Cyclosporine and bone mass

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ABSTRACT

Calcium homeostasis depends upon the interplay of intestinal calcium absorption, renal excretion, and skeletal mobilisation or uptake of calcium, mediated though bone formation and resorption which are closely 'coupled' in the adult skeleton under normal circumstances. Cyclosporine leads to the uncoupling of formation and resorption, with overall high bone turnover resulting. The effect of cyclosporine is due to multiple effects on calcium metabolism and a knowledge of its mechanism of actions is necessary to understand its skeletal effects.

Introduction

The introduction of cyclosporine A (CyA) to transplantation immunosuppressive regimens in the 1980s played a pivotal role in reducing the frequency and severity of post-transplant rejection episodes, as well as permitting the use of lower doses of corticosteroids. CyA has also been widely used in the treatment of various immunological and rheumatic diseases, but generally in lower doses than that used after organ transplantation. CyA use in the transplant setting was subsequently recognised to be associated with reductions in bone mineral density (BMD) and an increased risk of fracture post transplantation (1-3). In contrast, its use in the rheumatic disease setting has generally not been associated with significant concerns about increased fracture risk. This review will examine current knowledge about CyA effects on bone.

Animal studies

A major problem in examining the effects of CyA in the clinical setting is that it is virtually always used in combination with other agents that also affect bone metabolism, such as corticosteroids. For this reason, animal studies have been helpful in sorting out its independent effects on bone. Such animal studies have shown that CyA generally results in increased bone turnover (4). Thus, in young rats CyA administration results in significant elevation of serum osteocalcin and 1,25 dihydroxyvitamin D levels and produces high turnover osteopenia, particularly in trabecular bone due to enhanced bone resorption outpacing bone formation. In these studies, bone loss was dependent upon both the dose and duration of CyA (4-5). The effect of CyA appears to be generally greater in young rats than in older rats (5), the latter having a lower rate of bone remodelling, but older rats are also at risk with higher doses (6). The effect is not seen with all cyclosporins. For example, cyclosporine H (CsH), a D-N-MeVal11 analog of CyA, is not immunosuppressive, and in contrast to CyA, it neither binds to cyclophilin nor alters cytokine activity. In rat models, CsH does not result in any significant increase in serum osteocalcin or serum 1,25(OH)2D levels (7), unlike CyA-treated rats. In rat models, CyA has also been shown to decrease serum testosterone, but its effects on bone appear not to be related to the degree of hypogonadism (8) and cannot be prevented by the concomitant administration of testosterone replacement (9).

The immunosuppressive action of CyA is thought to be primarily due to the inhibition of the antigen/mitogen-induced secretion of lymphokines at the transcriptional level from T cells. The inhibition of calcium-dependent signaling pathways by cyclophilin-cyclosporin complexes in T cells appears to shut down lymphokine-gene transcription. It is therefore of interest that animal studies suggest that T lymphocytes appear to be a prerequisite for the development of CyA-induced osteopenia. Thus Buchinsky *et al.* (10) observed that the administration of CyA to athymic nude rats and age-matched immunocompetent rats for 28 days resulted in high turnover bone loss in the latter group, whereas the nude rats were largely unaffected by the drug. Some drugs that have been used for the treatment of human osteoporosis have

also been examined in animal studies for their protective effects on CyA-induced bone loss. A study of alendronate treatment in young rats found that it decreased serum osteocalcin and largely prevented CyA adverse effects on bone, particularly with respect to the maintenance of trabecular bone volume, presumably by decreasing bone remodeling (11). CyA, when administered to oophorectomized rats, exacerbates the high turnover osteopenia associated with estrogen deficiency. Raloxifene, like estradiol, can prevent the high turnover osteopenia caused by oophorectomy. A study of raloxifene given to CyA-treated oophorectomized rats found that it was able to partially attenuate the skeletal effects of CyA (12).

Human studies

Most studies in humans of CyA have been conducted in organ transplant patients and, as such, are complicated by the need to account for the concomitant administration of other bone active agents, such as corticosteroids, as well as of metabolic bone disease associated with the underlying disease for which the transplant was performed. In renal transplant patients, some histomorphometric studies have shown increased bone turnover and the delayed repair of renal osteodystrophy (13), suggesting that the effect of CyA on bone turnover is independent of corticosteroids. In contrast, other studies after renal transplantation have reported the histomorphometric picture to be one of resolving secondary hyperparathyroidism which was considered to be more typical of corticosteroid osteoporosis (14). These differences may reflect variations in the dose and duration of CyA and corticosteroid therapy after transplant (15).

Secondary hyperparathyroidism appears to be common in the transplant situation. Elevated parathyroid hormone levels have been reported in some (2, 16-17) but not all studies of cardiac transplant bone loss (18). Guo *et al.* (17) found elevated serum osteocalcin, serum bone specific alkaline phosphatase and urinary N telopeptide of type I collagen levels in a cross-sectional study of 50 men after cardiac transplantation, consistent with increased turnover. Because serum PTH levels correlated positively with serum osteocalcin and serum creatinine, whereas serum creatinine correlated negatively with serum 1,25 dihydroxy vitamin D3 levels, the authors concluded that the bone loss was related primarily to corticosteroids. However, the increased remodelling was attributed to secondary hyperparathyroidism due to renal impairment from CyA therapy.

Others have also found relationships between markers of bone turnover and CyA use. Withold (19) measured a variety of markers after bone marrow transplantation and found that both the duration of CyA therapy and the time since transplantation were independent predictors of serum bone alkaline phosphatase values ($p < 0.01$), whereas the duration of CyA therapy was the only independent predictor of C-terminal propeptide serum concentrations ($p < 0.01$).

Although cyclosporin is generally used with other immunosuppressive therapy in most transplant situations, a number of studies have attempted to examine the effects of monotherapy in humans. Cuerto-Manzo *et al.* (20), in a study of renal transplant patients on average 127 months after transplant, found that CyA monotherapy treated subjects had a significantly lower appositional rate than patients treated with azathioprine plus prednisolone. Long-term renal transplant-patients treated with CyA monotherapy showed reduced BMD in both trabecular and cortical bone, but this reduction in BMD was not as severe as suggested by shorter term reports. CyA was associated with osteoclastic stimulation, osteoblast suppression and retardation of mineral apposition and bone formation rates (20).

Ebeling *et al.* (21) examined patients after allogeneic bone marrow transplantation and found that bone loss correlated best with the cumulative prednisolone dose at the spine and hip. Bone loss was also negatively related to the duration of CyA therapy for graft versus host disease and baseline deoxypyridinoline concentrations.

Aroldi *et al.* (22) studied 53 adults with first kidney transplants in a randomized trial to analyze the efficacy of three different immunosuppressive regimens: CyA alone (group 1), CyA plus steroids (group 2), and CyA plus steroids plus azathioprine (group 3). At 18 months, premenopausal transplant recipients showed a lesser decrease of lumbar bone mineral density than male transplant patients. In transplant recipients given CyA with corticosteroids, lumbar spine BMD decreased significantly, while it increased significantly in patients given CyA alone.

Osteoclast differentiating factor (ODF) is a type II transmembrane receptor TNF super family member, also known as RANKL (receptor activator of NF-KB ligand) or OPGL (OPG-ligand). ODF signalling has been implicated in the differentiation and function of dendritic cells, the antigen presenting cell of the immune system (23, 24). Osteoprotegerin (OPG) is a soluble TNF receptor family member that inhibits osteoclast differentiation and inhibits bone resorption by neutralising OPG-L.

A recent study has shown that CyA can decrease OPG mRNA in human marrow stromal cells (25) and this may be a further mechanism of CyA-induced bone loss. Since OPGL regulates T lymphocyte differentiation and its receptors are expressed on T lymphocytes (23,24), this is of particular interest given the animal studies referred to above suggesting CyA effects on bone are mediated via T lymphocytes (10).

A schematic diagram of summarising these possible mechanisms of CyA on bone is shown in Figure 1.

Most studies of CyA use in rheumatic disease, where doses generally do not exceed 5 mg/kg/day (26), have been reassuring with no reports of bone related toxicity. Recent trials of CyA effects on radiological progression in RA over 12 months reported reduction in erosions and joint damage with CyA with no reports of local or systemic osteoporosis being increased by CyA therapy (27, 28). In longer term follow-up studies out to 3 years, albeit in smaller numbers of subjects, no adverse events related to bony complications have been reported (29), although BMD was not measured in any of these studies. However, Ferriaccioli *et al.* (30) studied patients with early erosive, aggressive RA and a poor previous response to a 6-month course of methotrexate to assess the effect of adding

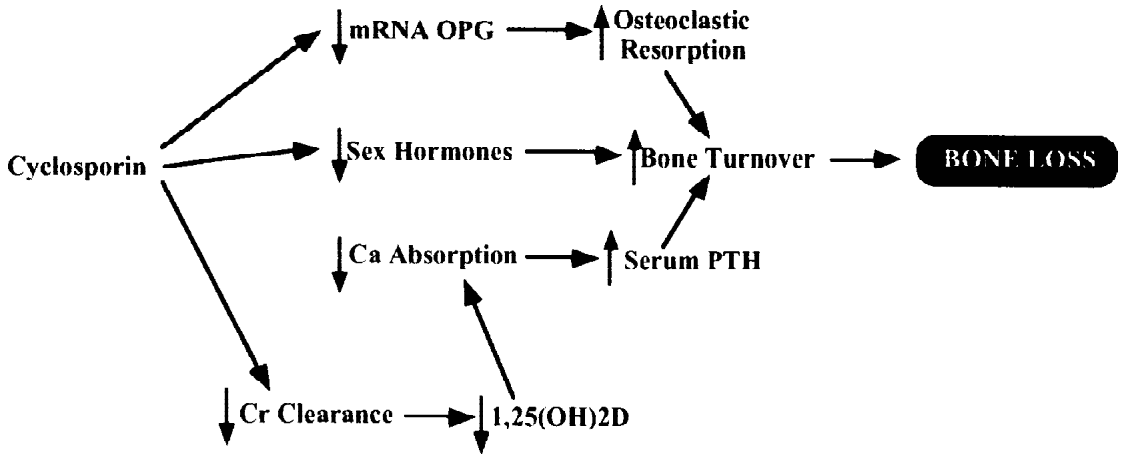


Fig. 1. Mechanisms of cyclosporine-induced bone loss.

CyA. An average BMD decline of 4% occurred in the first 6 months of methotrexate treatment, along with a significant decline in the serum levels of IGF-1 (-24.8%), DHEAS (-21.6%), and osteocalcin (-19.7%). After adding CyA in a dose of 3 mg/kg daily for 6 months, BMD increased by 3.9 %, serum IGF-1 by 42.4%, serum DHEAS by 34.2% and serum osteocalcin by +34.3%. These changes mirrored the clinical variables and acute phase reactants, which improved significantly.

In summary, CyA therapy has been shown to cause increased bone turnover with the uncoupling of resorption over formation in both animal models and human studies. This effect appears to be mediated by T cells and possibly the OPG/OPGL pathways. The effect is dose-dependent and, whilst resulting in an increased risk of fracture post transplantation, its use in the rheumatic disease setting at a dose < 5 mg/kg has generally not been associated with clinically significant adverse effects on bone.

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