inhaled beclomethasone 200 µg/Q.I.D.(Group A; 8 men and 4 women), inhaled budesonide 200 µg/Q.I.D.(Group B; 6 men and 5 women), or not requiring steroids (Group C; 6 men and 4 women). Both inhaled corticosteroids decreased serum concentrations of the osteoblastic markers osteocalcin and carboxy terminal propeptide of type I collagen (PICP). The osteoclastic marker cross-linked carboxyterminal telopeptide of type I collagen (ICTP) increased significantly more in patients on beclomethasone than in those on budesonide. The decrease in bone mineral density (BMD) was more pronounced in patients treated with beclomethasone (1.1% in the spine; 1.7% in the hip; $p < 0.05$) compared to those treated with budesonide (0.6% in both spine and hip) or in the control group. Inhaled corticosteroids affect biochemical bone markers and bone mineral density, but there is different effect for the two corticosteroids evaluated in the present study.

KEY WORDS

Beclomethasone; bone markers; BMD; budesonide; osteoporosis; steroids.

INTRODUCTION

High doses of glucocorticoids constitute effective treatment of bronchial asthma and chronic obstructive pulmonary disease (COPD). Unfortunately they decrease intestinal absorption of calcium and increase calcium excretion in urine, both in animals and in man [1-7]. This negative calcium balance leads to secondary hyperparathyroidism with increased bone resorption and decreased bone formation [8-10].
Previous studies in asthmatic patients simultaneously treated with oral prednisolone and inhaled glucocorticosteroids, demonstrated a marked decrease in bone mass and changes in several parameters of bone homeostasis [11-13].
We measured the effects of inhaled beclomethasone and budesonide on bone mass and several biochemical parameters of bone homeostasis in patients with chronic obstructive pulmonary disease (COPD).

PATIENTS AND METHODS

Between February 1st, 1991 and January 31st, 1992, 39 patients with COPD living in the Rotterdam area, and not previously treated with glucocorticosteroids were recruited. Patients were excluded if they had any disease affecting bone or calcium metabolism, or were taking medication such as vitamin D, estrogens, calcitonin, bisphosphonates, or fluoride. A complete history was taken and physical examination performed in each patient before the initiation of therapy; laboratory evaluations included serum chemistry and urinary analysis. If a patient dropped out of the study, the scheduled protocol will be followed (intention to treat). The procedures followed were in accord with guidelines of the Helsinki declaration for physician-initiated studies.
After giving their informed consent, patients requiring glucocorticoids were randomised to beclomethasone aerosol 200 μg four times daily (Group A) or budesonide 200 μg four times daily (Group B); patients not requiring glucocorticoids constituted Group C. Patients were considered as drop-out if they needed oral glucocorticoids during the study, if measurements of BMD or of blood chemistry were not performed according to protocol, or if patients in group C were treated with either oral or inhaled glucocorticoids

during follow-up.
Vertebral (L1-L4) and hip bone mineral density (BMD) were measured by dual energy X-ray absorptiometry (HOLOGIC QDR 1000) by a technician blinded to treatment assignment, at baseline and after 6 and 12 months of treatment.
Serum calcium and alkaline phosphatase were measured with an Autoanalyser. Carboxy-terminal propeptide of type I collagen (PICP) and cross-linked carboxy terminal telopeptide of type I collagen (ICTP) were determined by radioimmunoassay (Orion Farmos, Turku, Finland), with an intra- and interassay CV of respectively 4.7% and 5.3% for PICP and 6.2% and 7.9% for ICTP. Osteocalcin was determined by radioimmunoassay (Inestar Corporation; Stillwater MN, USA; intra- and interassay CV were respectively of 3.8% and 4.3%).

STATISTICS

Data shown as mean ± SEM; one-sample t-tests for between groups changes (Minitab 5.1, Minitab Inc, State College of Pennsylvania) were used, confidence interval 95%, $p < 0.05$ is considered statistical significant.

RESULTS

Initially 39 patients during the study period were included. One patient from Group A and one from Group B were treated with oral glucocorticoids and one patient in Group C with inhaled glucocorticoids; one patient in Group B and two in Group C were excluded because of incomplete laboratory analyses and BMD determinations. The remaining 33 patients completed the study. Beclomethasone and budesonide were well tolerated; no discontinuation of study drugs because of side effects was observed. The efficacy of beclomethasone and budesonide was the same in both treatment groups. Group A consisted of 8 men and 4 women (1 postmenopausal) between 36 and 75 years old (mean age: 46 years) and received beclomethasone. Group B consisted of 6 men and 5 women (1 postmenopausal) between 34 and 79 years old (mean age: 49 years) and received budesonide. Group C included 6 men and 4 women (2 postmenopausal), between 28 and 81 years old (mean age: 50 years), and received no glucocorticoids. Inhaled corticosteroids were prescribed mainly to patients with asthma with an extrinsic component. The patients were able to perform all daily activities without help from others and the mobility in the differ-

ent groups was about the same.
Mean serum concentrations of calcium and of bone markers are listed in table 1. Osteoblastic markers (alkaline phosphatase, osteocalcin, and PICP) decreased in groups A and B and were unchanged in Group C. The osteoclastic marker ICTP increased only in Group A.
Changes in biochemical bone markers after 12 months of treatment are given as percentages in table 2. All measurements were unchanged in Group C. Alkaline phosphatase, osteocalcin, and PICP decreased similarly and significantly in both groups on inhaled glucocorticosteroids. ICTP increased only in patients on beclomethasone. BMD data are listed in table 3. Mean BMD slightly decreased in all groups; no change was statistically significant. A statistically significant percentual decrease in lumbar spine and hip BMD was observed only in patients on beclomethasone (fig.1).

DISCUSSION

Osteoporosis is a well-known and serious complication of prolonged oral treatment with glucocorticoids, but only limited data are available for patients inhaling these drugs [11-13]. Cross-sectional studies on bone density lack properly matched controls, and include confounding variables such as previous courses of oral glucocorticoids. A decrease in osteoblastic markers (alkaline phosphatase and osteocalcin) and an increase in the osteoclastic marker hydroxyproline have been reported [12]. This suggested, depressed bone formation associated with increased bone resorption.
Previous studies [11-13] on the effects of inhaled glucocorticoids on bone mass and biochemical markers of bone and calcium homeostasis have documented an 8% decrease in bone mass in asthmatic patients simultaneously treated with oral prednisolone [11], and a decrease in the osteoblastic marker serum osteocalcin after 8 days of treatment with inhaled glucocorticosteroids in 8 healthy volunteers [12]. Inhaled budesonide and beclomethasone decreased osteoblastic function and increased bone resorption, with a significant decrease in serum alkaline phosphatase and an increase in the urinary hydroxyproline/creatinine ratio [13]. We also found a decrease in bone formation associated with an increase in bone resorption. The decrease in osteocalcin concentrations seems to be a dose-dependent effect of inhaled glucocorticoids [14]. The percentual decrease in the osteoblastic marker PICP was more pronounced than the decrease in alkaline phosphatase or in osteocalcin; as PICP is formed in a 1:1 molar ratio to collagen, this is a more direct measure of on-going bone formation [14]. The more pronounced decrease observed in patients on beclomethasone

TABLE 1

Serum Levels of Calcium, Alkaline Phosphatase, Osteocalcin, PICP and ICTP Before and During Glucocorticosteroid Inhalation Therapy (mean ± SEM)

		Before	3 Months	6 Months	9 Months	12 Months
Serum Ca (mmol/l)	Beclomethasone	2.30 ± 0.02	2.30 ± 0.03	2.30 ± 0.02	2.31 ± 0.02	2.30 ± 0.02
	Budesonide	2.31 ± 0.02	2.30 ± 0.02	2.30 ± 0.02	2.32 ± 0.02	2.30 ± 0.02
	Control	2.28 ± 0.03	2.29 ± 0.02	2.31 ± 0.03	2.32 ± 0.02	2.31 ± 0.02
Alkaline phosphatase (U/l)	Beclomethasone	64 ± 4	57 ± 3	55 ± 4*	52 ± 5*	54 ± 4*
	Budesonide	64 ± 4	56 ± 3	63 ± 3	52 ± 3*	53 ± 3*
	Control	62 ± 5	65 ± 4	65 ± 4	64 ± 4	64 ± 4
Osteocalcin (µg/l)	Beclomethasone	2.85 ± 0.5	2.63 ± 0.4*	2.61 ± 0.4*	2.52 ± 0.4†	2.49 ± 0.5*
	Budesonide	2.79 ± 0.6	2.65 ± 0.5	2.51 ± 0.5*	2.45 ± 0.5†	2.50 ± 0.5*
	Control	2.76 ± 0.5	2.77 ± 0.4	2.74 ± 0.5	2.75 ± 0.4	2.78 ± 0.6
PICP (µg/l)	Beclomethasone	214 ± 14	171 ± 12*	171 ± 14*	168 ± 11†	148 ± 12†
	Budesonide	223 ± 10	188 ± 11*	185 ± 9*	164 ± 10*	161 ± 9*
	Control	204 ± 12	206 ± 10	200 ± 10	205 ± 11	208 ± 10
ICTP (µg/l)	Beclomethasone	3.0 ± 0.3	3.2 ± 0.3	3.2 ± 0.2	3.6 ± 0.2*	3.4 ± 0.3*
	Budesonide	2.9 ± 0.2	2.9 ± 0.3	3.1 ± 0.3	3.2 ± 0.4	3.0 ± 0.3
	Control	2.8 ± 0.2	2.9 ± 0.3	2.9 ± 0.2	3.0 ± 0.3	2.9 ± 0.2

Significance compared to the pre-treatment value: * P <0.05, † P <0.01.

TABLE 2

Percentage of Change (mean ± SEM) (12 Months) Compared to the Pre-treatment Value of Bone Markers in Patients Using Inhaled Glucocorticoids

	Alk. Phosphatase (U/l)	Osteocalcin (µg/l)	PICP (µg/l)	ICTP (µg/l)
Group A	-15.6 ± 1.0*	-12.6 ± 1.9*	-30.9 ± 3.7†	+13.3 ± 1.2‡
Group B	-17.2 ± 1.3*	-10.4 ± 1.8*	-28.7 ± 4.0†	+3.4 ± 1.9§
Group C	+3.2 ± 0.4	+0.7 ± 0.4	+2.0 ± 1.5	+3.6 ± 1.0

A = inhaled beclomethasone; B = inhaled budesonide; C = control group.
* P <0.05 compared to control group.
† P <0.01 compared to control group.
‡ P <0.05 compared to group B and control group.
§ P <0.05 group B compared to group A.

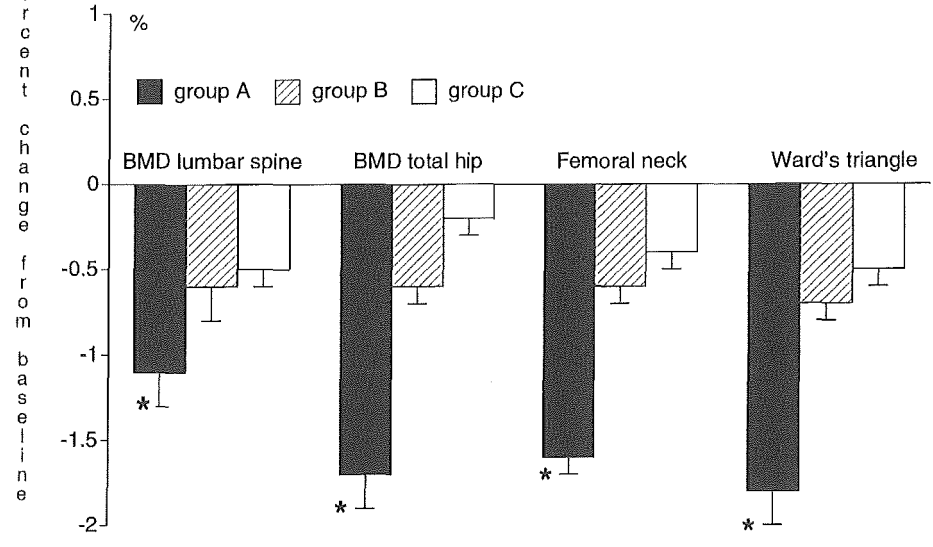
TABLE 3

BMD Values of the Lumbar Spine (mean ± SEM) and Total Hip Before Treatment and During Treatment in COPD Patients

	Before	6 Months	12 Months
BMD lumbar spine (g Ca/cm²)			
Group A	1.101±0.019	1.092±0.020	1.089±0.014
Group B	1.096±0.018	1.091±0.016	1.090±0.018
Group C	1.085±0.021	1.083±0.020	1.080±0.020
BMD total hip (g Ca/cm²)			
Group A	0.884±0.015	0.874±0.018	0.869±0.014
Group B	0.875±0.018	0.872±0.021	0.870±0.022
Group C	0.869±0.020	0.873±0.018	0.867±0.016

A = inhaled beclomethasone; B = inhaled budesonide; C = control group.

Fig. 1 **Mean (± S.E.M.) percentage of change in spine and hip BMD (12 months vs baseline) in patients using inhaled glucocorticosteroids (A = inhaled beclomethasone; B = inhaled budesonide; C = control group)**



* $p<0.05$. Percent change from baseline after five days.

suggests a stronger inhibition of bone formation compared to budesonide. However the number of patients are too small to draw straightforward conclusions. Serum osteocalcin also decreased more on beclomethasone than on budesonide [13]. The osteoclastic marker ICTP increased on beclomethasone, but not on budesonide.

The impact on bone mass of these different effects on osteoblast and osteoclast of these drugs was reflected in the significant percentual decrease in BMD of the lumbar spine and of the hip in patients on beclomethasone, not in those on budesonide. The decrease in BMD was similar in the femoral neck area, in Ward's triangle, and in the total hip bone, but less than the 8% decrease reported by Reid [10], whose patients were also on oral glucocorticoids. Budesonide seems to affect bone mass less than beclomethasone, probably because of its different bioactivity and four times faster degradation into inactive metabolites [15-20]. Although the number of patients are too small to draw straight-

forward conclusions. Our observations warrant a large prospective study with a long follow-up period to evaluate the influence of inhaled glucocorticoids on bone formation and resorption.

ACKNOWLEDGEMENTS

The authors wish to thank the clinical trail organisation Good Clinical Practice, Bergweg 66, 3036 BC, Rotterdam, for their technical assistance and Frans Hagemeyer for helping preparing the manuscript.

REFERENCES

1. Miravet L, Hioco D, Debeyre N, Dryll A, Ryckewaert A, de Seze S. La cinétique du calcium au cours de l'ostéoporose cortisonique. Effect d'un anabolisant de synthése. Sem Hop Paris (suppl.) 1966;42:60-64
2. Wajchenberg BL, Pereira VG, Kieffer J, Ursic S. Effect of dexamethasone on calcium metabolism and Ca kinetics in normal subjects. Acta Endocrinol (Kbh) 1969;61:173-192
3. Gallagher JC, Aaron J, Horsman A, Wilkinson R, Nordin BEC. Corticosteroid osteoporosis. Clin Endocrinol Metab 1973;2:355-368
4. Teotia M, Teotia SPS, Raman T, Sharma NL, Singh RK. Intestinal absorption of calcium and phosphorus; experiments on interaction between vitamin D and adrenocortical hormone. Indian J Pediatr 1975;42:1-9
5. Lukert BP, Adams JS. Ca lcium and phosphorus homeostasis in man; effect of corticosteroids. Arch Intern Med 1976;136:1249-1253
6. Klein RG, Arnaud SB, Gallagher JC, DeLuca HF, Riggs BL) Intestinal calcium absorption in exogenous hypercortisolism; role of 25-hydroxy-vitamin D and corticosteroid dose. J. Clin Invest 1977;60:253-259
7. Hahn TJ, Halstead LR, Teitelbaum SL, Hahn BH. Altered mineral metabolism in glucocorticoid-induced osteopenia; effect of 25-hydroxy-vitamin D administration. J Clin Invest 1979;64:655-665
8. Frast HM, Villaneuva AR. Human osteoblastic activity. III The effect of cortisone on lamellar osteoblastic activity. Henry Ford Hosp Med Bull 1961;9:97-99
9. Jowsey J, Rigs BL Bone formation in hypercortisolism. Acta Endocrinol (Kbh) 1970;63: 21-28
10. Reid DM, Nicol JJ, Smith MA, Higgens B, Turchill P, Noki G. Corticosteroids and bone mass in asthma; comparisons with rheumatoid arthritis and polymyalgia rheumatica.BMJ.1986;293: 1436-1466
11. Bradley BWD, Ansell BM Fractures in Still's disease. Ann Rheum Dis 1960;19:135-142
12. Pouw EM, Prummel MF, Oosting H, Roos CM, Endert E. Beclomethasone inhalation decreases serum osteocalcin concentrations. BMJ 1991;302:627-628
13. Morrison D, Ale NJ, Routlodge PA, Capwell S. Bone turnover in patients with chronic obstructive airways disease during short course prednisone therapy. Thorax 1990;45:791
14. Leech JA, Hodder RV, Ooi DS, Gay J. Effects of short term inhaled budesonide and beclomethasone dipropionate on serum osteocalcin in premenopausal women. Am.Rev.Respir Dis 1993;148:113-115
15. Ryrfeldt A, Andersson P, Edsbacker S. Pharmokinetics and metabolism of budesonide, a selective glucocorticoid. Eur J Respir Dis Suppl 1982;122:86-95

16. Ryrfeldt A, Edsbacker S, Pauwels R. Kinetics of epimeric glucocorticoid budesonide. Clin Pharmacol Ther 1984;35(4):525-530
17. Clissold SP, Heel RC. Budesonide, a preliminary review of its pharmacodynamic properties and therapeutic eficacy in asthma and rhinitis. Drugs 1992;44:375-407
18. Martin LE, Harrison C, Tanner RJ. Metabolism of beclomethasone dipropionate by animals and man. Postgrad Med J 1975;51(suppl 4):11-20
19. Martin LE, Tanner RJ, Clark TJ. Absorption and metabolism of orally administered beclomethasone dipropionate. Clin Pharmacol Ther 1974;15:267-275
20. Small P, Black M, Frenkiel S Beclomethasone dipropionate in the management of rhinitis. A review. Ann Allergy 1982;49:127-130

PART 4

The Effects of Etidronate in the Prevention and Treatment of Glucocorticoid-Induced Osteoporosis

CHAPTER 7

ACUTE EFFECTS OF ETIDRONATE ON GLUCOCORTICOID-INDUCED BONE DEGRADATION

A.Struijs[1], A. Smals[2], S.A. de Witte[1], W.H.L. Hackeng[3], H. Mulder[4]
[1]Erasmus Medical Centre, Rotterdam, [2]Division of Endocrinology, University Hospital, Nijmegen, [3]Praktijk voor Laboratorium Diagnostiek, Spijkenisse, [4]Site Management Organisation "Good Clinical Practice", Rotterdam

(accepted in Rheumatology)

SUMMARY

Objectives. To study the acute short term effects on the biochemical parameters of calcium and bone homeostasis in postmenopausal women treated with high dose of prednisone alone or with additional etidronate, before and during 5 days of treatment.
Methods. Serum calcium, phosphorus, creatinine, alkaline phosphatase activity, osteocalcin, carboxy terminal propeptide of type I procollagen (PICP), cross-linked carboxy terminal telopeptide of type I collagen (ICTP), PTH, 25 hydroxy vitamin D and urinary excretion of calcium over 24 hours were measured before and during 5 days of treatment in fourteen postmenopausal women treated with high dose of prednisone (60 mg/day) alone (group A) or combined with cyclical etidronate (group B).
Results. Significant differences from baseline were found in osteocalcin and urinary excretion of calcium in both groups and for ICTP in group B. Significant differences between groups were calculated at day 5 of the study for osteocalcin, ICTP and 24 hours of urine calcium excretion ($p < 0,01$). Urinary excretion of calcium over 24 hours increased in group A (+ 14.7%; $p < 0,05$) and decreased in group B (- 22.1%; $p < 0,01$). Osteocalcin levels decreased in group A (-38.1%) and increased in group B (+27.4%; both $p < 0,01$). ICTP decreased only in group B (-19.4%; $p < 0,01$).

Conclusions. The results are consistent with the fact that etidronate is able to acutely prevent bone resorption due to corticosteroids. The increase in osteocalcin in the etidronate treated group is a new feature. A direct or indirect (PTH, 1,25 vitamin D) stimulatory effect of etidronate on the osteoblast cannot be excluded.

KEY WORDS

Prednisone, corticosteroid, glucocorticoid, etidronate, bisphosphonate, osteoporosis, calcium metabolism and bone markers.

INTRODUCTION

Osteoporosis and pathological fractures due to use of prednisone, in a daily dosage of more than 7,5 mg for periods longer than 6 months have been recognised for a long time [1-3]. However the contribution of different pathofysiological mechanisms is still unclear. Supraphysiologic doses of glucocorticosteroids, given to animals and men result a negative calcium balance [4-7], which is associated with secondary hyperparathyroidism contributing to further calcium and bone loss [7-9]. Also direct effects of glucocorticosteroids on osteoblasts have been described [10-12]. Nowadays it becomes clear, that with the use of glucocorticosteroids for shorter or longer time period's [13], an uncoupling between bone resorption and formation occurs. Chronic use of glucocorticosteroids increases resorption and decreases formation, which leads to the development of osteopenia and fractures [2,3,14].

Different treatment modalities as hormone replacement therapy, calcitonin, fluor and bisphosphonates have been used to prevent or treat low bone mass due to glucocorticosteroids [15-24]. We and others found a pronounced increase of BMD in patients with steroid induced osteoporosis treated with etidronate or alendronate [21-24]. The results suggested an additional effect of bisphosphonates apart from decreased resorption alone. No data however have been published dealing with the acute effect of bisphosphonates on glucocorticoid- induced changes in circulating bone markers. The objective of the current study was to determine the short-term effect of the bisphosphonate etidronate on glucocorticoid-induced changes in biochemical parameters of calcium and bone homeostasis in postmenopausal women with temporal arteriitis treated with high dosage of glucocorticosteroids.

PATIENTS AND METHODS

Study design

This prospective randomised open-label study was conducted in an outpatient clinic. Within a time period of twelve months fourteen postmenopausal patients with histological proven temporal arteriitis were randomly allocated to the following treatments: high dose of prednisone (60 mg/daily) alone (group A) or high dose prednisone (60 mg/daily with etidronate 400 mg daily (group B) for five days. Patients with any disease or medication that could interfere with calcium or bone metabolism were excluded from the study. All patients gave informed consent to the study. The procedures followed were in adherence with the guidelines of the Helsinki declaration for physician initiated studies.
The study participants took their prednisone at 8.00 a.m.. The patients in group B received 400 mg etidronate one hour before prednisone intake. Breakfast was served at 9.00 a.m. The patients had a dietary calcium intake of 400-800 mg before the study and they were kept on this level during the study.

Measurements

Blood and urine were collected at 7.00 a.m. daily for five days starting one day before any medication was taken. The following biochemical parameters were evaluated before and during the first five days of treatment: serum calcium, phosphorus, creatinine, alkaline phosphatase activity, osteocalcin, carboxy terminal propeptide of type I procollagen (PICP), cross-linked carboxy terminal telopeptide of type I collagen (ICTP), PTH and 25 hydroxy vitamin D. Urinary excretion of calcium over 24 hours was measured daily from day –1 to day 5. The serum and urine calcium, serum phosphorus, alkaline phosphatase and creatinine were measured by autoanalyzer techniques. The samples for hormonal measurements were immediately frozen and stored at –20°C until use. All hormonal measurements were performed in a one assay session.
The radio immunoassay (RIA) used for osteocalcin measurements was the commercial available kit of Incstar Corporation, Stillwater Minnesota, USA (intra-interassay coefficient of variation (CV) 3,8% and 4.3% respectively; sensitivity of the assay was 0,5 µg/l; normal values 1,8-6,6 µg/l). PICP and ICTP results were obtained with RIA produced by Farmos Diagnostica (Orion Corporation Farmos P.O. Box 425, SF-20101 Turku Finland). The intra- and interassay CV were 4,7% and 5.3% for PICP and 6.2% and 7.9% for ICTP.
Sensitivity was 1,2 µg/l for P1CP and 0,43 µg/l for 1CTP; normal values P1CP, 50-170 µg/l and 1CTP 1,4-5,4 µg/l. PTH was measured by a two-side RIA. The first step involved extraction and concentration of plasma PTH using solid phase anti-amino-terminal antibodies. After elution, the PTH immunoextract was analyzed using a sensitive mid- and

C-region immunoassay [25]. The intra- and interassay CV were 8.3% and 10.2% respectively; the sensitivity of the assay was 0.8 pmol/l (normal values 0.8-5.0 pmol/l).
Serum 25- OH-D was quantified, after acetonitrile extraction, using a direct commercial RIA (Incstar Corporation, Stillwater, MN, USA). The intra- and interassay CV were 6.3% and 13.0% respectively (normal values 25-125 nmol/l).

Statistics

Data were expressed as mean ± SEM, with 95% confidence intervals of the mean. The significance of change from baseline was determined by a paired t-test, SPSS windows version 8.0 (p denoted by p). The significance of difference between means of biochemical parameters, at the end of the study (day 5), was determined by an independent two-sample t-test (p denoted by p*). Also analysis of covariance (ANCOVA, with mean pre-treatment values determinations and time as covariable) was performed to compare the differences between groups at day 5 of the study. Differences were considered statistically significant when $p < 0.05$.

RESULTS

Fourteen postmenopausal women were randomised to group A and group B. Each group comprised seven women. At baseline the two treatment groups were comparable in age and biochemical parameters. Group A; n= 7; mean age 72 yr., range 59-82 yr. Group B; n=7; mean age 74 yr., range 63-82 yr. The results are given in Table 1 and illustrated for osteocalcin (Fig.1), urinary excretion of calcium over 24 hours (Fig. 2) and the telopeptide ICTP (Fig. 3). In both groups of prednisone alone and combined prednisone and etidronate treatment serum calcium and phosphate levels, alkaline phosphatase PTH and 25 OH vitamin D levels virtually remained unchanged.
In the prednisone alone group, serum osteocalcin levels significantly decreased from 2.7 ± 0.3 mg/L at baseline to 1.7 ± 0.1 mg/L after 5 days (- 38,1%, $p < 0.01$). In contrast to the combined treatment group serum osteocalcin levels steadily increased from 2.4 ± 0.3 mg/L at baseline to 3.0 ± 0.4 mg/L after 5 days (+ 27.4%, $p < 0.01$). The Δ change in osteocalcin between the two groups was statistically significant from day 3 ($p^* < 0.05$). The procollagen I levels remained unchanged in both treatment groups.
The bone resorption marker ICTP remained virtually unchanged during prednisone treatment alone, but instead gradually decreased in the bisphosphonate group to levels 19.4% below baseline at day 5 ($p < 0.01$). At that time the difference between both groups was statistically significant ($p^* < 0.01$). Urinary calcium excretion gradually increased over

Figure 1. Time course of serum osteocalcin (mean ± SEM).
(group A: prednisone; group B: prednisone and etidronate)

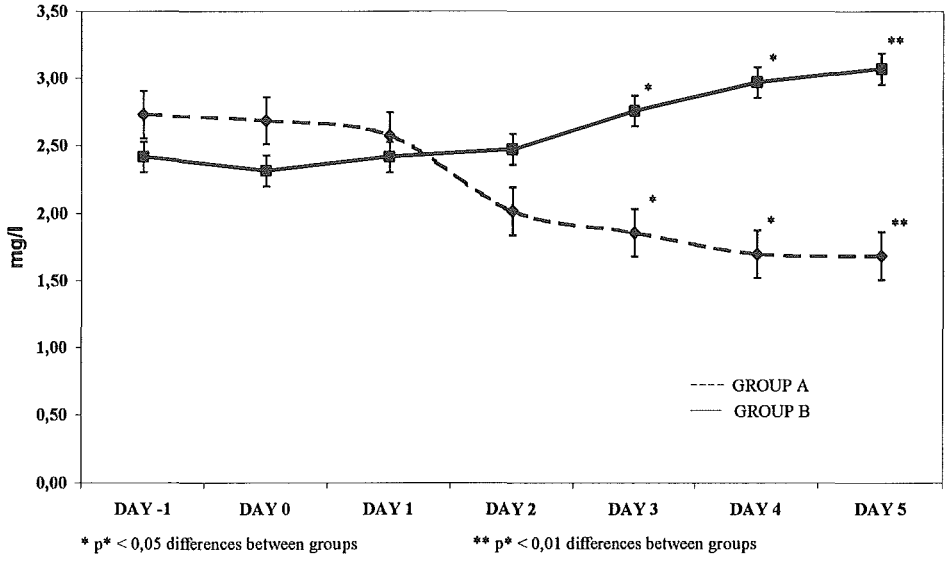


Figure 2. Time course of 24 hours of urinary calcium excretion (mean ± SEM).
(group A: prednisone; group B: prednisone and etidronate)

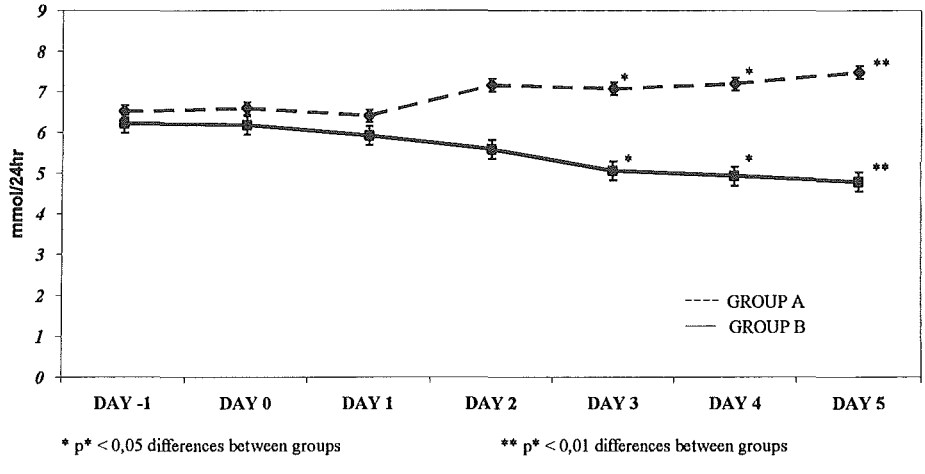
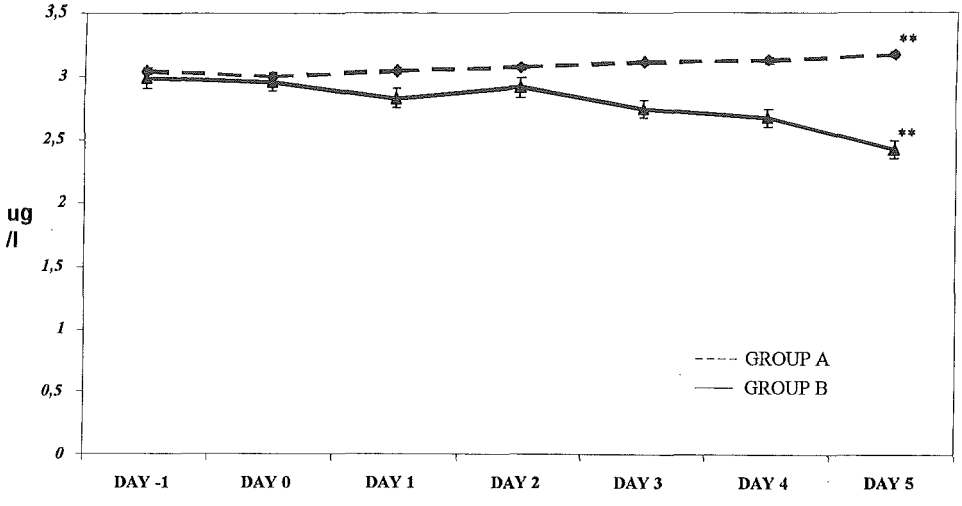


Table 1. Biochemical markers of calcium and bone homeostasis (mean ± SEM and 95% CI).
(group A: prednisone ; group B: prednisone and etidronate)

Group		Before	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5
SeCa (mmol/l)	A	2.36± 0.04(2.26-2.46)	2.35± 0.04(2.26-2.43)	2.37± 0.04(2.26-2.47)	2.35±0.05(2.24-2.46)	2.35±0.04(2.26-2.43)	2.34±0.04(2.24-2.44)	2.34±0.04(2.25-2.44)
	B	2.34± 0.05(2.23-2.45)	2.32± 0.05(2.20-2.44)	2.30± 0.04(2.21-2.39)	2.33±0.04(2.23-2.41)	2.34±0.04(2.24-2.43)	2.33±0.04(2.23-2.42)	2.34±0.05(2.23-2.45)
SePO4 (mmol/l)	A	1.12±0.07(0.95-1.28)	1.12±0.06(0.98-1.26)	1.14±0.06(1.00-1.28)	1.18±0.06(1.02-1.33)	1.10±0.07(0.94-1.27)	1.12±0.07(0.95-1.29)	1.11±0.07(1.00-1.21
	B	1.10±0.08(0.91-1.28)	1.08±0.07(0.91-1.25)	1.10±0.06(0.94-1.26)	1.07±0.07(0.91-1.24)	1.11±0.07(0.93-1.29)	1.08±0.06(0.92-1.24)	1.03±0.06(0.88-1.17)
Uca (mmol/l)	A	6.5 ± 0.6 (5.1-7.9)	6.6 ± 0.7 (4.8-8.4)	6.4 ± 0.8 (4.4-8.4)	7.2 ± 0.7 (5.4-8.9)	7.1 ± 0.9 (4.9-9.3)	7.2 ± 0.9 (5.0-9.3)	7.5 ± 0.8 (5.5-9.4) [2,]
	B	6.2 ± 0.8 (4.3-8.1)	6.2 ± 0.7 (4.3-8.0)	5.9 ± 0.9 (3.7-8.1)	5.6 ± 0.8 (3.7-7.4)	5.0 ± 0.6 (3.5-6.6)	4.9 ± 0.7 (3.1-6.7)	4.8 ± 0.6 (3.4-6.1) [1,3]
Alk Phosph (U/l)	A	62 ± 6.3 (46 - 77)	61 ± 5.8 (47 - 75)	61 ± 5.0 (49-73)	62 ± 5.7 (49 - 77)	62 ± 6.2 (46 - 77)	60 ± 5.9 (46 - 75)	61 ± 5.8 (47 - 75)
	B	61 ± 4.3 (51 - 72)	61 ± 3.9 (52 - 71)	62 ± 4.3 (52-79)	62 ± 5.1 (51 - 76)	61 ± 4.7 (50 - 73)	63 ± 3.9 (53 - 72)	64 ± 4.1 (54 - 74)
Osteocalcin (mg/l)	A	2.7 ± 0.3 (1.9-3.5)	2.7 ± 0.3 (1.9-3.5)	2.6 ± 0.3 (1.9-3.3)	2.0 ± 0.2 (1.4-2.6)	1.9 ± 0.2 (1.5-2.2)	1.7 ± 0.2 (1.3-2.1)	1.7 ± 0.1 (1.4-2.0) [1,3]
	B	2.4 ± 0.3 (1.6-3.2)	2.3 ± 0.3 (1.6-3.0)	2.4 ± 0.3 (1.7-3.1)	2.5 ± 0.3 (1.7-3.2)	2.8 ± 0.3 (1.9-3.6)	3.0 ± 0.4 (2.0-4.0)	3.0 ± 0.4 (2.2-3.9) [1,3]
Procollagen1 (µgr)	A	217 ± 41 (116-318)	213 ± 38 (119-307)	207 ± 35 (120-293)	206 ± 43 (102-310)	213 ± 37 (123-303)	220 ± 45 (108-331)	219 ± 36 (131-307)
	B	266 ± 53 (136-395)	260 ± 52 (133-388)	268 ± 54 (136-400)	275 ± 49 (155-395)	277 ± 53 (148-405)	272 ± 49 (153-391)	279 ± 54 (144-409)
Telopeptide (µgr)	A	3.0 ± 0.4 (2.0-4.1)	3.0 ± 0.4 (2.1-3.9)	3.0 ± 0.4 (2.1-4.0)	3.1 ± 0.4 (2.2-4.0)	3.1 ± 0.4 (2.1-4.1)	3.1 ± 0.3 (2.3-4.0)	3.2 ± 0.3 (2.4-4.0) [3]
	B	3.0 ± 0.6 (1.5-4.4)	3.0 ± 0.6 (1.4-4.5)	2.8 ± 0.5 (1.5-4.2)	2.9 ± 0.6 (1.5-4.3)	2.4 ± 0.6 (1.4-4.1)	2.7 ± 0.6 (1.3-4.1)	2.4 ± 0.5 (1.2-3.7) [1,3]
PTH (Pmol/l)	A	1.9 ± 0.3 (1.1-2.6)	2.0 ± 0.3 (1.1-2.8)	2.0 ± 0.4 (1.1-2.8)	2.1 ± 0.3 (1.3-2.9)	2.0 ± 0.3 (1.2-2.8)	2.2 ± 0.4 (1.3-3.1)	2.1 ± 0.4 (1.2-3.0)
	B	1.6 ± 0.2 (1.0-2.1)	1.6 ± 0.2 (1.1-2.1)	1.6 ± 0.2 (1.1-2.2)	1.5 ± 0.2 (1.1-1.9)	1.6 ± 0.2 (1.2-2.1)	1.7 ± 0.2 (1.3-2.2)	1.9 ± 0.2 (1.5-2.4)
Kreatinine (µmol/l)	A	94 ± 4.5 (83-105)	95 ± 4.7 (83-106)	93 ± 5.4 (79-106)	93 ± 3.1 (86-101)	92 ± 4.9 (80-105)	91 ± 4.8 (78-103)	95 ± 4.3 (84-105)
	B	93 ± 4.2 (83-104)	90 ± 3.4 (82- 99)	93 ± 3.7 (84-102)	90 ± 3.8 (81-100)	94 ± 4.2 (84-104)	93 ± 4.4 (82-104)	92 ± 3.2 (85-100)
25 OHVit D (nmol/l)	A	42 ± 7.4 (24 - 60)	41 ± 6.9 (23 - 58)	42 ± 6.4 (27 - 58)	42 ± 7.1 (24 - 59)	42 ± 7.4 (24 - 60)	42 ± 7.8 (23 - 61)	43 ± 7.5 (25 - 62)
	B	43 ± 4.7 (31 - 54)	44 ± 4.7 (32 - 55)	44 ± 4.4 (33 - 54)	43 ± 5.4 (30 - 56)	41 ± 4.6 (30 - 52)	42 ± 5.5 (28 - 56)	43 ± 4.6 (31 - 54)

[1] P < 0.01 difference from baseline after five days
[2] P < 0.05 difference from baseline after five days
[3] P* < 0.01 difference between groups after five days

Figure 3. Time course of telopeptide (mean ± SEM).
(group A: prednisone; group B: prednisone and etidronate)



** p* < 0,01 differences between groups

five days in the prednisone alone group (+ 14.7%, $p < 0.05$) but decreased in the combined prednisone etidronate group (- 22.1%, $p < 0.01$). The difference in urinary calcium excretion between both groups was already statistically significant from day 3 on. Every patient completed the study and the study drug was well tolerated.

DISCUSSION

Short term high dose glucocorticoid therapy in postmenopausal women with florid arteritis temporalis resulted in acute changes in bone formation and resorption as reflected by a 38% decrease of osteocalcin –not PICP or Alkaline Phosphatase- and a 15% increase in urinary calcium excretion. The glucocorticoid induced changes in bone remodelling markers have been attributed to a decrease in differentiated function of mature osteoblast leading to a decrease in osteocalcin transcription and –not in this study- a decrease in

type I collagen expression and increase in its degradation [10-12,26,31]. Other studies administering 10 to 20 mg of prednisone daily for 1 week to healthy men found a similar decrease in osteocalcin (-35%) and increase in urinary calcium (+45%) as we did, however, they reported a decrease in PICP [27,28] and both decreased [27] and increased [28] ICTP has been reported. This could be explained by the different dosages of prednisone and/or the difference in study population. Similar results were obtained, in patients with rheumatoid arthritis given high dose of dexamethasone [13], in patients with multiple sclerosis given huge intravenous doses of methyl prednisolone [29] and after high dose corticosteroid inhalation [30]. From the present study it appears that combined etidronate and glucocorticoid administration acutely reversed glucocorticoid induced suppression of bone formation as reflected by a 27% increase in osteocalcin within five day's. Urinary calcium excretion decreased from + 14.7% in the prednisone alone group to –22.1% in the combined therapy group, indicating attenuation of bone resorption. This was also illustrated by a decrease in ICTP. This decrease of bone resorption markers is well known in chronically bisphosphonate treated patients on glucocorticoids, but never has been reported after such short time interval as in this study.

The increase in osteocalcin levels during short term etidronate administration in glucocorticoid treated patients is, however remarkable, although we must be careful to extrapolate our short-term study results to the long-term benefits.

There are five possible mechanisms by which this increase of osteocalcin could be explained: first decreased elimination of osteocalcin by the kidney during etidronate therapy. This is unlikely since in most studies on postmenopausal osteoporosis etidronate induces a decrease in osteocalcin. Secondly altered degradation, thirdly increased production of osteocalcin, fourthly an indirect effect of etidronate on bone turnover by influencing the inflammatory process [32] and fifthly initial displacement of osteocalcin from hydroxyapatite by EHDP as shown by Price et al in rats [33]. This could also explain the initial rise of osteocalcin found by Tobias et al in the first two weeks of treatment of postmenopausal women with HRT or a bisphosphonate [34].

A direct or indirect stimulatory effect of bisphosphonates on the osteoblast could be postulated especially in high turnover bone disease as in patients with Paget's disease, in whom an increase in osteocalcin occurs after APD treatment [35]. Also in vitamin D deficient postmenopausal osteoporotic women chronic etidronate therapy has been reported to induce a rise in osteocalcin [36]. Bisphosphonates may exert their effects on the osteoblast indirectly by at least two different mechanisms. One is the modulation in release of cytokines by the osteoclast and the release of local signals due to the effects on the osteoclast function, resulting in decreased resorption [37]. The other is through stimulation of the production of the calciotropic hormones 1,25 dihydroxyvitamin D and

PTH, which could increase during bisphosphonate treatment [6,38,39]. Recently Staal et al [40] and Zhang et al [41] reported increased expression of plasma osteocalcin by 1,25 OH vitamin D_3 which is inhibited by glucocorticoids [42]. Our hypothesis is that etidronate could reverse this mechanism. Unfortunately we have no data on plasma 1,25 vitamin D_3 levels during the short term administration of glucocorticoid alone or combined glucocorticoid and etidronate. During the short term treatment period, serum phosphate and PTH levels remained virtually unchanged in both groups. Therefore it seems unlikely that they trigger changes that could explain the increase in osteocalcin. Finally etidronate could have a direct stimulatory effect on osteoblast function in a state of glucocorticoid induced high bone turnover.

In conclusion the present study is the first demonstrating an acute beneficial effect of etidronate on glucocorticoid induced bone degradation as reflected by prevention of bone resorption and in addition stimulation of the bone formation marker osteocalcin by a thus far unknown direct or indirect effect (PTH,1.25 OH D_3) on the osteoblast. A study with more individuals, a longer follow-up period with new more specific markers and vitamin D metabolites is warranted.

ACKNOWLEDGEMENTS

We would like to thank Prof. Huibert A.P. Pols and Prof. Jonathan H. Tobias for their critical review and advice. We are grateful to K.Kragt and W.J. Jansen for their assistance with the data and statistics.

REFERENCES

1. Meunier PJ. Is steroid-induced osteoporosis preventable? N Eng J Med 1993; 328: 1781-1782.
2. Cooper C, Coupland C, Mitchell M. Rheumatoid arthritis, corticosteroid therapy and the risk of hip fracture. Ann Rheum Dis 1995; 54: 49-52
3. Lems WF, Jahangier ZN, Jacobs JWG, Bijlsma JWJ. Vertebral fractures in patients with rheumatoid arthritis treated with corticosteroids. Clin Exp Rheum 1995; 13: 293-297.
4. Need AG, Philcox JC, Hartley TF, Nordin BE. Calcium metabolism and osteoporosis in corticosteroid-treated postmenopausal woman. Aust NZ J Med 1986; 16: 341.
5. Colette C, Monnier L, Herbute NP, Blotman F, Miruoze J. Calcium absorption in corticoid treated subjects: effects of single oral dose of calcitriol. Horm Metab Res 1987; 19: 335-338.
6. Findling JW, Adams NS, Lemann J, Gray RW, Thomas CJ, Tyrrell JB. Vitamin D metabolites and parathyroid hormone in Cushing's syndrome: relationship to calcium and phosphorus homeostasis. J Clin Endocrinol Metab 1982; 54: 1039-1044.
7. Suzuki Y, Ichikawa Y, Saito E, Homma M. Importance of increased urinary calcium excretion in the development of secondary hyperparathyroidism of patients under glucocorticoid therapy. Metabolism 1983; 32: 151-156.
8. Lukert BP, Adams JS. Calcium and phosphorus homeostasis in man: Effect of corticosteroids. Arch Int Med 1976; 136: 1249-1253.
9. Gennari C, Imbimbo B, Montagnani M, Bernini M, Avioli LV. Effects of prednisone and deflazacort on mineral metabolism and parathyroid hormone activity in humans. Calcif Tiss Int 1984;36:245-252.
10. Delany AM, Dong Y, Canalis E. Mechanisms of glucocorticoid action in bone cells. J Cell Biochem 1994; 56: 295-302.
11. Chevalley T, Strong DD, Mohan S, Baylink DJ, Linkhart TH. Evidence for a role for insulin-like growth factor binding proteins in glucocorticoid inhibition of normal human osteoblast-like cell proliferation. Eur J Endocrin 1996; 134: 591-601.
12. Morrison NA, Shine J, Fragonas JC. 1.25-dihydroxyvitamin D-responsive element and glucocorticoid repression in the osteocalcin gene. Science 1989; 146: 1158-1161.
13. Lems WF, Gerrits MI, Jacobs JWG, Vugt RM, van Rijn HJM, Bijlsma JWJ. Changes in markers of bone metabolism during high dose corticosteroid pulse treatment in patients with rheumatoid arthritis. Ann Rheum Dis 1996;55:288-293.
14. Dempster DW. Bone histomorphometry in glucocorticoid-induced osteoporosis. J Bone Min Res 1989; 4: 137-141.

15. Sambrook P, Birmingham J, Kelly P, et al. Prevention of corticosteroid osteoporosis N Eng J Med 1993; 328: 1747-1752.
16. Murphy S, Khaw KT, Cassidy A, Compston JE. Sex hormones and bone mineral density in elderly men. Bone Miner 1993; 20: 133-140.
17. Bagatell CJ, Bremner WJ. Androgens in men- uses and abuses. N Engl J Med 1996; 334:707-714.
18. Reid IR, King AR, Alexander CJ, Ibbertson HK. Prevention of steroid induced osteoporosis with (3-amino-1-hydroxypropylidene)-1, 1-bisphosphonate (APD). Lancet 1988; 1: 143-146.
19. Lukert BP, Johnson BE, Robinson RG. Estrogen and progesterone replacement therapy reduces glucocorticoid-induced bone loss. J Bone Miner Res 1992; 7: 1063-1069.
20. Montemurro L, Schiraldi G, Fraioli P, Tosi G, Riboldi A, Rizzato G. Prevention of corticosteroid-induced osteoporosis with salmon calcitonin in sarcoid patients Calcif Tissue Int 1991; 49: 71-76.
21. Mulder H, Struijs A. Intermittent cyclical etidronate in the prevention of corticosteroid-induced bone loss. Br J Rheumatol 1994; 33: 348-350.
22. Struijs A, Snelder AA, Mulder H. Cyclical etidronate reverses bone loss of the spine and proximal femur in patients with established corticosteroid-induced osteoporosis. Am J Med 1995; 99: 235-242.
23. Adachi JD, Bensen WG, Brown J, et al. Intermittent etidronate therapy to prevent corticosteroid-induced osteoporosis. N Eng J Med 1997; 337: 382-387.
24. Saag KG, Emkey R, Schnitzer TJ, et al. Alendronate for the prevention and treatment of glucocorticoid-induced osteoporosis. N Engl J Med 1998; 339: 292-299.
25. Hackeng WHL, Lips P, Netelenbos C, Lips CJM. Clinical implications of estimation of intact parathyroid hormone (PTH) versus total immunoreactive PTH in normal subjects and hyper-parathyroid patients. J Clin Endocrinol Metab 1986; 63:447-453.
26. Lems WF, Jacobs JWG, van den Brink HR, van Rijn HJM, Bijlsma JWJ, Transient decrease in osteocalcin and markers of collagen type 1 formation during corticosteroid pulse therapy. Br J Rheum 1993;32:787-790.
27. Gram J, Junker P, Nielsen HK, Bollerslev J. Effects of short-term treatment with prednisolone and calcitriol on bone and mineral metabolism in normal men. Bone 1998; 23:297-302.
28. Lems WF, Jacobs JWG, Rijn HJM van, Bijlsma JWJ. Changes in calcium and bone metabolism during treatment with low dose prednisone in young, healthy, male volunteers. Clinical Rheumatology 1995;14:420-424.
29. Cosman F, Nieves J, Herbert J, Shen V, Lindsay R. High-dose glucocorticoids in multiple sclerosis patients exert direct effects on the kidney and skeleton. J Bone Miner Res 1994;9:1097-1105.
30. Barnes PJ, Pedersen S, Busse W. Efficacy and safety of inhaled corticosteroids. Am J Resp Crit Care Med 1998;157:S1-S53.
31. Beresford JN, Gallagher JA, Poser JW, Russel RGG. Production of osteocalcin by human bone cells in vitro. Effects of $1,25(OH)_2D_3$, $24,25(OH)_2D_3$, parathyroid hormone and glucocorticoids. Metab. Bone Dis Rel Res 1984; 5: 229-234.
32. Bijvoet OLM, Frijlink K, Jie K et al. APD in Paget's disease of bone. Role of the mononuclear phagocyte system? Arthritis and Rheumatism; 1980;23:1143-1203.
33. Price PA, Williamson MK, Baukol SA. The vitamin K-dependent bone protein and the biological response of bone to 1,25 dihydroxyvitamin D_3. In Veis A (ed) The chemistry and biology of mineralised connective tissues. Elsevier/North Holland, New York 1981; 327-335.
34. Tobias JH, Laversuch CJ, Chambers TJ, Gallagher AC. Aminohexane bisphosphonate suppresses bone turnover in post menopausal women more rapidly than oestrogen therapy. Brit J Rheumatol

1996;35:636-641.

35. Papapoulos SD, Frohlich M, Mudde AH, Harinck HIJ, Berg H, Bijvoet OLM. Serum osteocalcin in Paget's disease of bone: Basal concentrations and response to bisphosphonate treatment. J Clin Endocrinol Metab 1987;65:89-94.
36. Koster JC, Hackeng WHL, Mulder H. Diminished effect of etidronate in vitamin D deficient osteopenic postmenopausal women. Eur J Clin Pharmacol 1996;51(2): 145-147
37. Boonekamp PM, vd Wee-Pals LJA, v Wijk-v Lennep MML, Thesingh CW, Bijvoet OLM. Two modes of action of bisphosphonates on osteoclastic resorption of mineralised matrix. Bone Mineral 1986;1: 27-39.
38. Adami S, Frijlink WB, Bijvoet OLM, O'Riordan JHL, Clemens TL, Papapoulos SE. Regulation of calcium absorption by 1,25-dihydroxyvitamin D. Studies of the effect of a bisphosphonate treatment. Calcif Tissue Int 1982;34:317-320.
39. Papapoulos SE, Harinck HIJ, Bijvoet OLM, Gleed JH, Fraher LJ, O'Riordan JHL. Effects of decreasing serum calcium on circulating parathyroid hormone and vitamin metabolites in no mocalcaemic and hypercalcaemic patients treated with APD. Bone Mineral 1986; 1: 69-78.
40. Staal A, van den Bemd GJ, Birkenhager JC, Pols HAP, van Leeuwen JP. Consequences of vitamin D receptor regulation for the 1,25 dihydroxyvatamin D_3 –induced 24-hydroxylase activity in osteoblast-like cells: initiation of the c24- oxidation pathway. Bone 1997;20:237-243.
41. Zhang R, Ducy P. Karsenty G. 1,25 dihydroxyvitamin D_3 inhibits osteocalcin expression in mouse through an indirect mechanism. J Biol Chem 1997;272:110-116.
42. Lian JB, Shalhoub V, Aslam F, Frenkel B, Green J, Hamrah M, Stein GS, Stein JL. Species specific glucocorticoid and 1,25 dihydroxyvitamin D_3 responsiveness in mouse mc3t3-E1 osteoblast: dexamethasone inhibits osteoblast differentiation and vitamin D down-regulates osteocalcin gene expression. Endocrinology 1997;138:2117-2127.

CHAPTER 8

THE PREVENTION OF GLUCOCORTICOID-INDUCED OSTEOPOROSIS WITH ETIDRONATE IN POSTMENOPAUSAL WOMEN WITH TEMPORAL ARTERITIS

H. Mulder, M.D. and A. Struys, M.D.

(Published as; "Etidronate Prevents Steroid-Induced Bone Loss" in Br J Rheumatol 1994;33:348-350)

SUMMARY

We conducted a prospective study about the effect of etidronate on corticosteroid-induced bone loss in postmenopausal women with temporal arteritis for whom high-dose prednisone therapy was indicated. Group A (n = 10) received etidronate (400 mg/day for 2 weeks, then 11 weeks off etidronate; four cycles total) and prednisone; Group B (n = 10) received only prednisone. Vertebral bone mineral density (BMD) was measured blinded by dual x-ray absorptiometry.

At 3, 6 and 12 months, vertebral BMD was significantly ($p < 0.01$) increased in Group A and decreased in Group B, based on mean actual and percent changes in BMD and mean changes in BMD Z-score from baseline. Between-group comparisons were also significant ($p < 0.002$) at each time point. No adverse events related to etidronate treatment were reported.

Our results suggest that instituting intermittent cyclical etidronate therapy when high-dose prednisone therapy is begun may prevent corticosteroid-induced bone loss. Further research into bisphosphonate use in corticosteroid-induced bone loss (with larger patient populations, longer follow-up and fracture assessment) is warranted.

KEY WORDS

Bone loss; Corticosteroids; Etidronate; Prednisone, Temporal arteritis

INTRODUCTION

The concept that excess endogenous glucocorticoid can lead to an increased tendency to fracture was first described by Cushing in 1932 [1]. Soon after the introduction of cortisone as a therapeutic agent, it became clear that exogenous hypercortisolism was also deleterious to the skeleton [2]. Both bone quantity and bone quality are decreased with the use of corticosteroids. Prospective studies of bone mass during corticosteroid treatment have confirmed that their use can cause progressive bone loss [2]. Data suggest that up to 50% of patients taking glucocorticoids long-term will develop osteoporosis and experience atraumatic fractures due to the resultant compromised bone quality [2-4].

The pathogenesis of corticosteroid-induced bone loss is associated with decreased bone formation and increased bone resorption [2]. Decreased intestinal calcium absorption and increased urinary calcium excretion are also attributable to corticosteroid use and contribute to a negative calcium balance [2]. This negative balance in turn leads to secondary hyperparathyroidism that results in increased bone resorptive activity [2]. These effects are manifested by accelerated bone loss early in treatment (within the first 6-12 months), followed by continuing losses and slower rates in subsequent years [2,5].

Although the effects of steroids on bone and calcium balance are well described, no fully safe and effective therapeutic or prophylactic regimen for corticosteroid-induced bone loss has been established. However, antiresorptive agents such as estrogen [6], calcitonin (given either parentally or nasally) [7], and the bisphosphonate pamidronate [8,9] have been reported to be effective in preventing or reversing corticosteroid-induced bone loss. In addition, the prophylactic use of calcitriol and calcium, with or without calcitonin has been shown to prevent corticosteroid-induced bone loss [10].

The bisphosphonate etidronate, given in an intermittent cyclical regimen, has been demonstrated to increase vertebral bone mass significantly and reduce the incidence of vertebral fractures in patients with postmenopausal osteoporosis [9,11]. The objective of the current study was to determine whether this antiresorptive agent, when administered in a similar manner and at the outset of corticosteroid therapy, could prevent the early accelerated bone loss associated with pharmacological doses of prednisone.

MATERIALS AND METHODS

Study Design

This study was conducted as a prospective 1-year clinical trial according to the guidelines of the Helsinki declaration for physician-initiated studies. Patients were recruited from one investigation site (IJsselland Hospital, the Netherlands) from April 15, 1990 to February 1, 1991. The study enrolled postmenopausal women with a biopsy-confirmed diagnosis of temporal arteritis for whom high-dose prednisone therapy was indicated. Patients were excluded from the study if they had any disease or were receiving any medication, with the exception of the study drugs, that would interfere with calcium or bone metabolism. A complete history and physical examination including routine haematology, serum chemistry, and urinalysis evaluations were performed before treatment.

Patients were alternately assigned to one of two treatment groups. Group A received intermittent cyclical oral etidronate disodium 400 mg (Didronel®, Procter & Gamble Pharmaceuticals, Norwich, New York) once daily for 2 weeks, followed by 11 weeks off therapy (four cycles total) in conjunction with appropriate prednisone therapy. Group B received only prednisone therapy. The daily prednisone dosage in both groups was 60 mg once daily for 4 weeks, followed by dose adjustment (decrease) to maintain an erythrocyte sedimentation rate (ESR) below 30 mm/hour. Patients had a daily dietary calcium intake of 400-800 mg; no supplemental calcium was administered.

Assessment

Vertebral (L1-L4) bone mineral density (BMD) was measured by dual x-ray absorptiometry (Hologic QDR 1000) at baseline and at 3, 6 and 12 months by a technician blinded to treatment assignment. Serum alkaline phosphatase (a biochemical marker of bone turnover) was measured at baseline and 12 months. Patients were judged eligible for evaluation if they followed the prescribed regimens and had the scheduled BMD determinations during the 12-month study.

Statistics

Vertebral BMD data, serum alkaline phosphatase, and the respective actual and percent changes from baseline in these clinical variables are displayed as mean ± SEM. Statistical comparisons (Minitab, Release 5.1, Minitab, Inc., State College, Pennsylvania) were performed by one-sample t-tests for between-groups changes, respectively. Z-scores (comparing individual patients' BMD with the mean of BMD of age-matched normal women) were calculated at baseline and at 3, 6 and 12 months; T-scores (comparing indi-

vidual patients' BMD with the mean BMD of young normal women) were calculated only at baseline. Statistical significance was assigned as $p < 0.05$. Ninety-five percent confidence intervals (95% CI) were calculated by a two-sample t procedure.

RESULTS

Ten women received intermittent cyclical etidronate concurrently with the prescribed prednisone regimen (Group A); 10 women received only the prednisone regimen (Group B). The mean age of the patients receiving etidronate and prednisone treatment was 74 years (range, 62-82 years), and the mean age of the control patients was 72 years (range 58-84 years). Baseline BMD (Table I) was not statistically significantly different between the groups. At baseline, Z-scores were 0.00±0.33 in Group A and 0.21±0.20 in Group B (95% CI: -1.02, 0.61); T-scores were -2.26±0.45 in group A and -1.80±0.32 in Group B.

Each of the patients responded to the prednisone therapy for temporal arteritis. Mean prednisone dosages of the 12 month study period were 11.0 mg/day in Group A and 10.7

TABLE 1

Vertebral Bone Mineral Density

Measurement	Length of treatment (Months)	Group A	Group B	95% CI*
BMD (g/cm^2)	Baseline	0.798 ± 0.050	0.850 ± 0.035	-0.180, 0.076
	3	0.802 ± 0.050	0.829 ± 0.034	-0.155, 0.101
	6	0.807 ± 0.050	0.815 ± 0.033	-0.136, 0.121
	12	0.809 ± 0.050	0.807 ± 0.032	-0.125, 0.129
Change from baseline (g/cm^2)	3	0.005 ± 0.001†‡	-0.020 ± 0.002‡	0.02, 0.030
	6	0.009 ± 0.003†‡	-0.035 ± 0.004‡	0.034, 0.054
	12	0.011 ± 0.003†‡	-0.042 ± 0.006‡	0.040, 0.068
Percentage change from baseline (%)	3	0.58 ± 0.14†‡	-2.36 ± 0.25‡	2.33, 3.57
	6	1.10 ± 0.31†‡	-4.09 ± 0.45‡	4.02, 6.36
	12	1.42 ± 0.45†‡	-4.95 ± 0.64‡	4.71, 8.03
Change from baseline (Z-score)	3	0.06 ± 0.01†‡	0.01 ± 0.00‡	0.02, 0.07
	6	0.11 ± 0.02†‡	0.10 ± 0.03‡	0.14, 0.29
	12	0.16 ± 0.03†‡	-0.15 ± 0.04‡	0.20, 0.41

Group A (N=10) received intermittent cyclical etidronate in conjunction with high-dose prednisone; Group B (N=10) received high-dose prednisone only. Vertebral BMD.

*95% CI applies to the difference between treatment groups and was calculated by a two-sample *t*-procedure.

†P <0.002, compared with Group B; two sample *t*-test.

‡P <0.01, compared with baseline; one-sample *t*-test.

mg/day in Group B.

At 3, 6 and 12 months, vertebral BMD was significantly ($P < 0.01$) increased in Group A and decreased in Group B, based on mean actual and percent changes from baseline in BMD and mean changes from baseline in BMD Z -score (table I). Between-group comparisons were also significant ($P < 0.002$) at each time point. At 12 months, the mean percent change from baseline in BMD was +1.42±0.45% in Group A and -4.95 ±0.64% in Group B; the 95% CI showed that the difference between the two treatment groups was at least 4.71% and as much as 8.03%. Nine of the 10 patients treated with etidronate and prednisone had increases in bone mass, whereas all 10 patients treated with only prednisone had decreases (Fig.1).

At 12 months, the change from baseline in serum alkaline phosphatase was 6.2±3.9% in Group A and -5.3±3.0% in group B ($p < 0.05$, between groups). No clinically significant changes in routine clinical and laboratory evaluations were noted. Etidronate was well tolerated, and no adverse events related to etidronate treatment were reported.

DISCUSSION

Our study demonstrated that intermittent cyclical etidronate can prevent the negative effects of continuous prednisone use on bone mass. The study patients were at high risk of developing osteoporosis, being elderly postmenopausal women requiring first-time high-dose prednisone treatment for 6 months or longer. The significant and progressive decreases from baseline in BMD in the group treated with prednisone alone confirmed a continued loss of bone mass induced by prednisone in excess of losses due to age or menopause. In contrast, the patients who received concurrent etidronate exhibited significant increases in bone mass that were comparable to those seen in studies of etidronate in the treatment of postmenopausal osteoporosis [11,12]. Significant increases from baseline in BMD Z-scores in this group further suggest a beneficial protection from or prevention of these progressive corticosteroid-induced and age-related losses in bone mass. This is especially important because of the recognised strong inverse relationship between low bone mass and an increased risk of atraumatic vertebral fractures [13].

Controversy currently exists about the overall influence of clinical factors (such as age, sex, underlying disease, dose, treatment duration etc.) on corticosteroid-induced bone loss. Studies in heterogeneous patient populations have shown variable results as to the contribution of these and other factors. However, because we studied a highly uniform population, the conclusions drawn from our results can be interpreted independent of other possibly confounding clinical influences.

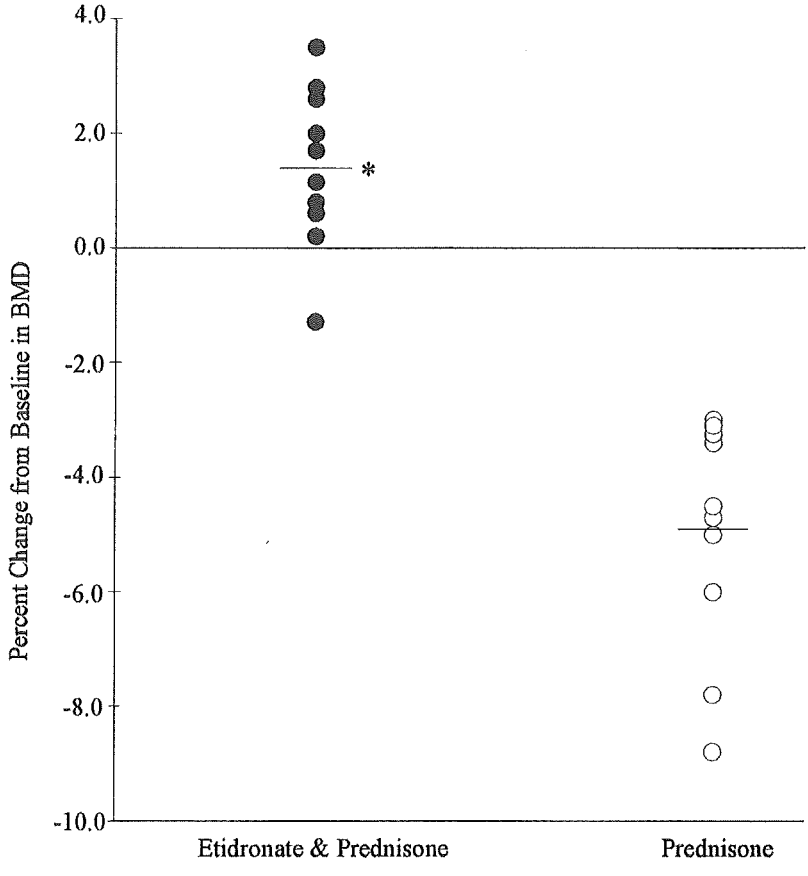


Figure 1

Percent change from baseline In vertebral BMD after twelve months of treatment with intermiitent cyclical etidronate in combination with high dose prednisone (N=10) or high dose prednisone only (N=10). Means are shown as horizontal bars.

*$p < 0.001$, compared with high dose prednisone; two-sample *t*-test

The result of this study of etidronate and those of Reid et al [8,9] in their study of another bisphosphonate, pamidronate, suggest that this class of antiresorptive drugs can counteract corticosteroid-induced bone loss by producing an overall increase in bone mass. It is also important to note that, in our study, etidronate treatment was initiated at the same time as the patients' first exposure to prednisone. This is in contrast with the Reid et al. investigations, in which all patients had already received long-term corticosteroid treatment and likely experienced bone loss before pamidronate intervention was begun [8,9]. Our results suggest that instituting intermittent cyclical etidronate therapy as soon as high-dose prednisone therapy is begun can effectively prevent this drug-induced bone loss. However, because our study involved a limited number of patients and was restricted to only those with temporal arteritis, further studies to confirm the efficacy of bisphosphonates in corticosteroid-induced bone loss (i.e. with larger patient populations, longer follow-up, and assessment of fractures) is warranted.

ACKNOWLEDGEMENT

The authors gratefully acknowledge the valuable contribution of Mrs. H.A.A. Snelders, Director of Clinical Research, Good Clinical Practice, Bergweg 66, Rotterdam, for performing bone mass measurements.

REFERENCES

1. Cushing H. The basophil adenomas of the pituitary body and their clinical manifestations (pituitary basophilism). Bull John Hopkins Hosp 1932; 1:137-92.
2. Lukert BP, Raisz LG. Glucocorticoid-induced osteoporosis: Pathogenesis and management. Ann Intern Med 1990; 112:352-64.
3. Adinoff AD, Holister JR. Steroid-induced fractures and bone loss in patients with asthma. New Engl J Med 1983; 309:265-8.
4. Ruegsegger P, Medici TC, Anliker M. Corticosteroid-induced bone loss. A longitudinal study of alternate day therapy in patients with bronchial asthma using quantitative computed tomography. Eur J Clin Pharmacol 1983; 25:615-20.
5. Silverman SL. Management of corticosteroid-induced osteoporosis: A clinician's perspective (editorial). Calcif Tissue Int 1992; 50:101-3.
6. Lukert BP, Johnson BE, Robinson RG. Estrogen and progesterone replacement therapy reduces glucocorticoid-induced bone loss. J Bone Miner Res 1992; 7:1063-9.
7. Montemurro L, Schiraldi G, Fraioli P, Tosi G, Riboldi A, Rissato G. Prevention of corticosteroid-induced osteoporosis with salmon calcitonin in sarcoid patients. Calcif Tissue Int 1991; 49:71-6.
8. Reid IR, King AR, Alexander CJ, Ibbertson HK. Prevention of steroid-induced osteoporosis with (3-amino-1-hydroxypropylidene)-1, 1-bisphosphonate (APD). Lancet 1988; 1:143-6.
9. Reid IR, Heap SW, King AR, Ibbertson HK. Two year follow-up of bisphosphonate (APD) treatment in steroid osteoporosis. Lancet 1988; 2:1144.
10. Sambrook P, Birmingham J, Kelly P, Kempler S, Ngnyen T,Pocock N, Eisman J. Prevention of corticosteroid osteoporosis. N Engl J Med 1993; 328:1747-52.
11. Storm T, Thamsborg G, Steiiche T, Genant H, Sorensen OH. Effect of intermittent cyclical etidronate therapy on bone mass and fracture rate in women with postmenopausal osteoporosis. N Engl J Med 1990; 322:1265-71.
12. Watts NB, Harris ST, Genant HK, et al. Intermittent cyclical etidronate treatment of post-menopausal osteoporosis. N Engl J Med 1990; 323:73-9.
13. Ross PD, Wasnich RD, Vogel JM. Detection of pre-fracture spinal osteoporosis using bone mineral absorptiometry. J Bone Miner Res 1988; 3:1-11.

CHAPTER 9

INTERVENTION WITH ETIDRONATE IN PATIENTS WITH GLUCOCORTICOID-INDUCED OSTEOPOROSIS

Ard Struys, M.D., Anneke A. Snelder, R.N., Henk Mulder, M.D., PhD

(Published as "Cyclical Etidronate Reverses Bone Loss of the Spine and Proximal Femur in Patients with Established Corticosteroid-Induced Osteoporosis" in Am J Med1995; 99:235-242)

SUMMARY

The purpose of the study was to compare the bone-mass effects of calcium supplementation and intermittent cyclic etidronate in patients with established corticosteroid-induced osteoporosis.

Eighteen male and 21 female patients who had established corticosteroid-induced osteoporosis and were receiving chronic prednisone therapy (> 10 mg/d) were enrolled in a prospective 12-month, open-label study. In addition to continuing prednisone therapy, patients received continuous calcium supplementation 500 mg/d (n = 20) or four cycles of intermittent cyclic etidronate therapy consisting of etidronate 400 mg/d for 14 days, followed by calcium 500 mg/d for 76 days (n = 19). Bone mineral density (BMD) of the spine (L1-L4) and proximal femur (total hip, femoral neck, trochanter, Ward's triangle) was measured by dual-energy x-ray absorptiometry (DEXA) at baseline, 6 months, and 12 months by staff blinded to the treatment. Serum calcium, phosphorus, and alkaline phosphatase were also measured at the these times.

Treatment with intermittent cyclic etidronate for 12 months resulted in significant increases of 5.7% and 6.8% in BMD of the spine and proximal femur (total hip), respectively ($p < 0.02$ versus baseline; $p < 0.001$ versus calcium group). Calcium supplementation alone did not prevent significant losses of 3.4% and 4.1% in BMD of those respec-

tive sites ($p < 0.02$ versus baseline). Z scores at the end of study reflected significant increases in BMD of the spine and proximal femur (all regions) in the etidronate group ($p < 0.01$) and significant decreases at the spine, proximal femur, and trochanter in the calcium group ($p < 0.01$). After 12 months, the difference between the groups was 9.1% ($p < 0.01$; 95% CI: 6.3% to 11.9%) at the spine and 10.9% ($p < 0.01$; CI 7.8% to 14.1%) at the proximal femur (total hip). Seventeen (89%) of the etidronate-treated patients had increases in BMD of both skeletal sites, whereas only 2 (10%) and 3 (15%) of the calcium-treated patients had positive changes in BMD of the spine and proximal femur (total hip), respectively ($p < 0.01$). Serum calcium, phosphorus, and alkaline phosphatase levels did not change significantly during the study in either treatment group. Both treatment regimens were well tolerated, with no interactions between prednisone therapy and the study medications. Analyses of response by subgroups (female/male, pulmonary/non-pulmonary) showed no significant attribute-dependent changes during the 12-month study. At baseline, women had significantly lower baseline BMD of the spine and proximal femur (total hip) ($p < 0.01$), and patients with pulmonary disease had significantly longer duration of prednisone therapy and cumulative prednisone dose ($p < 0.03$).
We concluded that, intermittent cyclic etidronate reversed the progressive loss of bone mineral density of the spine and proximal femur in female and male patients with established osteoporosis secondary to chronic corticosteroid (prednisone) therapy for pulmonary and non-pulmonary diseases. Calcium supplementation alone did not prevent or attenuate these corticosteroid-induced losses.

KEY WORDS

Bone mass, calcium, corticosteroid, cyclic, etidronate, osteoporosis, prednisone, proximal femur, spine

INTRODUCTION

The osteoporosis and pathological fractures associated with prolonged therapy with corticosteroids have been recognised for over four decades. Corticosteroid-induced osteoporosis may result from two concomitant mechanisms of basic bone function: inhibition of bone formation through direct corticosteroid action on osteoblasts and increased bone resorption due to indirect enhancement of osteoclastic activity [1,2]. Corticosteroids also inhibit the absorption of calcium in the gut [3]. The resulting negative bone balance pro-

duces bone loss, particularly at skeletal sites rich in trabecular (or cancellous) bone [4]. This bone loss is rapid in the first few months and seems to proceed at a slower rate after 6 to 12 months on therapy [5].

The means for the prevention and treatment of corticosteroid-induced osteoporosis remain poorly investigated. Among the treatment modalities proposed for this serious medical condition are supplemental calcium at doses up to 1500 mg/d, administered with or without vitamin D and/or thiazide [2,6-8]; estrogen replacement therapy for postmenopausal women who require prolonged corticosteroid therapy [9]; calcitonin [8,10,11]; bisphosphonates [12,13]; and fluoride [14]. Estrogen, calcitonin, and bisphosphonates are pharmacologically bone antiresorptive agents, and thus, their study in the clinic appears to have a rational basis.

Controlled clinical studies of the bisphosphonates have shown that these agents can increase vertebral bone mass both in patients with postmenopausal (primary) osteoporosis and in those with corticosteroid-induced (secondary) osteoporosis. In patients with established postmenopausal osteoporosis, an intermittent cyclic regimen of oral etidronate produced significant increases in bone mineral density of the spine and also reduced the rate of vertebral fractures [15-17]. We also recently reported that a similar regimen of etidronate prevented vertebral bone loss when therapy was initiated at the same time as corticosteroid treatment [18]. In a study of oral pamidronate, patients with established corticosteroid-induced osteoporosis experienced a significant increase in vertebral bone mineral density after 12 months of treatment [12]. Other investigators have also reported encouraging preliminary results in the treatment of established corticosteroid-induced osteoporosis with intermittent cyclic regimens with oral etidronate [19-22] and an intermittent regimen with intravenous pamidronate [22].

We conducted a 12-month, prospective clinical study to compare the bone-mass effects of an intermittent cyclic etidronate regimen with those of calcium supplementation in patients with established corticosteroid-induced osteoporosis. The results presented in this report suggest that the intermittent cyclic etidronate regimen is an effective treatment for established corticosteroid-induced osteoporosis.

PATIENTS AND METHODS

This prospective open-label study was undertaken at a private internal medicine and endocrinology/metabolism ambulatory clinic. Patients with established corticosteroid-induced osteoporosis were recruited from Rotterdam and the immediate environs during the 1-year period beginning 1 November 1989. Corticosteroid-induced osteoporosis was

defined as a vertebral bone mineral density two standard deviations or more below mean peak bone mass for young normal subjects, i.e., T score ≤ 2.0 SD. The patient dosing and observation period began 1 December 1989 and concluded 30 November 1991.

To be eligible for study, patients must have been receiving chronic uninterrupted corticosteroid therapy for at least 1 year or have received a cumulative prednisone dose > 4.0 gram, equivalent to at least 1 year of prednisone 10 mg/d. Patients were also in stable condition with regard to the primary corticosteroid-treated disease and in general good health, with no other metabolic bone disease or insulin-dependent diabetes mellitus. Patients were excluded if, within the year preceding the study, they had received calcium supplements of greater than 1000 mg/d, pharmacological doses of vitamin D or one of its metabolites, estrogen, calcitonin, bisphosphonates, or fluoride. Patients were alternately assigned to treatment groups according to their study eligibility (including informed consent) and random appearance at the clinic. The procedures followed were in accord with guidelines of the Helsinki declaration for physician-initiated studies.
Patients continued prednisone therapy and received either calcium supplementation 500 mg/d as calcium citrate (Cacit® 500, Procter & Gamble Pharmaceuticals, Norwich, NY) or intermittent cyclic etidronate therapy (Didrokit®, Procter & Gamble Pharmaceuticals, Norwich, NY). The etidronate cyclic regimen (Didrokit) consisted of etidronate disodium 400 mg/d for 14 days, followed by calcium 500 mg/d (as calcium citrate) for 76 days; with a total of four repetitions of the cycle during the 12-month study.
A complete history and physical examination, routine haematology, and multiphase serum chemistry were performed at study entry. Serum calcium, phosphorus, and alkaline phosphatase were measured at baseline and at 6 and 12 months.
Bone mineral density (BMD) (g/cm^2) of the lumbar spine (expressed as the mean of the four vertebrae measured, L1-L4) and proximal femur (total hip, femoral neck, trochanter, and Ward's triangle) was measured by dual-energy x-ray absorptiometry (DXA; Hologic QDR1000, (Hologic Inc., Waltham MA) at baseline and at 6 and 12 months. Bone mass measurements were conducted at an independent clinical research organisation (Good Clinical Practice, Rotterdam, The Netherlands) whose staff was blinded to patient treatment. The coefficient of variation of BMD measurements was 0,50%.
Baseline demographic characteristics, expressed as mean ± standard deviation (SD), were tested for between-group interactions by analysis of variance. Vertebral and hip BMD measurements were displayed as mean ± standard error of the mean (SEM) for each treatment group. Normative BMD scores for each patient at each time point were calculated from the mean BMD and standard deviation for normal subjects provided by the DXA instrument manufacturer, with Z score representing the number of standard

deviations between a patient's BMD and the age-matched mean BMD and T score representing the number of standard deviations between a patient's BMD and the young normal mean BMD. Changes from baseline in BMD (g/cm^2 and percentage) were calculated for each patient at each time point. Mean ± SEM for Z scores, T scores (reported at baseline only), and changes from baseline were then determined for each treatment group. Statistical comparisons within groups were performed by one-sample Student's t-tests and between groups, by two-sample Student's t-tests using a statistical package for personal computers (Minitab, Release 5.1, Minitab, Inc., State College, PA). Ninety-five percent confidence intervals were calculated by a two-sample t procedure. Statistical significance was set at $p < 0.05$.

RESULTS

Patient Characteristics

The initial study population comprised 41 older adults. Two patients dropped out of the study before the 6-month visit, one in the calcium group because of the discontinuation of prednisone therapy and one in the etidronate group because of epigastric distress related to the study drug; baseline data for these two patients are excluded. This report includes data only on patients who completed the study.

Thirty-nine patients with a mean age of 64 years (range 38 to 88) completed the study, 20 in the calcium group and 19 in the etidronate group. Nineteen (90%) of the 21 women enrolled in the study were postmenopausal, with a mean of 17.5 years since menopause. The mean duration of corticosteroid (prednisone) therapy was 6.0 years with a mean cumulative dose of 29.3 g. At the start of the study, the mean daily prednisone dose was 10.8 mg. Mean (± SD) spine and proximal femur (total hip) BMD Z scores for the completed study population were -2.5 ± 0.12 and -1.9 ± 0.1, respectively, and mean T scores were -3.7 ± 0.12 and -3.1 ± 0.13, respectively.

The baseline characteristics of the completed study population are presented by treatment group in Table I. At baseline, the two treatment groups were comparable in age, height, weight, ratio of female to male patients, duration of exposure to and cumulative dose of prednisone therapy before study entry, and spine and proximal femur (total hip) BMD. The diseases for which the study patients were receiving prolonged prednisone therapy are shown in Table II. Twenty-two of the 39 patients in the study had pulmonary diseases (chronic asthma; chronic obstructive pulmonary disease). The treatment groups were comparable in the types of disease and the number of patients in both the pulmonary and non-pulmonary disease categories.

TABLE I

Demographic Characteristics, Prednisone Exposure, and Bone Mineral Density of Patients at Baseline

	Calcium (N = 20)	Etidronate (N = 19)
Demographic characteristics		
Age (y)	64.6 ± 13.4	62.3 ±14.5
Height (cm)	165 ± 7.5	168 ±10.5
Weight (kg)	68.9 ± 10.4	73.3 ±13.3
Female:male ratio	13:7	8:11
Prednisone exposure		
Duration of exposure (y)	6.1 ± 4.8	5.9 ±2.5
Cumulative dose (g)	31.9 ± 26.5	26.6 ±9.8
Bone mineral density		
Spine (L1-L4)		
g/cm²	0.656±0.111	0.656±0.142
Z-score*	-2.3 ± 0.69	-2.7 ±0.84
T-score*	-3.7 ± 0.94	-3.8 ±1.16
Percent of young normal mean	61.7 ± 10.0	61.0 ±12.3
Proximal femur (total hip)		
g/cm²	0.647±0.099	0.609±0.120
Z-score*	-1.7 ± 0.70	-2.2 ±0.65
T-score*	-2.9 ± 0.83	-3.4 ±0.76
Percent of young Normal mean	63.2 ± 9.6	59.2 ±9.0

Data reported as mean ± standard deviation unless otherwise indicated.
*Z-score, bone mineral density normalised to age-matched mean; T-score, bone mineral density normalised to young normal mean.

TABLE II

Diseases Requiring Chronic Corticosteroid Treatment in Study Population

Disease Category	Calcium	Etidronate	Total
Pulmonary*	12	10	22
Non-pulmonary			
Gastrointestinal†	5	5	10
Cardiovascular‡	2	2	4
Other§	1	2	3
Total	20	19	39

Data reported as number of patients.
*Asthma, chronic obstructive pulmonary disease.
†Crohn's disease, chronic hepatitis, ulcerative colitis.
‡Giant cell arteritis.
§Hemolytic anemia, polychondritis, thrombocytopenia.

TABLE III
Changes in Bone Mineral Density of the Spine and Proximal Femur (Total Hip) After Treatment With Calcium or Intermittent Cyclic Etidronate

	N	Baseline (g/cm²)	Absolute Values (g/cm²)		Percent Change from Baseline	
			6 Months	12 Months	6 Months	12 Months
Spine						
Calcium	20	0.656 ± 0.025	0.642 ± 0.023*	0.633 ± 0.023*	-2.0 ± 0.6*	-3.4 ± 0.7*
Etidronate	19	0.656 ± 0.032	0.672 ± 0.031*†	0.689 ± 0.030*†	2.7 ± 0.6*†	5.7 ± 1.2*†
Proximal femur (total hip)						
Calcium	20	0.647 ± 0.022	0.637 ± 0.022*	0.620 ± 0.021*	-1.5 ± 0.6*	-4.1 ± 0.7*
Etidronate	19	0.609 ± 0.028	0.635 ± 0.027*†	0.648 ± 0.027*†	4.6 ± 1.0*†	6.8 ± 0.4*†

Data reported as mean ± standard error of the mean.
*P <0.02 compared with baseline.
†P <0.001 compared with calcium group.

TABLE IV
Changes in Bone Mineral Density of the Spine and Proximal Femur (Total Hip) After Treatment With Calcium or Intermittent Cyclic Etidronate: Characterisation According to Sex

	Calcium			Etidronate		
		% Δ from Baseline			% Δ from Baseline	
Skeletal Site	Baseline (g/cm²) (N)	6 Months	12 Months	Baseline (g/cm²) (N)	6 Months	12 Months
Spine						
Female	0.620 ± 0.030 (13)*	-2.5 ± 0.8†	-3.6 ± 0.9†	0.545 ± 0.018 (8)*	3.9 ± 1.2†‡	8.1 ± 1.8†‡
Male	0.722 ± 0.035 (7)	-1.1 ± 0.5§	-3.2 ± 1.1†	0.732 ± 0.040 (11)	1.9 ± 0.6†‡	3.9 ± 1.4†‡
Proximal femur (total hip)						
Female	0.638 ± 0.033 (13)	-1.6 ± 0.8	4.6 ± 0.9†	0.494 ± 0.019 (8)*	6.1 ± 1.2†‡	8.7 ± 2.6†‡
Male	0.665 ± 0.015 (7)	-1.5 ± 0.7	-3.3 ± 1.0†	0.693 ± 0.023 (11)	3.5 ± 1.3†‡	5.4 ± 1.4†‡

Data reported as mean ± standard error of the mean.
* P <0.05 compared with men.
† P <0.02 compared with baseline.
‡ P <0.01 compared with calcium group.
§ P <0.05 compared with baseline.

TABLE V
Changes in Bone Mineral Density of the Spine and Proximal Femur (Total Hip) After Treatment With Calcium or Intermittent Cyclic Etidronate: Characterisation According to Disease Category

	Calcium			Etidronate		
		% Δ from Baseline			% Δ from Baseline	
Skeletal Site	Baseline (g/cm²) (N)	6 Months	12 Months	Baseline (g/cm²) (N)	6 Months	12 Months
Spine						
Pulmonary	0.633 ± 0.025 (12)	-2.0 ± 0.6*	-3.6 ± 0.8*	0.653 ± 0.041 (10)	1.5 ± 0.4*†	3.8 ± 1.4*†
on-pulmonary	0.690 ± 0.490 (8)	-2.1 ± 1.2	-3.3 ± 1.4‡	0.659 ± 0.054 (9)	4.1 ± 1.1*†	7.7 ± 1.8*†
Proximal femur (total hip)						
Pulmonary	0.618 ± 0.024 (12)	-1.2 ± 0.8*	-3.5 ± 0.9*	0.575 ± 0.033 (10)	6.2 ± 1.5*†	9.2 ± 2.1*†
Non-pulmonary	0.692 ± 0.038 (8)	-2.0 ± 0.8‡	-5.1 ± 0.8*	0.647 ± 0.044 (9)	2.9 ± 0.9*†	4.1 ± 1.4*†

Data reported as mean ± standard error of the mean.
* P <0.02 compared with baseline.
† P <0.01 compared with calcium group.
‡ P <0.05 compared with baseline.

The subgroups of women and men differed significantly with respect to height (mean ± SD, 162.5 ± 7.4 cm versus 171.3 ± 8.9 cm; $p < 0.01$), weight (67.3 ± 8.3 kg versus 75.4 ± 14.2 kg ; $p < 0.05$), baseline BMD (0.592 ± 0.10 g/cm^2 versus 0.731 ± 0.12 g/cm^2; $p < 0.01$) and T score (- 4.14 ± 0.87 versus - 3.27 ± 1.04; $p < 0.01$) of the spine, and baseline BMD of the proximal femur (total hip) (0.583 ± 0.12 g/cm^2 versus 0.682 ± 0.07 g/cm^2; $p < 0.01$). They were comparable with respect to all other characteristics (data not shown).

The subgroups of patients with pulmonary disease and those with non-pulmonary disease were significantly different with respect to duration of exposure to prednisone (mean ± SD, 7.1 ± 4.0 years versus 4.6 ± 2.9 years, respectively; $p < 0.03$) and cumulative prednisone dose (35.0 ± 22.7 g versus 22.0 ± 13.6 g; $p < 0.03$). They were comparable with respect to all other characteristics (data not shown).

Bone Mineral Density

Treatment with intermittent cyclic etidronate regimen for 12 months resulted in significant increases of 5.7% and 6.8% from baseline in BMD of the spine and proximal femur (total hip), respectively ($p < 0.02$). Calcium supplementation alone did not prevent significant corticosteroid-induced losses of 3.4% and 4.1% from baseline in BMD of those respective sites ($p < 0.02$) (Table III; Figure 1). The changes from baseline in the etidronate group differed significantly from those in the calcium group after both 6 and 12 months of study ($p < 0.001$). After 12 months, the difference between the groups was 9.1% (95% CI 6.3% to 11.9%; $p < 0.01$) at the spine and 10.9% (95% CI 7.8% to 14.1%; $p < 0.01$) at the proximal femur (total hip).

The changes from baseline in Z scores of the spine and proximal femur (total hip) were comparable within each treatment group after 12 months (mean ± SD, etidronate group: 0.34 ± 0.06 and 0.34 ± 0.06, respectively; calcium group: - 0.17 ± 0.04 and - 0.19 ± 0.04, respectively) (Figure 2). The mean changes in Z score of the spine and proximal femur (total hip, femoral neck, trochanter, and Ward's triangle) regions were significantly different between treatment groups ($p < 0.01$). The changes in BMD Z score of these skeletal sites were uniformly positive in the etidronate group and uniformly negative in the calcium group.

A significantly higher proportion of patients in the intermittent cyclic etidronate group had increases from baseline in BMD of the spine and proximal femur (total hip) after 12 months of treatment ($p < 0.01$) (Figure 3). Seventeen (89%) of 19 etidronate-treated patients had increases in BMD of both skeletal sites, whereas only 2 (10%) and 3 (15%) of the calcium-treated patients experienced positive changes at the spine and proximal femur (total hip), respectively.

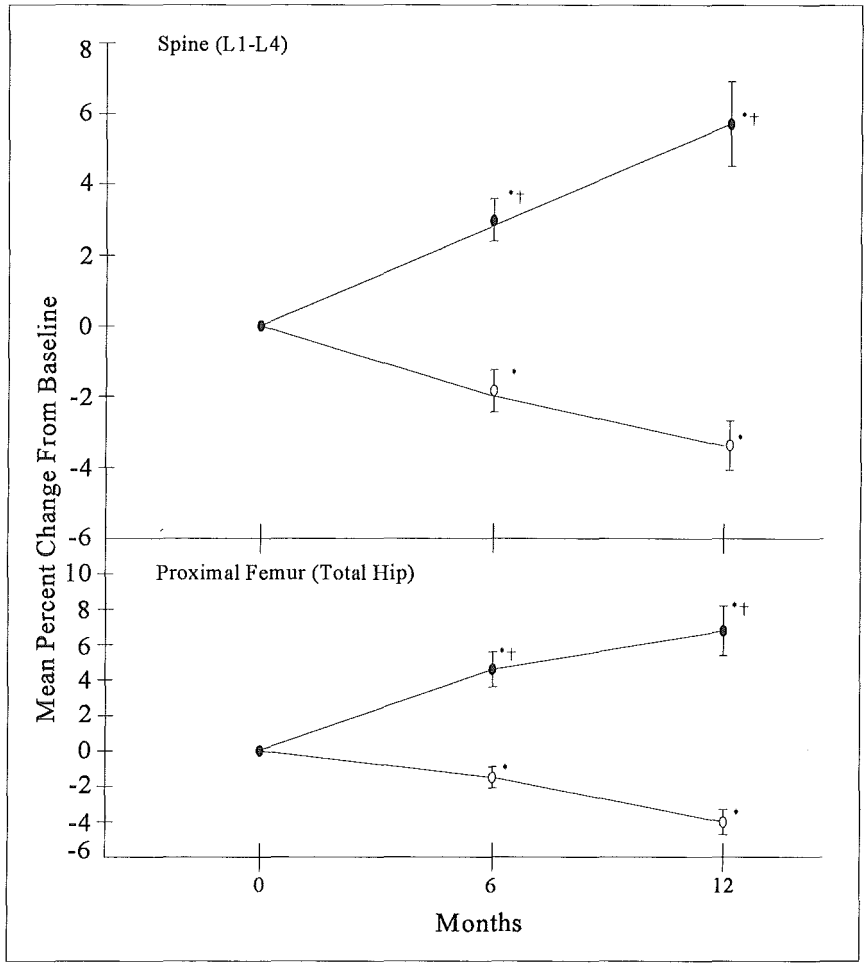


Figure 1. Mean (± SEM) percent change from baseline in bone mineral density of the spine (L1_L4) and proximal femur (total hip) during 12 months of treatment with calcium (n = 20; open circles) or intermittent cyclic etidronate (n = 19; solid circles). * $p < 0.02$ compared with baseline; † $p < 0.001$ compared with calcium group.

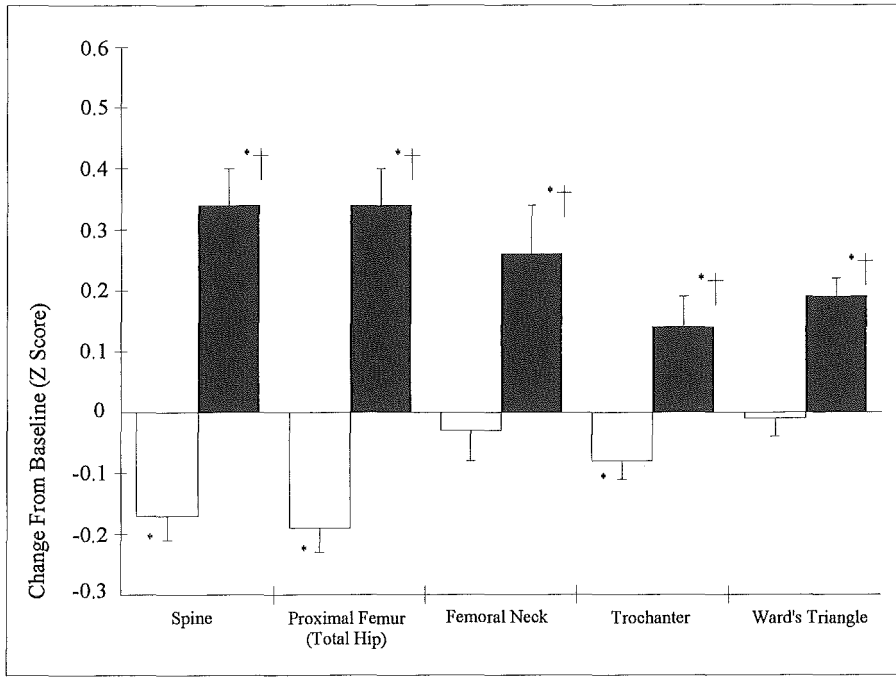


Figure 2. Mean (± SEM) in Z score from baseline in bone mineral density of the spine (L1_L4) and proximal femur (total hip, femoral neck, trochanter, and Ward's triangle) after 12 months of treatment with calcium (n = 20; open bars) or intermittent cyclic etidronate (n = 19; solid bars). * $p < 0.01$ compared with baseline; † $p < 0.01$ compared with calcium group.

Analyses of response by subgroups (female/male, pulmonary/non-pulmonary) showed no significant attribute-dependent changes. Women and men did not differ significantly in the extent of corticosteroid-induced bone loss or the response to etidronate treatment during the study period (Table IV). Not surprisingly, women had significantly lower baseline BMD of the spine than men in both treatment groups ($p < 0.05$); baseline BMD of the proximal femur (total hip) was significantly lower in women in the etidronate group

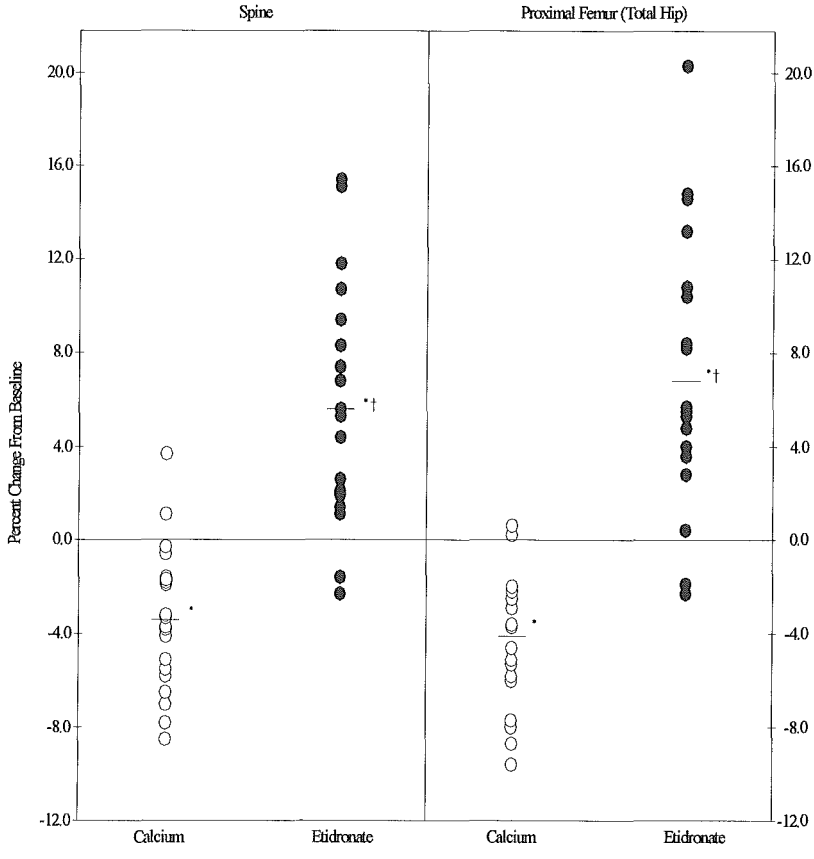


Figure 3. Percent change from baseline in bone mineral density of the spine (L1_L4) and proximal femur (total hip) in individual patients after 12 months of treatment with calcium (n = 20; open circles) or intermittent cyclic etidronate (n = 19; solid circles). Horizontal bars mark mean values. * $p < 0.02$ compared with baseline; † $p < 0.001$ compared with calcium group.

($p < 0.01$). Corticosteroid-treated patients with pulmonary disease and those with non-pulmonary disease also had comparable responses to calcium supplementation (losses in BMD) and to etidronate treatment (increases in BMD) at the spine and proximal femur (total hip) (Table V).

Serum calcium and phosphorus and haematological indices remained stable in both groups during the study period (data not shown). Serum alkaline phosphatase, a clinical-

ly useful marker of bone remodelling, did not change significantly after 6 and 12 months of treatment with calcium (mean change: 0.2% and 0.9%, respectively) or etidronate (0.1% and - 3.9%, respectively).

Side Effects

Both treatment regimens were well tolerated. There were no untoward reactions, unexpected side effects, or toxicity attributable to either treatment. One patient in the etidronate group dropped out of the study because of epigastric distress. No clinically apparent interactions were observed between corticosteroid treatment on the primary disease and the study medications.

DISCUSSION

The results of this study demonstrate the ability of intermittent cyclic etidronate to reverse progressive bone loss in the spine and proximal femur in patients with established corticosteroid-induced osteoporosis. In the entire population and the subgroups studied (women and men, pulmonary disease and non-pulmonary disease), intermittent cyclic etidronate therapy for 12 months produced significant increases in BMD of the spine and proximal femur (total hip), as compared both with baseline and with the calcium group ($p < 0.02$). The gains in bone mass in the etidronate group were similar at the two skeletal sites, with mean changes of about 6 to 7% in BMD and mean changes of + 0.34 in BMD Z score.

The effects of etidronate on BMD of the spine in our study are comparable to those reported for other bone antiresorptive agents tested for prevention of bone loss in early corticosteroid treatment [8,11] and established corticosteroid-induced osteoporosis associated with chronic treatment [9,13]. However, the positive effects of intermittent cyclic etidronate on BMD of the proximal femur (total hip) are, to our knowledge, the first reported for a therapeutic agent at this clinically important skeletal region. A recent clinical study in which treatment with calcitriol alone or in combination with calcitonin was concurrently initiated with corticosteroid therapy reported effectiveness in the prevention of bone loss in the lumbar spine but not in the femoral neck [23]. A recent review of clinical studies of the treatment of corticosteroid-induced osteoporosis or early bone loss [1] included no data for bone mass of the proximal femur, most likely because the studies were conducted when instruments for the accurate assessment of bone mass of the hip were not available.

The similarity we observed in the degree of change in BMD Z score at the spine and

proximal femur (total hip) after 12 months of treatment has important clinical implications. The inverse relationship between bone mass and the risk of spinal and hip fractures in osteoporosis is well recognised [23-26]. Although this relationship can predict hip fractures from BMD measurements at most skeletal sites [25,26], Cummings et al. [26] found the bone mass of the proximal femur to be the strongest predictor of hip fractures. A decrease in BMD of the proximal femur (total hip, femoral neck, trochanter, or Ward's triangle) by one standard deviation increases the relative age-adjusted risks of hip fractures by about threefold. In epidemiological studies, Ross et al. have also estimated similar increases in the risk of spinal fracture for each standard deviation (age-matched) decrease in BMD of the spine [23]. Maintenance of bone mass at these two sites is therefore clinically prudent, especially in postmenopausal women and other patients at risk for primary osteoporosis for whom chronic moderate- to high-dose corticosteroid therapy is ongoing or planned. In our study, the group of patients who received only calcium supplementation had pre-existing compromised bone mass of the spine and proximal femur (total hip), with Z scores of - 2.3 SD and - 1.7 SD, respectively (Table I), and subsequently lost bone mass equivalent to an additional - 0.17 and - 0.19 standard deviations during the 12-month study period. The group of patients who received intermittent cyclic etidronate treatment had comparably low bone mass of the spine and proximal femur (total hip) at baseline, with Z scores of - 2.7 SD and - 2.2 SD, respectively (Table I), but in contrast gained bone mass equivalent to + 0.34 standard deviation at both skeletal sites. The resulting difference between the calcium and etidronate groups in BMD of the spine and total hip at the end of the study was approximately half of a standard deviation (i.e., a 0.5 SD change in Z score). The relationship between changes in bone mass (expressed as increments of standard deviation) and the risk of fracture is exponential [23]. Thus, the potential for reductions in fracture risks as a result of these effects of etidronate treatment could be substantial and clinically significant.

Corticosteroids have been previously reported to have different effects on the bone mass of different skeletal regions, that is, the axial skeleton (e.g., lumbar spine), which is predominantly trabecular or cancellous bone, and the appendicular or peripheral skeleton (e.g., radius or forearm, hip), which is predominantly cortical bone [3,28-30]. However, more recently, Sambrook et al. [27] observed comparable losses in bone mineral density of the spine and hip (femoral neck, trochanter, and Ward's triangle) in patients receiving either acute or chronic corticosteroid therapy. Our results are consistent with those findings, with comparable corticosteroid-induced bone loss occurring in the spine and proximal femur (total hip) in the calcium-treated group after 12 months of study (Figure 2).

Corticosteroid-induced bone loss has also been reported to occur at a higher rate and to be more substantial during the first 6 to12 months of corticosteroid therapy as compared

with subsequent losses, which occur at a much slower rate and may be negligible [2,5]. In another study from our clinic [18], postmenopausal women with temporal arteritis experienced a loss of 5.0% in bone mineral density of the spine in the first year of prednisone therapy (mean daily dose: 10.7 mg). In the current study, long-term prednisone therapy (range of duration: 1-18 years) had already resulted in moderately advanced bone loss in our patients, as evidenced by the mean BMD Z scores for the spine (- 2.5 SD) and proximal femur (total hip) (- 1.9 SD). However, we continued to observe relatively high rates of bone loss in the calcium-treated patients, with mean changes of - 3.4% in BMD of the spine and - 4.1% in BMD of the proximal femur during the 12-month study (Table III). The results of both studies confirm that chronic, continuous corticosteroid administration of 10 mg/day or more substantially affects skeletal bone mass. This underscores the need to maintain chronic corticosteroid doses at the lowest levels necessary for therapeutic effects, since corticosteroid-induced bone loss has been reported to be less likely at daily doses of less than 7.5 mg [1]. However, in patients whose clinical conditions require chronic corticosteroid therapy at daily doses of at least 10 mg of prednisone (or its equivalent), therapeutic intervention to prevent or attenuate bone loss should be considered.

In our study, intermittent cyclic etidronate reversed losses in bone mass of the spine and proximal femur in a majority of female and male patients with established osteoporosis secondary to chronic corticosteroid therapy for pulmonary and non-pulmonary diseases. Calcium supplementation alone did not prevent or attenuate the sustained bone loss at the spine and proximal femur that was associated with long-term continuous corticosteroid therapy. Controlled, blinded studies are needed to confirm the encouraging results of this study of intermittent cyclic etidronate in the treatment of corticosteroid-induced osteoporosis.

ACKNOWLEDGEMENTS

The authors wish to thank Hilary J. Dijkstra, M.D., William Y.W. Au, M.D., and P.J. (Cilla) Davis, E.L.S., of Procter & Gamble Pharmaceuticals, for assistance in the collection and analysis of data and the preparation of the manuscript.

REFERENCES

1. Lukert, BP, Raisz LG. Glucocorticoid-induced osteoporosis: pathogenesis and management. Ann Intern Med 1990; 112: 352-64.
2. Silverman SL. Management of corticosteroid-induced osteoporosis: a clinician's perspective [editorial]. Calcif Tissue Int 1992; 50: 101-3.
3. Nordin BEC, Marshall DH, Francis RM Crilly RG. The effect of sex steroid and corticosteroid hormones on bone. J Steroid Biochem 1981; 15: 171-174.
4. Seeman E, Wahner HW, Offord KP, Kumar R, Johnson WJ, Riggs BL. Differential effects of endocrine dysfunction on the axial and the appendicular skeleton. J Clin Invest 1982; 69: 1302-9.
5. LoCascio V, Bonucci E, Imbimbo B, et al. Bone loss in response to long-term glucocorticoid therapy. Bone Miner 1990; 8: 39-51.
6. Reid IR, Ibbertson HK. Calcium supplements in the prevention of steroid-induced osteoporosis. Am J Clin Nutr 1986; 44: 287-90.
7. Hahn TJ, Halstead LR, Teitelbaum SL, Hahn BH. Altered mineral metabolism in glucocorticoid-induced osteopenia. Effect of 25-hydroxyvitamin D administration. J Clin Invest 1979; 64: 655-65.
8. Sambrook P, Birmingham J, Kelly P, et al. Prevention of corticosteroid osteoporosis. A comparison of calcium, calcitriol, and calcitonin. New Engl J Med 1993; 328: 1747-52.
9. Lukert BP, Johnson BE, Robinson RG. Estrogen and progesterone replacement therapy reduces glucocorticoid-induced bone loss. J Bone Miner Res 1992; 7: 1063-9.
10. Ringe J-D, Welzel D. Salmon calcitonin in the therapy of corticoid-induced osteoporosis. Eur J Clin Pharmacol 1987; 33: 35-9.
11. Montemurro L, Schiraldi G, Fraioli P, Tosi G, Riboldi A, Rizzato G. Prevention of corticosteroid-induced osteoporosis with salmon calcitonin in sarcoid patients. Calcif Tissue Int 1991; 49: 71-6.
12. Reid IR, King AR, Alexander CJ, Ibbertson HK. Prevention of steroid-induced osteoporosis with (3-amino-l-hydroxypropylidine)-1,1-bisphosphonate (APD). Lancet 1988; 1: 143-6.
13. Reid IR, Heap SW, King AR, Ibbertson HK. Two-year follow-up of bisphosphonate (APD) treatment in steroid osteoporosis [letter]. Lancet 1988; ii: 1144.
14. Meunier PJ, Briancon D, Chavassieux P, et al. Treatment with fluoride: bone histomorphometric findings. In: Christiansen C, Johansen JS, Riis BJ, editors. Osteoporosis 1987. Proceedings of the International Symposium on Osteoporosis; 1987 Sept 27-Oct 2; Aalborg (Denmark). Volume 2. Copenhagen (Denmark): Osteopress; 1987: 824-8.
15. Storm T, Thamsborg G, Steiniche T, Genant HK, Sørensen OH. Effect of intermittent cyclical etidronate therapy on bone mass and fracture rate in women with postmenopausal osteoporosis.

New Engl J Med 1990; 322: 1265-71
16. Watts NB, Harris ST, Genant HK, et al. Intermittent cyclical etidronate treatment of post menopausal osteoporosis. New Engl J Med 1990; 323: 73-9.
17. Harris ST, Watts NB, Jackson RD, et al. Four-year study of intermittent cyclic etidronate treatment of postmenopausal osteoporosis: three years of blinded therapy followed by one year of open therapy. Am J Med 1993; 95:557-67.
18. Mulder H, Struys A. Intermittent cyclical etidronate in the prevention of corticosteroid-induced bone loss. Br J Rheumatol 1994; 33:348-50.
19. Lane NE, Genant HK, Kinney JH, Engleman E. Effect of intermittent cyclic etidronate (ICT) therapy for glucocorticoid-induced osteoporosis in rheumatoid arthritis (RA): interim analysis. J Bone Miner Res 1993; 8(Suppl. 1):S262 (#583).
20. Miller PD, Hamilos DL, McIntyre DO, et al. Cyclical etidronate (CE) increases bone mass in patients with established steroid-induced osteoporosis (SIOP). J Bone Miner Res 1993; 8(Suppl. 1):S338 (#885).
21. Bensen WG, Bianchi F, Cividino A, et al. Intermittent cyclic etidronate therapy (ICT) prevents corticosteroid-induced bone loss. In: Christiansen C, ed. Proceedings of the Fourth International Symposium on Osteoporosis and Consensus Development Conference, Hong Kong, March 27-April 2, 1993. Rødovre, Denmark: The Fourth International Symposium on Osteoporosis, 1993:118 (#434).
22. Gallacher S, Anderson K, Speekenbrink T, Bryden F, Banham SW, Boyle IT. A densitometric assessment of the value of cyclical oral etidronate and intermittent intravenous pamidronate in corticosteroid-dependent lung disease. In: Christiansen C, ed. Proceedings of the Fourth International Symposium on Osteoporosis and Consensus Development Conference, Hong Kong, March 27-April 2, 1993. Rødovre, Denmark: The Fourth International Symposium on Osteoporosis, 1993:111 (#405).
23. Ross PD, Davis JW, Vogel JM, Wasnich RD. A critical review of bone mass and the risk of fractures in osteoporosis. Calcif Tissue Int 1990; 46: 149-61.
24. Melton LJ III, Kan SH, Frye MA, Wahner HW, O'Fallon WM, Riggs BL. Epidemiology of vertebral fractures in women. Am J Epidemiol 1989; 129: 1000-11.
25. Hui SL, Slemenda CW, Johnston CC. Baseline measurement of bone mass predicts fracture in white women. Ann Intern Med 1989; 111: 355-61.
26. Cummings SR, Black DM, Nevitt MC, et al. Bone density at various sites for prediction of hip fractures. Lancet 1993; 341: 72-5.
27. Sambrook P, Birmingham J, Kempler S, et al. Corticosteroid effects on proximal femur bone loss. J Bone Miner Res 1990; 5: 1211-6.
28. Adinoff AD, Hollister JR. Steroid-induced fractures and bone loss in patients with asthma. N Engl J Med 1983; 309: 265-8.
29. Schaadt O, Bohr H. Bone mineral in lumbar spine, femoral neck and femoral shaft measured by dual photon absorptiometry with 153-gadolineum in prednisone treatment. Adv Exp Med Biol 1984; 171: 201-8.
30. Dykman TR, Gluck OS, Murphy WA, Hahn TJ, Hahn BH. Evaluation of factors associated with glucocorticoid-induced osteopenia in patients with rheumatic diseases. Arthritis Rheum 1985:28: 361-8.

PART 5

General Discussion

GENERAL DISCUSSION

I. NEW HYPOTHESIS AND ANIMAL MODELS IN THE PATHOPHYSIOLOGY OF GLUCOCORTICOID-INDUCED OSTEOPOROSIS

Bone loss is one of the major side effects of glucocorticoid treatment. Histologically glucocorticoid-induced osteoporosis is characterised by a decreased bone formation rate, decreased wall thickness of trabeculae and increased bone resorption (see chapter 1). Increased bone resorption, decreased osteoblasts proliferation and biosynthetic activity, sex hormone deficiency, as well as hyperparathyroidism resulting from decreased intestinal calcium absorption and hypercalciuria due to altered vitamin D metabolism, all have been proposed as mechanisms for the loss of bone that ensues with glucocorticoid excess [1]. However, none of these mechanisms provides a satisfactory explanation for the cardinal histological features of glucocorticoid-induced osteoporosis and the evidence to support these mechanisms appeared to be weak and conflicting (see chapter 1). Therefore continuing research is necessarily to clarify the pathophysiological mechanisms of glucocorticoid-induced osteoporosis. Weinstein et al recently presented new insights in the pathofysiology of glucocorticoid-induced osteoporosis by using the mouse as a model. They claimed that the mouse, unlike the rat and other previously examined laboratory animals, is a faithful animal model of glucocorticoid-induced bone loss in humans. They reported that, after only 7 days of treatment, the same histomorphometric changes, seen in biopsies from patients receiving long-term glucocorticoid therapy, could be observed in mice [2]. They suggest that decreased production of osteoclasts can explain the reduction in bone turnover with chronic glucocorticoid excess, whereas decreased production and apoptosis of osteoblasts can explain the decline in bone formation and trabecular width. According to these authors, increased apoptosis of osteocytes is also involved in the pathofysiology of glucocorticoid-induced osteoporosis. These cells embedded in the bone matrix are suggested to play a role in the detection of micro-damage and the subsequent transmission of the signal that leads to repair by remodelling [3]. This animal

model could be useful in further research, studying new therapies like anabolic agents (PTH) and/or inhibitors of bone resorption.

II. BONE MINERAL MEASUREMENTS: WHAT TO DO?

In the prevention study we measured only lumbar spine BMD. We now know that DXA measurement both at the lumbar spine and femoral neck are indicated in glucocorticoid treated patients. Especially in our first study, we should have performed scans of the femoral neck because measurements of the lumbar spine may be unreliable due to age-related degenerative changes like osteophytes, osteochondrosis, decreased disk space and aortic calcifications.

BMD measured by DXA can be expressed in different ways. In all studies we expressed our results as an areal density (g/cm^2). For comparison with age- and sex-matched controls we also used Z-scores to underscore the fact that we regard glucocorticoid-induced osteoporosis as a secondary form of osteoporosis. The use of T-score results in an overestimation of the effect of age, especially in elderly subjects.

During the period our studies were performed, the field of bone densitometry has developed rapidly and many non-invasive techniques are currently available for clinical use. Especially three techniques are now widely used: DXA, QCT, QUS.

The DXA technique makes it possible to quantify the amount of bone in the lumbar spine, proximal femur, forearm or entire skeleton with minimum radiation exposure and with high degree of accuracy and precision. However, as discussed earlier, with DXA bone mass is expressed as an areal density. Therefore collapse of one or more vertebral bodies in the region of interest can lead to falsely increased BMD values. Furthermore it is also impossible to differentiate between cortical and trabecular bone. Also osteophytes and aortic calcifications could lead to a false increase in BMD. To avoid this problem other scanning positions have been proposed. Using lateral scanning of the lumbar spine, the posterior processes of the vertebral body are excluded from the measurement, with the potential advantage that it provides a better insight in changes in trabecular bone [4]. However lateral scans of the vertebral bodies, particularly in the decubites position, are significantly less precise than AP scans. Furthermore the window for this technique is limited in case of deformities of especially lumbar vertebrae.
QCT is also a precise and accurate method of measuring bone mass, with the major

advantage that a volumetric density is measured instead of an areal density. Another advantage, is the possibility to differentiate between cortical and trabecular bone. A relatively disadvantage is the high radiation exposure, its lower availability and the impossibility to measure hip BMD.
QUS is a more recent development. It variously examines the velocity, attenuation or reflection of ultrasound. The interest in its use is that it does not involve ionising radiation and may give some information concerning the structural organisation of bone in addition to bone mass. However, this technique needs additional study both with respect to the diagnosis and follow-up of treatment of patients with osteoporosis.

The frequency of fractures in glucocorticoid-treated subjects suggests that physicians should be more aware of the devastating effects of glucocorticoids on the skeleton. Bone mineral density measurements provide an opportunity to quantify this effect. At present mostly T-scores are used to express BMD results. However it is important to realise that both T- and Z-scores reflects different forms of information which might effect the ultimate actions taken in patient with glucocorticoid-induced osteoporosis. With respect to glucocorticoid-induced osteoporosis it remains questionable whether the T-score is the appropriate way to express and define this kind of osteoporosis. Given the fact that glucocorticoid-induced osteoporosis is a secondary form of osteoporosis a Z score seems more appropriate. Of course interpretation can be combined with T-scores, to get insight in the age-related component of osteoporosis.
Currently, it is still difficult to identify the individuals at risk. The use of the computer program, as described in chapter 5, might provide a tool to identify subjects on long-term maintenance treatment with glucocorticoids. This instrument could also be useful to recruit populations to study in more detail the relationship between BMD and fracture in glucocorticoid-treated patients.

Taken together, the fact that glucocorticoids mostly effect trabecular bone has consequences for the interpretation of BMD results. Furthermore, when interpreting the changes in bone one need to distinguish between effects due to the disease itself and those that are caused by glucocorticoid treatment. Another aspect of interest are potential differences in the response of QUS versus BMD parameters in subjects with glucocorticoid-induced osteoporosis. Since QUS parameters and BMD might provide different forms of information (chapter 2), combination of these techniques could be a valuable option for the future. Further studies on this area have to be awaited and till then QUS should not be used in daily clinical practice to diagnose and follow-up patients with glucocorticoid-induced osteoporosis.

In conclusion:
- DXA of lumbar spine and hip are still the best way to quantify glucocorticoid-induced osteoporosis
- A combination of T- and Z- scores should be used in expressing BMD results
- Whether a Z score below –1 SD and T-score below –1.5 SD are real cut-off points for treatment needs further study.
- QUS should only be used as an experimental option in glucocorticoid-induced osteoporosis

III. INHALED CORTICOSTEROIDS: FROM GOOD TO BAD?

Over the past years, glucocorticoids have become established as a cornerstone of treatment of asthmatic patients, because of their demonstrated clinical efficacy. However the indications and duration of use are not so well defined. It is clear that glucocorticoids should be given in the lowest possible dose. This is not only the case in oral glucocorticoids but also in case inhaled glucocorticoids are used. The studies on the effects of inhaled glucocorticoids on BMD are not conclusive. In our own study we reported a decrease in BMD. This was expected because different studies reported a decrease in markers of bone formation (chapter 3). However, not every patient using inhaled glucocorticoids is at risk. Grassi et al divided them in three groups:
- "Low risk", inhaled budesonide less than 800 microgram a day
- "Moderate risk", inhaled budesonide more than 800 microgram a day
- "High risk", inhaled glucocorticoids combined with courses of oral glucocorticoids more than four times a year.

Till now the absence of knowledge on the relation between duration, dose and device of inhaled glucocorticoids used and fracture risk must lead to a restrictive use of these drugs. Nevertheless the effects of inhaled glucocorticoids on bone mass in asthmatic patients are much smaller compared to oral glucocorticoids. Therefore we should use them if indicated, for a short period of time, at the lowest possible dose and with instructions to prevent ingestion. Studies combining data about, bone mass, fracture risk, dose and duration of inhaled glucocorticoids, device used and underlying disease are urgently needed. By adjusting our computer program for patients taking inhaled glucocorticoids, also these patients could be identified.

IV. TREATMENT OF GLUCOCORTICOID-INDUCED OSTEOPOROSIS

Calcium, vitamin D and its metabolites, thiazide diuretics, fluoride, estrogen, testosterone, medroxyprogesterone, nandrolone decanoate, growth hormone, deflazacort, calcitonin and bisphosphonates have all been proposed as therapy for glucocorticoid-induced osteoporosis. Based on our data concerning BMD and the more recent data about fracture risk, bisphosphonates are the only therapeutic options with "proven" fracture reduction and must, therefore considered as the first choice in the prevention and treatment of glucocorticoid-induced osteoporosis. Are other therapeutic options become worthless?
Especially in glucocorticoid-induced osteoporosis, vitamin D induced calcium absorption is decreased due to the mechanisms described in chapter 1. Data reported by Koster et al showed that calcium or vitamin D deficiency decreased the efficacy of a bisphosphonate [5]. Therefore, calcium and vitamin D are a prerequisite for any therapy concerning osteoporosis. If we take a closer look at the studies that reported fracture reduction, it appeared that the control and treatment group both received calcium and vitamin D supplements. So the first line of this paragraph could better be: "Combined therapy of bisphosphonates with vitamin D and calcium can be considered as the first choice in prevention and treatment of glucocorticoid-induced osteoporosis".

Combination of different treatment modalities and target specific glucocorticoids are future treatment options that needs further study. Given the decreased bone formation and the increased resorption in glucocorticoid-induced osteoporosis a combination of a stimulator of bone formation with an inhibitor of bone resorption seems to be a logical therapy. A potential stimulator of bone formation is PTH. Our own data suggested that the combination of magnesium with etidronate, in women with low serum magnesium, increased bone mass more than etidronate alone. [6]. This extra gain in bone mass can be explained by the increase in PTH when magnesium is added in those patients. A recent study with PTH and HRT showed spectacular results on BMD [7]. From other studies it appeared that prevention of osteoblast and osteocyte apoptosis is the principle mechanism for the anabolic effect of PTH on bone [8,9]. Therefore, it seems that PTH and, perhaps in the future PTH mimetics represent a rational therapy especially in glucocorticoid-induced osteoporosis. The reduction in fracture rate has to be awaited, as an increase in BMD will not always reflect a decrease in fracture risk, as shown in the case of fluoride [10,11]. Also the longterm safety of PTH is still unclear. However the combination of bone formation stimulators with anti-resorptive compounds like bisphosphonates is certainly an option which deserves further study.
From the recent studies with risedronate and alendronate the numbers needed to treat

have been calculated. It was suggested that less than 10 percent needed to be treated for one year to prevent one vertebral fracture. If this is true this makes bisphosphonates a potential cost-efective treatment of CIOP [12].

HRT still have a prominent place in all guidelines. However, the question remains if HRT has the same beneficial effects in glucocorticoid-induced osteoporosis as it has in early postmenopausal osteoporosis. The efficacy and safety is still disputable and no evidence on fracture reduction is provided (see chapter 4). Therefore the main indication for using HRT in CIOP should be limited to proven hypogonadism. Adler [13] stated that all patients with glucocorticoid-induced hypogonadism should be considered for sex hormone replacement therapy. He advises to measure serum oestradiol in premenopausal women and testosterone in men, to determine which patients are functionally hypogonadal. Nevertheless, HRT can be considered in the first years after menopause as second choice after bisphosphonates.

Deflazacort –an oxazoline derivate of prednisone- is discussed in chapter 4 and seems not to be a well-validated option in the reduction of glucocorticoid-induced bone loss. Recently new "designer glucocorticoids" have been developed that exhibit anti-inflammatory activity *in vivo* as potently as classical glucocorticoids, without requiring glucocorticoid receptor-DNA binding and transactivation which could be the signal for cell apoptosis [14,15]. Further studies on these drugs have to be awaited.

V. SUGGESTIONS FOR FURTHER RESEARCH

In this dissertation we have presented data, about the use of etidronate in the prevention and treatment of glucocorticoid induced osteoporosis. Meanwhile these results have been confirmed by others using other bisphosphonates and also data have been obtained which support fracture reduction. The question "Who and When to Treat" remains prominent. Therefore, prospective studies are needed to identify better tools to select those subjects at highest risk for glucocorticoid-induced osteoporosis. Furthermore, we have to put more effort in the design of model systems to understand the pathophysiological mechanisms underlying glucocorticoid-induced osteoporosis. In this respect the mouse model developed by Weinstein et al is promising and certainly deserves further study. Only the exact description of factors involved in the development of glucocorticoid-induced osteoporosis will provide the insights necessary to design new strategies to prevent and treat this devastating disease.

REFERENCES

1. Reid IR, Glucocorticoid-induced osteoporosis: Assessment and treatment. J Clin Densitometry, 1998. 1(1): p. 65-73.
2. Weinstein RS, Jilka RL, Parfitt AM, and Manolagas SC, Inhibition of osteoblastogenesis and promotion of apoptosis of osteoblasts and osteoclasts by glucocorticoids: potential mechanisms of their deleteroius efects on bone. J Clin Invest, 1998. 102: p. 274-282.
3. Manologas SC and Weinstein RS, New developments in the pathogenesis and treatment of steroid-induced osteoporosis. J Bone Miner Res, 1999. 14: p. 1061-1066.
4. Reid IR, Evans MC, and Stapleton J, Lateral spine densitometry is a more sensitive indicator of glucocorticoid-induced bone loss. J Bone Miner Res, 1992. 7: p. 1221-1225.
5. Koster JC, Hackeng WHL, and Mulder H, Diminished effect of etidronate in vitamin D deficient osteopenic postmenopausal women. Eur J Clin Pharmacol, 1996. 51(2): p. 145-147.
6. Struijs A and M. H, Treatment of postmenopausal osteoporosis and low serum magnesium with intermittent cyclical EHDP and magnesium. Osteoporosis Int, 1996. 6(Suppl 1): p. PTu 680.
7. Lane NE, Sanchez S, Modin GW, Genant HK, Pierini E, and Arnaud C, Parathyroid hormoner treatment can reverse corticosteroid-induced osteoporosis. J Clin Invest, 1998. 102: p. 1627-1633.
8. Jilka RL, Weinstein RS, Roberson P, Shelton R, Swain F, Smith C, Kirchner j, Parfitt AM, and Manolagas SC, PTH increases bone formation by postponing osteoblast apoptosis, not by increasing precursor proliferation. Bone, 1998. 23: p. S 519.
9. Bellido T, Plotkin L, Han L, Manolagas SC, and Jilka RL, PTH prevents gucocorticoid-induced apoptosis of osteoblasts and osteoclasts in vitro: Direct interference with a private death pathway upstream from caspase-3. Bone, 1998. 23: p. S518.
10. Riggs BL, Hodgson SF, and O'Fallon WM, Effects of Fluor treatment on the fracture rate in postmenopausal women with osteoporosis. New Engl J Med, 1990. 322: p. 802-809.
11. Kleerekoper M, Peterson EL, Nelson DA, Phillips E, Schork MA, Tilley BC, and Parfitt AM, A randomised trail of sodium fluoride treatment for postmenopausal osteoporosis. Osteoporosis Int, 1991. 1: p. 155-161.
12. Adami S and di Valeggio COC, New oral bisphosphonates in the prevention and treatment of corticosteroid-induced osteoporosis. Osteoporosis Int, 1999. 9: p. S18.
13. Adler RA and Clifford JR, Glucocorticoids and osteoporosis. Edocrinol Metab Clin North Am, 1994. 23: p. 641-654.
14. Reichardt HM, Kaestner KH, Tuckerman J, Kretz O, et al., DNA binding of the glucocorticoid receptor is not essential for survival. Cell, 1998. 93: p. 531-541.

15. Vayssiere BM, Dupont S, Choquart A, Petit F, Garcia T, Marchadeau C, Gronemeyer H, and Resche-Rigon M, Synthetic glucocorticoidsv that dissociate transactivation and AP1 transrepression exhibit anti-inflammatory activity *in vivo*. Mol Endocrinol, 1997. 11: p. 1245-1255.

PART 6

Summary

(English and Dutch)

Samenvatting

(Engels en Nederlands)

SUMMARY

Osteoporosis is a serious adverse event caused by the use of glucocorticoids. The impact of fractures of hip and vertebral body on the individual is superimposed on the already compromised quality of life due to the underlying disease for which glucocorticoid treatment was initiated. Also the cost of health care generated by these patients and the loss of economical productivity should be a challenge for the medical profession to find not only a solution for intervention strategies but also to create a signalling system to identify the patient at highest risk.
The impact of glucocorticoids on bone and the consequences for the patient are often underestimated or not recognised by the medical profession. Research by and education of, this medical profession has to be done to provide a safety net for those who needs glucocorticoids for their daily living. This thesis provides a compact overview about pathofysiology, treatment, past, present and future research in the field of glucocorticoid-induced osteoporosis.

Chapter 1 focuses on the pathofysiology of glucocorticoid-induced osteoporosis. It provides an overview of the literature on the effects of glucocorticoids at the cellular and skeletal level and describes the different pathophysiological mechanisms involved.

Chapter 2 describes the risk of fracture in patients treated with glucocorticoids. The assessment of risk is complicated because it consists out of many, sometimes independent components. The division in skeletal and non-skeletal components is the most logical one, however interactions between both components is complex and makes it difficult to clearly define prediction rules. BMD plays an important role in the skeletal component of fracture risk and the different methods of BMD measurements are discussed.

Chapter 3 gives an overview about the impact of inhaled glucocorticoids on calcium and bone homeostasis. The literature and the data provided appeared to be inhomogeneous and difficult to interpret. Inhaled glucocorticoids appeared to have less systemic adverse

events but in higher dosages it still has to be regarded as a risk factor for developing steroid-induced osteoporosis.

Chapter 4 gives an overview of the literature about the different treatment modalities and their rationale. It appeared that the combination of vitamin D, calcium and bisphosphonates is the only proven combination to reduce fractures in glucocorticoid-induced osteoporosis.

Chapter 5 describes the results of a search, using population based databases of general pharmacists, by a special designed computer program.
Twenty-two large population based databases from general pharmacists, randomised throughout the Netherlands were screened. The investigated population consisted of 248,169 people. This population consisted out of 157.461 subjects with a public insurance and 90.708 subjects with a private insurance. The age-specific prevalence of long-term prednisone use, was calculated in the population with a public insurance. An age-related increase of prednisone use was observed from 0.015% at the age of 0-10 yrs up to the highest prevalence (1.4%) in the age-group 80-89 yrs (overall prevalence of 0.27%). Nine-hundred sixty-four patients (532 women and 432 men, aged 4-98 years) receiving long-term prednisone (7.5 mg prednisone daily for $\geq$ 3 months), and thereby at risk for developing steroid induced osteoporosis, were identified. Concomitant data about prescription of drugs used for prevention and treatment of osteoporosis were obtained. Of these 964 patients, 351 (36%) were taking some form of anti-osteoporotic medication.
Our computer program appeared to be an effective signalling system in identifying patients at risk for steroid induced osteoporosis.

Chapter 6 describes the effects on bone mass and biochemical bone markers, in 33 prospectively studied patients with chronic obstructive pulmonary disease treated during one year with 800 µg/day inhaled beclomethasone, 800 µg/day inhaled budesonide or not requiring steroids. Both inhaled corticosteroids decreased serum concentrations of the formation markers osteocalcin and carboxy terminal propeptide of type I collagen (PICP). The resorption marker cross-linked carboxyterminal telopeptide of type I collagen (ICTP) increased significantly more in patients treated with beclomethasone compared to those treated with budesonide. The decrease in bone mineral density (BMD) was more pronounced in patients treated with beclomethasone (1.1% in the spine; 1.7% in the hip; $p < 0.05$) compared to those treated with budesonide (0.6% in both spine and hip) or in the control group. Taken together, inhaled corticosteroids in the dosages used appeared to affect both biochemical markers of bone turnover and bone mineral density.

Chapter 7 describes the acute short-term effects on biochemical parameters of calcium and bone homeostasis in fourteen postmenopausal women treated with high dose of prednisone (60mg/day) alone or with additional etidronate. Before and during 5 days of treatment, serum calcium, phosphorus, creatinine, alkaline phosphatase activity, osteocalcin, carboxy terminal propeptide of type I procollagen (PICP), cross-linked carboxy terminal telopeptide of type I collagen (ICTP), PTH, 25 hydroxy vitamin D and urinary excretion of calcium over 24 hours were measured. Significant differences from baseline were found in osteocalcin and urinary excretion of calcium in both groups and for ICTP in only one group. Significant differences between groups were calculated at day 5 of the study for osteocalcin, ICTP and 24 hours of urine calcium excretion. In the group additionally treated with etidronate, osteocalcin increased and urinary calcium excretion and ICTP decreased. In the prednisone alone treated group, osteocalcin decreased and urinary calcium increased.

Chapter 8 describes the results of a randomised clinical trial studying the preventive effects of etidronate on corticosteroid-induced bone loss in postmenopausal women with temporal arteritis treated with high dose of prednisone (mean dose 11mg/day). At 3, 6 and 12 months, vertebral BMD was significantly ($p < 0.01$) increased in the etidronate treated group and decreased in the group without etidronate , based on mean actual and percent changes from baseline in BMD and mean changes from baseline in BMD Z-score. Between-group comparisons were also significant ($p < 0.002$) at each time point. At 12 months, the mean percent change from baseline in BMD was $+1.42 \pm 0.45\%$ in the etidronate treated group and $-4.95 \pm 0.64\%$ in the group without etidronate. Nine of the 10 patients treated with etidronate and prednisone showed an increase in bone mass, whereas all 10 patients treated with only prednisone showed a decrease of BMD.
At 12 months, the change from baseline in serum alkaline phosphatase was $6.2 \pm 3.9\%$ in the etidronate treated group and $-5.3 \pm 3.0\%$ in the group without etidronate ($p < 0.05$, between groups). No clinically significant changes in routine clinical and laboratory evaluations were noted. Etidronate was well tolerated, and no adverse events related to etidronate treatment were reported.

Chapter 9 describes the results of a randomised clinical trial studying the effect of calcium supplementation and intermittent cyclic etidronate on BMD in patients with established corticosteroid-induced osteoporosis. Patients who had established corticosteroid-induced osteoporosis and received chronic prednisone therapy (> 10 mg/d) were enrolled in a prospective 12-month, open-label study. BMD of the spine (L1-L4) and proximal

femur (total hip, femoral neck, trochanter, and Ward's triangle) was measured by DEXA, at baseline, 6 months, and 12 months. Serum calcium, phosphorus, and alkaline phosphatase were also measured. Serum calcium, phosphorus, and alkaline phosphatase levels did not change significantly during the study in either treatment group. At baseline, women had significantly lower baseline BMD of the spine and proximal femur (total hip) and patients with pulmonary disease had significantly longer duration of prednisone therapy and cumulative prednisone dose. It appeared that, intermittent cyclic etidronate reversed the progressive loss of bone mineral density of the spine and proximal femur in female and male patients with established osteoporosis secondary to chronic corticosteroid (prednisone) therapy for pulmonary and non-pulmonary diseases. Calcium supplementation alone did not prevent or attenuate these corticosteroid-induced losses.

In part V of this thesis the main findings and clinical implications are discussed and further suggestions for research are given.

SAMENVATTING

Osteoporose is een ernstige bijwerking van het gebruik van glucocorticoiden. De gevolgen van heup en wervel fracturen beïnvloeden de kwaliteit van leven negatief. Een kwaliteit die al verminderd was ten gevolge van de oorspronkelijke ziekte waarvoor men glucocorticoiden moest gaan gebruiken. Ook de kosten van de medische behandelingen en het verlies aan productiviteit zouden een uitdaging moeten zijn, voor medici, om niet alleen behandelopties te bedenken, maar ook om strategieën te ontwerpen waarbij actief naar juist dit soort patiënten gezocht en gekeken kan worden zodat primaire preventie kan worden bedreven.
De gevolgen van het gebruik van glucocorticoiden voor de botten en de consequenties voor de patiënt worden vaak onderschat of worden nauwelijks herkend door medici. Onderzoek door en bijscholing van artsen, zal moeten leiden tot een veilig vangnet voor degenen die niet zonder glucocorticoiden kunnen. Dit proefschrift bevat een compact overzicht over de pathofysiologie, behandeling en research van glucocorticoid-geinduceerde osteoporose in het verleden, heden en toekomst .

In hoofdstuk 1 wordt een overzicht gegeven van de verschillende pathofysiologische mechanismen betrokken bij het ontstaan van glucocorticoid-geinduceerde osteoporose.

Hoofdstuk 2 beschrijft het fractuurrisico van patiënten die met glucocorticoiden behandeld worden. Het bepalen van het risico is erg moeilijk, omdat het is opgebouwd uit verschillende componenten die soms wel en soms ook niet met elkaar samenhangen. De verdeling in skelet gerelateerde en niet- skelet gerelateerde componenten lijkt een logische verdeling, echter de onderlinge afhankelijkheid van deze twee componenten, maakt het moeilijk om fractuurrisico te voorspellen. BMD speelt een belangrijke rol in de skelet gerelateerde zijde van de kans op fracturen. De verschillen tussen de BMD-meetmethoden, impliceren een zorgvuldige interpretatie van onderzoek gegevens gezien de gevaren voor onder- en overschatting. Deze worden in dit hoofdstuk besproken.

Hoofdstuk 3 geeft een overzicht van de literatuur over inhalatie steroïden en de effecten daarvan op de calcium- en botstofwisseling. De literatuur blijkt onvolledig en is door de zeer vele factoren die van invloed zijn op het bot moeilijk te interpreteren. In het algemeen kunnen we zeggen dat inhalatie steroïden minder systemische bijwerkingen hebben dan orale toedienings vormen. Echter in hogere doseringen vormen ze nog steeds een risico op glucocorticoid-geinduceerde osteoporose.

Hoofdstuk 4 geeft een overzicht van de potentiële behandelingsvormen die voor glucocorticoid-geinduceerde osteoporose beschikbaar zijn. Het blijkt dat er maar één combinatie is die daadwerkelijk fractuurreductie heeft aangetoond; dat is de combinatie: vitamine D, calcium en bisfosfonaten.

In hoofdstuk 5 worden de resultaten besproken van een screening van databases van apothekers m.b.v. een speciaal door ons ontwikkeld computerprogramma. Dit programma stelt ons in staat om de patiënten te selecteren die meer dan 7,5 mg prednison gebruiken gedurende drie maanden of langer. We hebben 22 grote databases kunnen screenen die random over Nederland verspreid waren. De onderzochte populatie bestond uit 248.169 mensen waarvan 157.461 personen ziekenfonds en 90.708 particulier verzekerd waren. Ook werden data verzameld over de andere medicatie van deze patiënten en dan met name medicamenten die gebruikt kunnen worden in de behandeling van osteoporose. De leeftijd specifieke prevalentie van langdurig, hoog corticosteroid gebruik kon worden berekend uit de populatie die ziekenfonds verzekerd was. De prevalentie verschilde van 0.015% in de groep van 0-10 jaar tot 1.4% in de groep van 80-89. De overall prevalentie was 0.27%. Van de onderzochte totale populatie bleken er 964 personen te zijn die meer dan 7,5 mg prednison gebruikten gedurende drie maanden of langer (532 vrouwen en 432 mannen). Van deze groep bleken 351 personen (36%) enige vorm van anti-osteoporotische medicatie voorgeschreven te krijgen.
Ons computerprogramma zou een effectief middel kunnen zijn om patiënten te identificeren die een verhoogd risico hebben op glucocorticoid-geinduceerde osteoporose.

Hoofdstuk 6 bevat de resultaten van een prospectieve pilotstudy van 33 patiënten met COPD behandeld met 800 µg beclomethason en budesonide per dag. De effecten van inhalatie steroïden op BMD en de markers van calcium en botstofwisseling worden beschreven. Het blijkt dat osteocalcin en PICP verlaagd worden door het gebruik van inhalatie steroïden. De marker die vooral de activiteit van osteoclasten aangeeft steeg significant meer in de groep die behandeld werd met beclomethason. Ook de BMD daalde in de met inhalatie steroïden behandelde groep. Het meest uitgesproken was de daling

in de groep behandeld met beclomethason (1.1% in de wervelkolom en 1.7% in de heup). Het blijkt dat inhalatie steroïden zowel de BMD als de markers van calcium en botstofwisseling negatief beïnvloeden.

Hoofdstuk 7 beschrijft de resultaten van een studie over het acute effect van etidronaat op de parameters van calcium en botstofwisseling van veertien postmenopauzale vrouwen met arteritis temporalis die met hoge doses prednison behandeld worden. Gedurende vijf dagen werden serum calcium, fosfaat, creatinine, alkalische fosfatase, osteocalcin, PICP, ICTP, PTH, 25 hydroxy vitamine D en 24 uur urine calcium verzameld en gemeten. Significante veranderingen ten opzichte van de basale waarden, werden gevonden voor osteocalcin en urine calcium excretie in beide groepen en ICTP in één groep. Verschillen tussen twee groepen werden berekend op dag vijf en waren significant voor osteocalcin, ICTP en urine calcium excretie. In de groep die behandeld werd met etidronaat steeg de osteocalcin en daalde de urine calcium excretie en de ICTP. In de groep aan de behandeld met prednison zonder de toevoeging van etidronaat daalde de osteocalcin en steeg de urine calcium excretie.

Hoofdstuk 8 beschrijft de resultaten van een prospectief gerandomiseerde klinische studie betreffende het preventieve effect van etidronaat op de BMD van postmenopauzale vrouwen die vanwege arteritis temporalis starten met behandeling met hoge doses prednison (gemiddelde dosis 11mg/dag). In de groep die met etidronaat behandeld werd steeg de botmassa (+ 1,42%) en in de groep zonder etidronaat daalde de BMD (- 4,95%) significant. Negen van de tien patiënten behandeld met etidronaat toonden een stijging van de BMD terwijl bij alle patiënten in de controle groep een BMD verlies werd gevonden. Na 12 maanden veranderde het alkalische fosfatase in de behandelde groep met + 6, 2% en in de controle groep daalde de alkalische fosfatase met 5,3%. Er werden geen bijwerkingen van de behandeling met etidronaat gemeld.

Hoofdstuk 9 beschrijft de resultaten van een prospectief gerandomiseerde klinische studie betreffende het effect van etidronaat en calcium op reeds bestaande glucocorticoidgeinduceerde osteoporose. Gedurende 12 maanden werden patiënten gerekruteerd die chronisch meer dan 10 mg prednison per dag gebruikten. BMD van de heup en wervelkolom werden gemeten m.b.v. DEXA op tijdstip 0, 6 en 12 maanden. Serum calcium, fosfaat en alkalische fosfatase werden ook bepaald. Het bleek dat deze biochemische parameters gedurende de studie niet veranderden. Het bleek dat vrouwen een significant lagere BMD van de heup en de wervelkolom hadden. Tevens was opvallend, dat de patiënten die glucocorticoiden gebruikten vanwege COPD, gedurende een langere tijd en

hogere dosering gebruikten dan de rest van de populatie. Het bleek dat etidronaat een positief effect had op de BMD van patiënten met osteoporose ten gevolge van chronisch prednison gebruik ongeacht de indicatie. Calciumsupplementen konden verdere progressie van botverlies niet voorkomen.

In deel V van dit proefschrift worden de belangrijkste bevindingen besproken en in een klinisch perspectief geplaatst. Voorts worden aanbevelingen geformuleerd voor verder onderzoek.

DANKWOORD

Het schrijven van dit dankwoord vervult mij met ambivalente gedachtenkronkels. Allereerst is het een opluchting om een dankwoord te kunnen schrijven omdat dit betekent dat het proefschrift afgerond is. Anderzijds behoort het dankwoord, samen met het curriculum vitae, waarschijnlijk tot het meest gelezen gedeelte van een proefschrift, hetgeen eisen stelt aan de kwaliteit ervan. Degene die nu een prachtig epistel verwacht, raad ik nu af verder te lezen.

Het gezegde "Beter laat dan nooit" is er wel een die opgaat voor dit proefschrift. Al met al is het een proefschrift "oude stijl" geworden, zeker gezien de manier waarop het tot stand gekomen is. Daarbij denk ik allereerst aan mijn "werkplek". Al in een zeer pril stadium van de conceptie van dit proefschrift werd ik gedwongen te verhuizen van mijn werkkamer naar de hoek van de huiskamer in verband met uitbreiding van het gezin. Deze plek had voor- en nadelen, al werden de nadelen pas in de laatste fase van de uitdrijving van dit proefschrift pijnlijk duidelijk.

Lieve Nellie, zonder jouw persoonlijk motiverend vermogen en verzorgende eigenschappen zou dit proefschrift er niet zijn gekomen. Ook het wegcijferen van jouw persoonlijke wensen en verlangens vanwege dit proefschrift, had niet veel langer moeten duren. Dank voor je 'gedwongen' visites buitenshuis, zodat ik rustig kon werken; je fantastische planning waardoor de kinderen zo lang mogelijk op bed lagen of weg waren zodat ik in alle rust kon werken. Ik hoop me de komende tijd ook meer aan jouw planning te kunnen houden. Herbert, ik moest steeds aan je klaagzin denken: "Papa, het is niet eerlijk, u zit nu al weer achter de computer. U mag er de hele dag al achter zitten. Wanneer mag ik nu eens? Voorlopig altijd, jongen!!! Al is het maar omdat ik een nieuwe computer heb moeten kopen.

Beste promotoren, ook voor jullie is dit een apart proefschrift geworden. Ton, bedankt voor het initieel vertrouwen, je inspanningen en bijna vaderlijke bemoedigingen in de

laatste fase van dit proefschrift. De waardering voor alles wat je hebt gedaan en de manier waarop is enorm. Huib, bewaker van kwaliteit, zo zal ik je zeker herinneren. Dankzij jou is het proefschrift geworden tot wat het nu is. Dank, dat je ondanks het feit dat je pas in een laat stadium betrokken raakte, je de moeite getrooste, het proefschrift nauwgezet te corrigeren en suggesties te geven, zodat het eindresultaat er mag zijn. In de gezamenlijke bespreek sessies heb ik veel geleerd over het hoe en waarom met betrekking tot research en het schrijven van artikelen. Beste promotoren, bedankt dat ik met jullie heb mogen samenwerken. De kennis die ik door jullie heb verkregen zal ik in de toekomst kunnen gebruiken en hopelijk verder uitbreiden.

Beste Henk, een verlies voor het internistencorps. Onze eerste tijd in het IJsselland ziekenhuis zal ik nooit vergeten. Samen met Sten begonnen aan onze eerste baan als artsassistent. Ik heb geluk gehad dat ik jou als supervisor trof. Je manier van opleiden was onopvallend en efficiënt. Je gaf me het idee dat ik de afdeling "runde", hoewel in feite jij dat deed. Echter het zelfvertrouwen en de ervaringen die ik in die tijd heb opgedaan zijn van veel betekenis geweest voor mijn latere loopbaan. In die tijd kwam ik ook via jou voor het eerst in aanraking met osteoporose. Terloops werd opgemerkt:" Ik doe nog iets met osteoporose, hier heb je wat literatuur, als het je wat lijkt heb ik nog wel wat voor je te doen". Awel, dat heb ik geweten !! Ik heb er inderdaad aardig wat werk aan over gehouden, letterlijk en figuurlijk. Ondanks mijn overgewicht en de samenwerking met jou, ben ik toch in opleiding gekomen tot internist, ben ik artikelen gaan publiceren en heb ik delen van de wereld gezien, die ik anders nooit zou hebben aanschouwd. Ook heb ik voordrachten kunnen geven en ben ik vertrouwd geraakt met de commerciële zijde van het medisch bedrijf en het klinisch geneesmiddelenonderzoek.
Tevens ben jij de motor geweest achter de artikelen die van onze hand verschenen zijn. Je geduld met mij als het gaat over de snelheid, of beter gezegd traagheid, waarmee de artikelen geschreven werden, is opmerkelijk te noemen. Ook heb ik bewondering voor je onnavolgbare wijze van het documenteren en archiveren van de originele onderzoeksresultaten. Al met al kun je dit proefschrift zeker ook zien als een soort tweede promotie voor jezelf. De meeste studies zijn opgezet zonder hulp van buitenaf, zowel in financiële als in intellectuele zin. Dit heeft geleid tot hypotheses en studies ontsproten uit de dagelijkse poliklinische praktijk; resulterend in kleine pilotstudy's met echter een aardige impact. Terugkijkend was de studie opzet niet altijd even mooi en compleet, maar we hebben het naast onze dagelijkse bezigheden wel voor elkaar gekregen. Beste Henk, jij bent nu een belangrijke directeur van een goed lopende site management organisatie, die de laatste jaren explosief gegroeid is. Ondanks dat, ben je echter nog steeds de Henk Mulder die ik ken van vroeger. Wat mij betreft is er een basis gelegd voor een samen-

werking die ik graag in de toekomst zal voortzetten.

Terugdenkend aan mijn eerste fase van de opleiding tot internist in het St. Clara ziekenhuis dank ik Albert, dat je al in een vroeg stadium de AMVB hebt willen doorvoeren. De daaruit volgende compensatie na diensten en de lagere werkdruk op de afdeling, hebben mij in staat gesteld in een traag doch gestaag tempo voort te gaan. Hoewel ik me misschien tot vervelens toe sterk heb gemaakt voor de naleving van de regels, heeft de vrije tijd geleid tot het schrijven van wetenschappelijke artikelen, die jij ook meer dan eens kritisch hebt doorgelezen. Je adviezen betreffende onderzoek, commercialiseren van onderzoek en het aanvragen van patenten zijn in goede aarde gevallen. Je enthousiasme waarmee je je IC-idealen uitdroeg hebben bijgedragen aan mijn keuze tot intensivist. Ik hoop in de toekomst onze samenwerking nog voort te zetten op het gebied van IC, research en commercie.
Ook de bibliothecaresse van het Clara ben ik ontzettend dankbaar. In het Dijkzigt heb ik zelf moeten ervaren hoeveel tijd het kost om artikelen op te zoeken, aan te vragen en te kopiëren. Dankzij jou heb ik met een minimum aan tijd, de meest onmogelijke artikelen, uit de meest exotische tijdschriften kunnen krijgen. Deze hoeveelheid aan artikelen bewees zijn waarde tijdens het schrijven van de introductie. Zo'n bibliothecaresse is voor iedere arts, die naast zijn dagelijks werk ook nog onderzoek doet, onmisbaar.

Nu ik het woord introductie heb laten vallen, denk ik direct aan mijn voorlaatste werkgevers. De groep mensen die in het AZR de grootste moeite doen om een volwaardige IC opleiding te behouden, hebben mij stuk voor stuk de mogelijkheid gegeven om dit proefschrift af te ronden en in de huidige vorm te gieten. Met name professor Bruining en Djo Hasan en de groep ICP hartelijk dank.
Sten, ik zal je missen nu je vertrokken bent naar warmere oorden in het zuiden van Frankrijk. Het lijkt wel een vakantie adres. Ik weet wel beter. Bedankt voor de vriendschap, collegialiteit en goede tijd die we samen gehad hebben. Ook hebben we veel lol gehad in het schrijven van de artikelen.
Onno en Michiel, dank voor de introductie in jullie apothekerswereld, die geleid heeft tot een wederzijdse vruchtbare samenwerking.
Joop, bedankt voor de "mooie" plaatjes van de botten en je ondersteunende rol in het voltooien van dit proefschrift.
Ook al mijn collegae vanuit de Clara, waaronder Arthur, Ernst-Jan, Sten, Henriette, Peter, Ron, Cees en degene die ik natuurlijk weer vergeet, nog bedankt. Peter, jouw promotie komt nog. Met plezier zal ik ook de fouten in jouw proefschrift opsporen, zoals jij dat ook bij mij hebt gedaan.

Jan en André, van je schoonfamilie moet je het maar hebben. Hartelijk dank voor de energie die jullie hebben gestopt in het opnieuw maken van de grafieken en tabellen die door het failliet gaan van het AVC waren verdwenen. Jullie hulp was onmisbaar.

Verder wil ik een ieder bedanken die zijn of haar bijdrage heeft geleverd en die ik niet bij name heb genoemd. Het is geen onwil of desinteresse maar onmogelijk om een ieder die een bijdrage heeft geleverd persoonlijk te noemen en te bedanken.

Nellie, het proefschrift is klaar, ik ben internist, intensivist en nog steeds met je getrouwd. Ik hoop hiervan nog lang te kunnen genieten. Zo, en nu een; nee geen Bavaria!, eerst afvallen.

CURRICULUM VITAE

De auteur werd geboren op 8 augustus 1965 te Vlaardingen. In 1984 behaalde hij het VWO diploma aan de scholengemeenschap "Guido de Brès" te Rotterdam. In datzelfde jaar begon hij met de studie geneeskunde aan de Erasmus Universiteit te Rotterdam. In 1991 behaalde hij het artsexamen. Daarna is hij AGNIO interne geneeskunde geweest in achtereenvolgens het IJsselland Ziekenhuis, AZR "Dijkzigt" en het St. Clara Ziekenhuis. Op 1 april 1993 begon hij zijn opleiding tot internist op de afdeling interne geneeskunde van het St. Clara Ziekenhuis te Rotterdam (opleider: dr. A.F. Grootendorst). Op 1 september 1997 werd de opleiding voortgezet in het AZR "Dijkzigt" (opleider : Prof. dr. M.A.D.H. Schalekamp). Op 1 april 1999 werd hij ingeschreven in het specialistenregister. Gedurende het laatste jaar van de opleiding tot internist heeft hij de opleiding tot internist-intensivist gevolgd op de verschillende I.C. afdelingen van het AZR "Dijkzigt". Op 1 september 1999 werd hij geregistreerd als subspecialist. Vanaf 1 september is hij als staflid verbonden aan de afdeling intensive care Thoraxchirurgie van het AZR "Dijkzigt". Het onderzoek dat heeft geleid tot dit proefschrift werd uitgevoerd in het IJsselland Ziekenhuis en het AZR "Dijkzigt". Tevens is hij oprichter van de DRCO en de EMCR-IC waarvan hij tevens voorzitter is.

LIST OF ABBREVIATIONS

DXA=Dual energy X-ray Absorptiometry
BMD=Bone Mineral Density
SD=Standard Deviation
WHO=World Health Organisation
IGF=Insulin-like Growth Factor
PTH=Parathyroid Hormone
BMU=Basic Multi-cellular Units
BRU=Bone Remodelling Units
Ca=Calcium
25-(OH)D_3=25 hydroxyvitamin D
1,25-$(OH)_2D_3$=1,25 dihydroxyvitamin D
LH=Luteneizing Hormone
LHRH=Luteneizing Hormone-Releasing Hormone
ACTH=Adrenal CorticoTrophin Hormone
FSH=Follicle Stimulating Hormone
DHEA=DiHydroxyEpiAndrosterone
GC=GlucoCorticoid
QCT=Quantative Computer Tomography
FEA=Finite Element Analysis
HPA=Hypothalamic-Pituitary-Adrenal
Beclo=Beclomethasone
Budeso=Budesonide
PICP=carboxy terminal propeptide of type I procollagen
ICTP=cross-linked carboxy terminal telopeptide of type I collagen
N=Number of participants
GE=Gastro-Enteral
BMI=Body Mass Index
SPA=Single Photon Absorptiometry
DPA=Dual Photon Absorptiometry
℞=medication
RA=Rheumatoid Arthritis
IM=Intra-Muscular
P=Prospective
R=Randomised
C=Controlled
PlC=Placebo Controlled
DB=Double Blind
PMR=Polymyalgia Rheumatica
SLE=Systemic Lupus
PMP=Postmenopausal
HRT=Hormone Replacement Therapy