The Effects of a Selective Androgen Receptor Modulator (SARM) ORM-11984 on Prevention of Osteoporosis in Rat Immobilization Model

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Introduction
Androgens play an important role in regulating bone development and bone metabolism. Selective androgen receptor modulators (SARMs) maintain the beneficial effects of androgens. A novel orally active non-steroidal androgen receptor (AR) agonist ORM-11984 has shown beneficial effects on primary osteoporosis induced by ovariectomy and orchiectomy in rats (1, 2). Disuse induced by limb immobilization acts as an external factor leading to secondary osteoporosis (3). Not much is known about the effects of SARMs on osteoporosis caused by immobilization.

Aim of the Study
The objective of this study was to investigate the effects of a novel orally active non-steroidal AR agonist ORM-11984 on secondary osteoporosis caused by disuse in the rat immobilization model.

Materials and Methods
Three-month-old male Sprague-Dawley rats were divided into the following groups (with 12 animals in each group): 1) Control group without immobilization receiving vehicle (0.5% methyl cellulose p.o.); 2) Immobilized group receiving vehicle; 3) Immobilized group receiving the reference compound clodronate 10 mg/kg/day s.c. and vehicle; 4) Immobilized group receiving ORM-11984 0.5 mg/kg/day p.o.; 5) Immobilized group receiving ORM-11984 3.0 mg/kg/day p.o. Left hind limbs of the animals in groups 2-5 were immobilized by a plaster cast in plantar flexion under anesthesia (Figure 1A). The casts were checked daily and renewed when necessary. Treatment was continued daily for 3 weeks, after which the animals were terminated and their hind left limbs were harvested. Follow-up of bone mineral density (BMD) and cross-sectional dimensions was performed by peripheral quantitative computed tomography (pQCT) in vivo from left tibia of each animal at days 0 and 21. At the end of the study, the tibiae were used for static and dynamic histomorphometry. For dynamic histomorphometry, tetracycline and calcein were administered s.c. at days 6 and 21 before the end of the study, respectively. Statistical analysis was performed with one-way analysis of covariance (ANCOVA) for follow-up measurements and with one-way analysis of variance (ANOVA) or Kruskal-Wallis test for endpoint measurements after checking assumptions for normality and homogeneity of variances. Linear contrasts of means or Mann-Whitney U-test were utilized for pairwise comparisons.

Immobilization and Body Weight

![Figure 1. Example of a rat with the left hind limb immobilized in plantar flexion by a plaster cast; B) Change of body weight during the study. The groups are: 1) Control group without immobilization receiving vehicle; 2) Immobilized group receiving vehicle; 3) Immobilized group receiving the reference compound clodronate 10 mg/kg/day s.c. and vehicle; 4) Immobilized group receiving ORM-11984 0.5 mg/kg/day p.o.; 5) Immobilized group receiving ORM-11984 3.0 mg/kg/day p.o.](image)

Body weight

![Figure 2. pQCT measurements.](image)

- Trabecular BMC of tibial metaphysis
- Trabecular bone mineral content (BMC) of tibial metaphysis
- Total BMC of tibial diaphysis
- Cortical thickness of tibial diaphysis

pQCT Measurements

![Figure 3. Static histomorphometry in trabecular bone of tibial metaphysis. A) Trabecular number (Tb.N); B) Number of osteoclasts per bone perimeter (N.Oc/B.Pm). The groups are: 1) Control group without immobilization receiving vehicle; 2) Immobilized group receiving vehicle; 3) Immobilized group receiving clodronate 10 mg/kg/day s.c. and vehicle; 4) Immobilized group receiving ORM-11984 0.5 mg/kg/day p.o.; 5) Immobilized group receiving ORM-11984 3.0 mg/kg/day p.o. One asterisk (*) indicates a statistically significant difference from group 1 with p < 0.05 and three asterisks (****) with p < 0.001. One b-letter (b) indicates a statistically significant difference from group 2 with p < 0.05 and three b-letters (bbb) with p < 0.001.](image)

Dynamic Histomorphometry

![Figure 4. Bone formation rate/bone surface (BFR/BS) is A) trabecular bone of tibial metaphysis; B) perisurface of tibial diaphysis; C) perisurface of total bone. The groups are: 1) Control group without immobilization receiving vehicle; 2) Immobilized group receiving vehicle; 3) Immobilized group receiving clodronate 10 mg/kg/day s.c. and vehicle; 4) Immobilized group receiving ORM-11984 0.5 mg/kg/day p.o.; 5) Immobilized group receiving ORM-11984 3.0 mg/kg/day p.o. One asterisk (*) indicates a statistically significant difference from group 1 with p < 0.05, two asterisks (**) with p < 0.01 and three asterisks (****) with p < 0.001. One b-letter (b) indicates a statistically significant difference from group 2 with p < 0.05, two b-letters (bb) with p < 0.01 and three b-letters (bbb) with p < 0.001.](image)

Static Histomorphometry

![Figure 2. Static histomorphometry in trabecular bone of tibial metaphysis. A) Trabecular number (Tb.N); B) Number of osteoclasts per bone perimeter (N.Oc/B.Pm). The groups are: 1) Control group without immobilization receiving vehicle; 2) Immobilized group receiving vehicle; 3) Immobilized group receiving clodronate 10 mg/kg/day s.c. and vehicle; 4) Immobilized group receiving ORM-11984 0.5 mg/kg/day p.o.; 5) Immobilized group receiving ORM-11984 3.0 mg/kg/day p.o. One asterisk (*) indicates a statistically significant difference from group 1 with p < 0.05 and three asterisks (****) with p < 0.001. One b-letter (b) indicates a statistically significant difference from group 2 with p < 0.05 and three b-letters (bbb) with p < 0.001.](image)

Summary
- Immobilization decreased body weight (Figure 1B) and induced osteoporotic changes as observed by pQCT measurements (Figure 2A, B) and dynamic histomorphometry (Figure 4).
- The reference compound clodronate showed anti-catabolic effects that were particularly strong in trabecular bone (Figures 2-4).
- ORM-11984 increased trabecular BMC and trabecular BMC in tibial metaphysis (Figures 2A and B). These results were supported by static histomorphometry, where ORM-11984 increased trabecular number (Figure 3).
- In dynamic histomorphometry, ORM-11984 showed anabolic effects in trabecular bone of tibial metaphysis, contrary to the anti-resorptive effects observed for the reference compound clodronate (Figure 4A).
- ORM-11984 increased total BMD in tibial diaphysis (Figure 2C) but showed no effects on cortical thickness (Figure 2D) or BFR/BS in either periosteal or endocortical surfaces of tibial diaphysis (Figures 4B and C).

Conclusions
Immobilization caused secondary osteoporosis that was effectively prevented by clodronate through anti-catabolic effects in particular trabecular bone, demonstrating that the study was performed successfully.

The orally active non-steroidal AR agonist ORM-11984 showed anabolic effects in trabecular bone in tibial metaphysis and on total BMD of tibial diaphysis in the rat immobilization model.

References

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