Research article

Association between alendronate, serum alkaline phosphatase level, and heterotopic ossification in individuals with spinal cord injury

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Context/objective: Only sparse evidence exists regarding the effectiveness of oral alendronate (ALN) in the prevention of heterotopic ossification (HO) in patients with spinal cord injury (SCI). The objective of this study is to investigate the protective effect of oral ALN intake on the appearance of HO in patients with SCI.

Study design: Retrospective database review.

Setting: A Spinal Cord Unit at a Rehabilitation Hospital.

Participants: Two hundred and ninety-nine patients with SCI during acute inpatient rehabilitation.

Interventions: Administration of oral ALN.

Outcome measures: The incidence of HO during rehabilitation was compared between patients with SCI receiving oral ALN ($n = 125$) and patients with SCI not receiving oral ALN ($n = 174$). The association between HO and/or ALN intake with HO risk factors and biochemical markers of bone metabolism were also explored.

Results: HO developed in 19 male patients (6.35%), however there was no significant difference in the incidence of HO in patients receiving oral ALN or not. The mean odds ratio of not developing versus developing HO given ALN exposure was 0.8. Significant correlation was found between abnormal serum alkaline phosphatase (ALP) levels and HO appearance ($P < 0.001$) as well as normal serum ALP and ALN intake ($P < 0.05$).

Conclusion: Even though there was no direct prevention of HO in patients with SCI by oral ALN intake, abnormal serum ALP was found more frequently in patients with HO development and without oral ALN intake. This evidence could suggest that ALN may play a role in preventing HO, especially in patients with acute SCI with increasing levels of serum ALP.

Keywords: Alendronate, Heterotopic ossification, Spinal cord injury

Introduction

Heterotopic ossification (HO) is the pathological formation of mature lamellar bone in muscle and soft tissues adjacent to joints. Although the mechanism for development of HO is not fully understood, it is thought to be associated with local inflammation in the soft tissues that induces transformation of mesenchymal stem cells into osteoblasts by prostaglandins.¹ Osteoblasts produce tropocollagen and alkaline

phosphatase (ALP). Tropocollagen polymerizes to form collagen fibers which are involved in the formation of the bone matrix, while ALP inactivates pyrophosphate allowing for calcium deposition and mineralization of the bone matrix which leads to development of ectopic bone.¹

HO is a common complication following central nervous system disorders. The incidence of HO in patients with spinal cord injury (SCI) varies from 10 to 78%.^{1–3} It most frequently occurs 3–12 weeks after SCI,^{2,4} is predominantly found in the hip^{5–7} and always occurs below the level of paralysis.^{2,6} Risk

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factors for the development of HO after SCI include complete lesions, thoracic injuries, pneumonia, spasticity, urinary tract infection (UTI), the presence of tracheostomy, pneumonia, and trauma.^{8,9}

HO typically presents with a non-specific inflammatory reaction including swelling, heat, pain (in cases with intact sensation), and erythema.³ As HO progresses, it can result in reduced range of motion and, eventually, joint ankylosis.^{5,10} It can also cause skin breakdown and peripheral nerve entrapment. In individuals with SCI, HO may result in further functional impairments, significantly impede acute rehabilitation, and ultimately impair quality of life.^{5,10} Therefore, prevention is key.

Although there are several options for treatment of HO including radiotherapy, bisphosphonates (etidronate), and surgery, there are few current options that exist for the prevention of HO. A conservative prevention program should include patient education regarding the possible complication of HO and a consistent program to maintain joint range of motion. To date, only non-steroidal anti-inflammatory drugs (NSAIDs) have substantial evidence to support their use to prevent HO. It has been shown that, when given early, NSAIDs can reduce the development of HO.^{2,11} They are proposed to work by reducing inflammation and blocking prostaglandin synthesis.¹² Nevertheless, this medication class may affect fracture healing and spinal fusion, therefore limiting its use in the acute SCI population.^{13,14}

Alendronate (ALN) is a potent N-containing second generation bisphosphonate. At our institution, ALN has been prescribed to individuals early after SCI to limit disuse osteoporosis. ALN is thought to inhibit bone resorption and additionally has a small effect on mineralization.^{15,16} It has also empirically been observed to decrease the rate of formation of clinically significant HO, but evidence supporting the effectiveness of bisphosphonates in primary prevention of HO after SCI is still lacking.^{15,16} The purpose of our study was to further investigate the role of ALN in the prevention of HO after SCI.

Material and methods

This is a retrospective study of patients admitted with SCI (ASIA A, B, C, and D) at Spaulding Rehabilitation Hospital (SRH) from 2006 to 2012. ALN 70 mg in a tablet form (by Tagipharma Company, a generic pharmaceutical supplier, South Beloit, IL, USA) was prescribed weekly in a number of patients when the patient was able to receive oral medications and could tolerate sitting position for at

least one and half-hour. The medication was administered either as treatment for osteoporosis or as antiosteoporotic preventive measure in patients with AIS A, B, or C. Patients were included if they had full data available for at least 3 months from the time of their admission to the rehabilitation unit. Patients presenting with neuropathies (Guillain-Barré syndrome, multiple sclerosis, syndromes, etc.) or myopathies, without cord deficit or with non-acute SCI (more than 3 months from time of injury incident) were excluded.

Data collected included demographics (age, sex), date of injury, date of admission at SRH, duration of follow-up, type of injury (traumatic or non-traumatic SCI), level (cervical or thoracic-lumbar-sacral) and completeness (motor complete or not) of injury, concomitant traumatic brain injury (TBI) or not, spinal fusion or not, date of ALN start and duration, early or late (more than 3 months from injury) ALN start, known osteopenia or osteoporosis (by dual-energy X-ray absorptiometry (DEXA) measurement), NSAIDs intake, warfarin intake, antispasmodic medications intake, maximum abnormal laboratory findings of bone-muscle metabolism indices (values for serum ALP, serum creatine phosphokinase CPK, serum parathyroid hormone PTH). The normal laboratory values for ALP is 40–150 U/l, for CPK 22–198 U/l, and for PTH 10–55 pg/ml.

Our primary outcome measure was documentation of HO in the medical record based on radiographic examinations (always plain X-rays done at our hospital, rarely triple phase bone scan as this was an offsite exam with increased radiation for the patient). Clinical notes and therapist reports mentioning joint stiffness-contractures were documented separately from HO incidence as contracture incidence without radiographic abnormality. Secondary outcome measures included time of HO appearance from the time of injury, joint affected (hip, knee, shoulder, elbow, spine, hand, ankle), and type of HO treatment (surgery-radiation-medications).

Statistical analysis was performed with SPSS version 10.0 (SPSS, Inc., Chicago, IL, USA). Descriptive statistics were used to present data. Numerical data were presented as mean $\pm$ standard deviation (SD). Correlations between the appearance of HO and ALN intake or other parameters (demographic, clinical, laboratory) were calculated with χ^2 test and Fisher's exact test. The Mantel-Haenszel *t*-test was used to estimate the odds ratio of developing versus not developing HO given ALN exposure. Differences of mean values of ALP were calculated with independent samples *t*-test (separate-variance *t*-tests for equality of means). All statistical tests were conducted at a 0.05 significance level (P-value).

Results

From the initial file of 754 patients with SCI admitted to the rehabilitation hospital between 2006 and 2012, we initially included 570 patients (the rest were missing from the system or did not have all data needed to be included). Two hundred and ninety-nine patients with SCI who presented within 3 months from their deficit and with at least 3 months follow-up time were finally included in the study. The average follow-up time was 626.72 ± 620.49 days. The mean age of the patients was 42.7 ± 18.36 years while male subjects were 226 of total. Patients were admitted to the rehabilitation department an average of 21.31 ± 17.16 days following their injury. HO (with bone formation evident in radiographs) was found in 19 (6.35%) patients, mostly hip joint (13 patients) with the remaining in the knee, shoulder, and elbow joint. Type of treatment until the last follow-up included NSAIDs in all cases, surgery in three (15.8%) cases, and radiotherapy in two (15.2%) cases. The diagnosis of HO was made in 290.1 ± 379.5 days from the initial injury. Contracture (defined as limitation of range of motion without radiographic evidence of osseous transformation) was reported in 26 patients (8.7%), mainly in the ankle and the metacarpophalangeal-interphalangeal joints of the hand.

ALN was prescribed in 125 patients (41.8%) for an average 267.2 ± 405.09 days or 38.17 ± 57.89 weeks with mean cumulative dosing of 2672 ± 4050 mg. Seventy-one of those received the drug as early as 3 months from the SCI. Table 1 shows the characteristics of patients with and without ALN intake. Patients who received ALN were more frequently ($P < 0.05$) males

Table 2 Odds ratio of not developing versus developing HO given ALN exposure

	Patients with HO	Patients without HO	Odds ratio (95% confidence interval)
Patients with ALN intake	7	118	0.8 (0.3–2)
Patients without ALN intake	12	162	
Total	19	280	

who had traumatic, complete, cervical cord injuries, were not operated and were prescribed with antispasmodic medications. There was no significant correlation between the diagnosis of HO and oral ALN intake (Table 2). However, there were more patients receiving ALN who developed contractures ($P < 0.001$).

The mean maximum serum ALP measurement was 171.29 ± 169.69 U/l. There was a significant difference ($P < 0.01$) of mean serum ALP level between patients with HO (307.61 ± 486.29) and patients without HO (158.65 ± 93.59). From the Fisher's exact test, significant correlations ($P < 0.05$) were found between HO appearance and male sex as well as between HO appearance and serum ALP abnormality ($P < 0.001$). Furthermore, ALN intake (125 patients, 41.8%) and normal serum ALP (201 patients, 67.2%) were positively correlated ($P < 0.05$).

No significant correlations ($P > 0.05$) were found between the development of HO and age <50 years old, traumatic SCI, level of SCI, spinal surgical procedure, co-existence of TBI, the presence of osteoporosis or antispasmodic medications intake, and abnormal values of serum CPK or PTH.

Table 1 Characteristics of patients with SCI with and without ALN oral intake

	174 patients without oral ALN intake	125 patients with oral ALN intake	P-value
Age	44.2 (18)	40.6 (18.7)	>0.05
Time of ALN start (days from admission)	21 (18.6)	21.8 (15)	>0.05
Duration (in days) of ALN use		267.2 (458)	>0.05
Max ALP serum level	165.7 (198.6)	181.4 (99.1)	>0.05
Male sex	140 (80.4)	86 (68.8)	<0.05
Traumatic SCI	117 (67.2)	102 (81.6)	<0.01
Cervical SCI	77 (44.2)	79 (63.2)	<0.005
Motor complete SCI	64 (36.8)	89 (71.2)	<0.001
Surgery for SCI	45 (25.9)	19 (15.2)	<0.05
TBI	17 (9.8)	20 (16)	>0.05
Osteoporosis	4 (2.3)	7 (5.6)	>0.05
Spasticity necessitating antispasmodic medications	111 (63.8)	96 (76.8)	<0.05
Anticoagulation (warfarin intake)	24 (13.8)	21 (16.8)	>0.05
Abnormal serum ALP	49 (28.2)	49 (39.2)	<0.05
Abnormal serum creatinophosphokinase	7 (4)	11 (8.8)	>0.05
Abnormal serum parathormone	3 (1.7)	4 (3.2)	>0.05
Contracture development	7 (4)	19 (15.2)	<0.001

Values are presented in mean (SD) for continuous variables (top four variables) and *n* (%) for categorical variables (bottom variables). P-values in bold indicate statistical significance.

Discussion

Pharmacological therapeutic options available for management of HO¹⁷ include NSAIDs,^{2,12} warfarin, and bisphosphonates. Specifically, the NSAID, indomethacin,² and rofecoxib,¹¹ delay and/or prevent development of HO after SCI presumably by inhibiting prostaglandins and inflammatory mediators required for transformation of mesenchymal cell into osteoblasts. Notably, NSAID use in acute SCI may affect fracture healing and spinal fusion, thereby limiting its widespread use. Even though etidronate is the “state of the art”-Food and Drug Administration approved bisphosphonate for the treatment and prevention of HO, its antiosteoporotic effect is lower than ALNs since etidronate’s main effect is the inhibition of bone mineralization and has minimal effect on bone matrix formation.^{15,16} In our retrospective study of patients with SCI exploring risk factors for HO development and possible inhibitory effect of ALN, there was no statistically significant prevention of HO in patients receiving oral ALN as antiosteoporotic treatment. However, there seems to be an indirect correlation between oral ALN and HO prevention in patients with SCI given the finding that patients with HO had abnormal serum ALP levels and oral ALN intake was strongly correlated with normal serum ALP levels.

The high incidence of clinically significant HO in patients with SCI is widely reported.^{10,18} Risk factors for HO formation in patients with SCI have been established as, spasticity, pressure ulcers, completeness of SCI, thoracic lesions, pneumonia, UTIs, and the presence of tracheostomy.^{9,18} Risk factors for HO recurrence are considered cognition deficits, spasticity, and massive HO.¹⁹ In our retrospective study, only male sex was confirmed as risk factor for HO formation and there was no statistical significant correlation with age, spasticity, level of SCI, or motor complete injuries.

There are no specific symptoms or diagnostic tests to predict the population of patients with SCI that will develop HO. The typical clinical presentation is pain, warmth, erythema, local swelling, decreased range of motion, and increased spasticity. Sometimes patients with SCI do not have pain. Therefore, skeletal biomarkers could help early HO diagnosis.^{16,19} Elevated levels of serum ALP,^{18,20} CPK,²⁰ C-reactive protein,²¹ erythrocyte sedimentation rate,^{18,21} and prostaglandin E2⁷ are associated with HO after SCI. Elevated levels of serum CPK but not ALP is correlated with the severity of HO.²⁰ Some researchers support that stabilization of ALP is an indicator of HO maturation,¹⁹ while others do not think the same.²² However, these biomarkers are

non-specific. Except from HO formation, other osseous causes of serum ALP elevation in acute SCI are fracture or bone surgery. Non-osseous causes of ALP elevation are hepatic, renal, gastrointestinal, metabolic, and general skeletal disorders.^{23,24} In our study, it was confirmed that abnormal serum ALP was positively correlated to HO formation.

An observational retrospective study suggested that warfarin is associated with a reduction in the incidence of HO after SCI.⁴ Warfarin inhibits vitamin K-dependent glutamate carboxylation which is required for osteocalcin protein that is needed for bone matrix production. However, despite the evidence suggesting that warfarin may be an effective prophylaxis against HO,⁴ in some patients the risk of hemorrhage could outweigh its potential benefit, therefore limiting its use. In our series of 299 patients with SCI, the preventive effect of warfarin on HO formation was not seen.

Bisphosphonates inhibit the enzyme, farnesyl pyrophosphate synthase, which prevents mineralization of the bone matrix.²⁵ Etidronate, a bisphosphonate, has been shown to delay development and progression of HO in its early stages.^{15,26–28} Surgical excision followed by administration of etidronate^{29,30} or pamidronate⁶ prevents the recurrence of HO. Stover *et al.*^{28,29} in 1976 concluded that disodium etidronate offered protection of HO recurrence in four patients operated for HO resection. Similarly, Banovac,¹⁵ in a 1.5–6 years follow-up study of 40 patients with SCI who received intravenous etidronate for 3 days and continued with oral etidronate for 6 months totally, confirmed the absence of high-grade HO in these patients and interpreted it as the beneficial effect of etidronate compared. Aminobiphosphonates, such as ALN and risedronate, offer more antiresorptive osseous effect and minimal inhibition of mineralization. Animal studies though have confirmed that ALN mainly acts as osteoclast inhibitor rather than bone formation cell activator.³¹ For this reason, aminobiphosphonates are more appropriate for the prevention and treatment of osteoporosis.³² There are sparse reports though that ALN also has a beneficial effect in HO prevention.³³ In this study, we explored the additional benefit of HO prevention in patients with SCI receiving oral ALN for osteoporosis prevention. However, no statistically significant correlation was found.

ALN has been shown to improve bone mineral density in hip or spine in elderly women. This has been associated with the reduction of bone turnover biochemical markers such as serum ALP.^{34,35} In addition, Braga de Castro Machado *et al.*³⁶ supported that

biochemical markers are a better way to monitor treatment with ALN than dual energy X-ray absorptiometry (DEXA) values. In our study, oral ALN intake by SCI patients was correlated with normal serum ALP and this may be indirectly associated with HO prevention. Also, since ALN intake was correlated with the appearance of joint contractures, we cannot be sure if these cases would have proceeded to HO if ALN was not prescribed and that ALN could prevent progression to severe HO-joint ankylosis.

A limitation to our study is the retrospective design with non-consistent start time or duration of ALN intake and different characteristics between the non-ALN and ALN patient groups. However, the results are valid as level III evidence. Since triple phase bone scan as gold standard for the diagnosis of HO was not used universally, there is a chance that we may have missed cases of subclinical or early HO. However, these cases were sparse, if any, based on the high index of suspicion of the physicians and therapists during their multiple and frequent patient evaluations. Furthermore, since the majority of patients in our population were of male sex, firm conclusions on the sex predilection of HO in patients with SCI cannot be drawn. Prospective randomized trials with controlled patient selection criteria as well as multicenter studies on the prevention of HO by oral ALN intake with bone-specific ALP measurement are warranted.

According to the results above, even though ALN intake does not seem to have direct preventive action for HO, it correlates with normal ALP levels and may indirectly influence HO. Patients with acute SCI with HO risk factors should have regular serum ALP measurements and when increasing ALP levels are found, ALN could be prescribed, if not contraindicated, for HO prevention.

Disclaimer statements

Contributors AP: Data collection, statistical analysis, manuscript writing. JD: Data collection and manuscript writing. MO: Data collection and manuscript writing. DC: Data collection and manuscript writing. JW: Data collection and manuscript writing. DS: Data collection. KOC: Review of manuscript.

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References

- van Kuijk AA, Geurts AC, van Kuppevelt HJ. Neurogenic heterotopic ossification in spinal cord injury. *Spinal Cord* 2002;40(7): 313–26.
- Banovac K, Williams JM, Patrick LD, Haniff YM. Prevention of heterotopic ossification after spinal cord injury with indomethacin. *Spinal Cord* 2001;39(7):370–4.
- Vanden Bossche L, Vanderstraeten G. Heterotopic ossification: a review. *J Rehabil Med* 2005;37(3):129–36.
- Buschbacher R, McKinley W, Buschbacher L, Devaney CW, Coplin B. Warfarin in prevention of heterotopic ossification. *Am J Phys Med Rehabil* 1992;71(2):86–91.
- Hardy AG, Dickson JW. Pathological ossification in traumatic paraplegia. *J Bone Joint Surg Br* 1963;45-B(1):76–87.
- Schuetz P, Mueller B, Christ-Crain M, Dick W, Haas H. Amino-bisphosphonates in heterotopic ossification: first experience in five consecutive cases. *Spinal Cord* 2005;43(10):604–10.
- Schurch B, Capaul M, Vallotton MB, Rossier AB. Prostaglandin e2 measurements: their value in the early diagnosis of heterotopic ossification in spinal cord injury patients. *Arch Phys Med Rehabil* 1997;78(7):687–91.
- Coelho CV, Beraldo PS. Risk factors of heterotopic ossification in traumatic spinal cord injury. *Arq Neuropsiquiatr* 2009;67(2B): 382–7.
- Citak M, Suero EM, Backhaus M, Aach M, Godry H, Meindl R, et al. Risk factors for heterotopic ossification in patients with spinal cord injury: a case-control study of 264 patients. *Spine (Phila Pa 1976)* 1976;37(23):1953–7.
- Garland DE, Orwin JF. Resection of heterotopic ossification in patients with spinal cord injuries. *Clin Orthop Relat Res* 1989; 67(8):169–76.
- Banovac K, Sherman AL, Estores IM, Banovac F. Prevention and treatment of heterotopic ossification after spinal cord injury. *J Spinal Cord Med* 2004;27(4):376–82.
- Banovac K, Williams JM, Patrick LD, Levi A. Prevention of heterotopic ossification after spinal cord injury with cox-2 selective inhibitor (rofecoxib). *Spinal Cord* 2004;42(12):707–10.
- Kurmis AP, Kurmis TP, O'Brien JX, Dalen T. The effect of non-steroidal anti-inflammatory drug administration on acute phase fracture-healing: a review. *J Bone Joint Surg Am* 2012;94(9): 815–23.
- Pountos I, Georgouli T, Calori GM, Giannoudis PV. Do nonsteroidal anti-inflammatory drugs affect bone healing? A critical analysis. *Scientific World Journal* 2012 (article id 606404):1–14.
- Banovac K. The effect of etidronate on late development of heterotopic ossification after spinal cord injury. *J Spinal Cord Med* 2000; 23(1):40–4.
- Garland DE, Alday B, Venos KG, Vogt JC. Diphosphonate treatment for heterotopic ossification in spinal cord injury patients. *Clin Orthop Relat Res* 1983;176:197–200.
- Teasell RW, Mehta S, Aubut JL, Ashe MC, Sequeira K, Macaluso S, et al. A systematic review of the therapeutic interventions for heterotopic ossification after spinal cord injury. *Spinal Cord* 2010; 48(7):512–21.
- Wittenberg RH, Peschke U, Botel U. Heterotopic ossification after spinal cord injury. Epidemiology and risk factors. *J Bone Joint Surg Br* 1992;74(2):215–8.
- Garland DE, Hanscom DA, Keenan MA, Smith C, Moore T. Resection of heterotopic ossification in the adult with head trauma. *J Bone Joint Surg Am* 1985;67(8):1261–9.
- Singh RS, Craig MC, Katholi CR, Jackson AB, Mountz JM. The predictive value of creatine phosphokinase and alkaline phosphatase in identification of heterotopic ossification in patients after spinal cord injury. *Arch Phys Med Rehabil* 2003;84(11): 1584–8.
- Estrores IM, Harrington A, Banovac K. C-reactive protein and erythrocyte sedimentation rate in patients with heterotopic ossification after spinal cord injury. *J Spinal Cord Med* 2004; 27(5):434–7.
- Kim SW, Charter RA, Chai CJ, Kim SK, Kim ES. Serum alkaline phosphatase and inorganic phosphorus values in spinal cord injury patients with heterotopic ossification. *Paraplegia* 1990; 28(7):441–7.
- Bernard GW. Ultrastructural localization of alkaline phosphatase in initial intramembranous osteogenesis. *Clin Orthop Relat Res* 1978;135:218–25.
- Siddique A, Kowdley KV. Approach to a patient with elevated serum alkaline phosphatase. *Clin Liver Dis* 2012;16(2):199–229.

- 25 Licata AA. Discovery, clinical development, and therapeutic uses of bisphosphonates. *Ann Pharmacother* 2005;39(4):668–77.
- 26 Banovac K, Gonzalez F. Evaluation and management of heterotopic ossification in patients with spinal cord injury. *Spinal Cord* 1997;35(3):158–62.
- 27 Banovac K, Gonzalez F, Renfree KJ. Treatment of heterotopic ossification after spinal cord injury. *J Spinal Cord Med* 1997;20(1):60–5.
- 28 Stover SL, Hahn HR, Miller JM III. Disodium etidronate in the prevention of heterotopic ossification following spinal cord injury (preliminary report). *Paraplegia* 1976;14(2):146–56.
- 29 Stover SL, Niemann KM, Miller JM III. Disodium etidronate in the prevention of postoperative recurrence of heterotopic ossification in spinal-cord injury patients. *J Bone Joint Surg Am* 1976;58(5):683–8.
- 30 Subbarao JV, Nemchausky BA, Gratzner M. Resection of heterotopic ossification and didronel therapy—regaining wheelchair independence in the spinal cord injured patient. *J Am Paraplegia Soc* 1987;10(1):3–7.
- 31 Yaffe A, Kollerman R, Bahar H, Binderman I. The influence of alendronate on bone formation and resorption in a rat ectopic bone development model. *J Periodontol* 2003;74(1):44–50.
- 32 Moran de Brito CM, Battistella LR, Saito ET, Sakamoto H. Effect of alendronate on bone mineral density in spinal cord injury patients: a pilot study. *Spinal Cord* 2005;43(6):341–8.
- 33 Ben Hamida KS, Hajri R, Kedadi H, Bouhaouala H, Salah MH, Mestiri A, *et al*. Myositis ossificans circumscripta of the knee improved by alendronate. *Joint Bone Spine* 2004;71(2):144–6.
- 34 Greenspan SL, Parker RA, Ferguson L, Rosen HN, Maitland-Ramsey L, Karpf DB. Early changes in biochemical markers of bone turnover predict the long-term response to alendronate therapy in representative elderly women: a randomized clinical trial. *J Bone Miner Res* 1998;13(9):1431–8.
- 35 Zehnder Y, Risi S, Michel D, Knecht H, Perrelet R, Kraenzlin M, *et al*. Prevention of bone loss in paraplegics over 2 years with alendronate. *J Bone Miner Res* 2004;19(7):1067–74.
- 36 Braga de Castro Machado A, Hannon R, Eastell R. Monitoring alendronate therapy for osteoporosis. *J Bone Miner Res* 1999;14(4):602–8.