Effects of strontium malonate (NB S101) on the compositional, structural and biomechanical properties of calcified tissues in rats and dogs

Strontium is known to have a positive effect on bone by concomitantly increasing bone formation and decreasing bone resorption, thereby providing a sustained skeletal benefit. Strontium ranelate (SrR) has been shown to reduce the risk of both vertebral and non-vertebral fractures in patients with postmenopausal osteoporosis. Strontium malonate (SrM) is currently being developed as a novel pharmaceutical for the treatment and prevention of osteoporosis. SrM potentially provides considerable advantages over SrR with respect to Sr content, bioavailability and ease of administration. SrM was tested in three animal studies: 1) a 4-week study in dogs using SrM doses of 0 (control), 300, 1000 and 3000 mg kg-1 day-1, 2) a 26-week study in rats, and 3) a 52-week study in dogs, both using SrM doses of 0 (control), 100, 300 and 1000 mg kg-1 day-1. Femurs, vertebrae, skullcaps and teeth from the treated animals were examined for treatment-related changes in concentrations of Sr, Ca, Mg and P using inductively coupled mass spectrometry (ICP-MS). Bone mineral density (BMD) was determined using dual energy X-ray absorptiometry (DEXA), and the biomechanical properties of the bones were assessed using bending and compression tests. A procedure was developed for determination of Mg, P, Ca and Sr in diluted serum using ICP-MS in combination with an Apex-Q desolvation unit. The Apex inlet system reduced the generation of oxides in the ICP and improved the sensitivity for Sr by a factor of 14 compared with a conventional cross-flow nebuliser. Rh was found to be a suitable internal standard for all four analytes. Reliable estimates of the measurement uncertainties were achieved by pooling calibration data obtained on different days. Treatment with SrM resulted in a dose-dependent increase in Sr contents in all analysed tissues. The highest concentrations were found in rat incisor, which contained an average of 45 ± 11 mg g-1 Sr in the highest dose group, corresponding to a 450-fold increase compared with placebo. The Sr/(Sr+Ca)mol% in the four groups were respectively 0.015, 1.2, 5.8 and 7.7 for incisors and 0.015, 0.6, 1.9 and 5.0 for femurs. Sr concentrations in rat incisor and rat femur were strongly correlated with approx. 30% less Sr found in the femurs. A strong correlation between serum Sr and incisor Sr was also observed. In dogs, the highest concentrations of Sr were found in skullcap after 52 weeks of treatment, with an average of 28 ± 12 mg g-1 Sr in the highest dose group, corresponding to a 350-fold increase compared with placebo. The Sr/(Sr+Ca)mol% in the four treatment groups were respectively 0.011, 2.5, 3.8 and 4.4 for femurs; 0.019, 3.3, 4.7 and 6.5 for calvaria and 0.013, 0.75, 1.1 and 1.6 for molar teeth. Sr concentrations in femur and molar were correlated. Sr concentrations in dog femur mid-shaft after 4 weeks were approx. 1/3 of the concentrations after 52 weeks, suggesting that initial incorporation of Sr proceeded by a faster mechanism than the long-term incorporation. In both rats and dogs, longterm treatment with SrM was associated with decreased concentrations of Ca and P in the mineralised tissues. EXAFS measurements indicated that Sr had substituted Ca in the bone apatite structure, but that Sr also coordinated to other atoms, most likely oxygen in collagen and water. The incorporation of Sr into the HAp increased with increasing dose. A simple stoichiometric model was proposed for incorporation of Sr into the hydroxyapatite (HAp) matrix of bones and teeth. The model showed excellent agreement with the analytical data and demonstrated that at least a part of the perceived loss of Ca and P was caused by the increased weight of HAp following adsorption and incorporation of Sr. It was found that respectively 70 – 72% of femur, 78% of rat incisor and 85% of dog molar was made up by HAp. The maximum strength of the dog femurs (three-point bending) was significantly reduced (19%) in the highest dose group, indicating a weakening of the apatite structure. A non-significant increase (9%) in strength was seen in the low and medium dose groups. Measurements of bone mineral density (BMD) using DEXA was found to be heavily influenced by presence of Sr in the mineralised matrix, and linear correction of 9% was determined for a 1:10 molar ratio of Sr:HAp. The corrected BMD increased significantly in dog femur mid-shaft in the group treated with 300 mg kg-1 day-1 SrM. The corrected BMD was found to be strongly correlated with bone strength, indicating that the increase in corrected BMD following SrM treatment was associated with concomitant increase of bone strength.