FACULTY OF HEALTH SCIENCES
UNIVERSITY OF COPENHAGEN

REGION H Glostrup Hospital

The P2X7 Receptor and Inflammation-Mediated Osteoporosis

T. M. Kvist [1,] , S. Syberg[1], A. G. Frederiksen[1], N. R. Jørgensen[1], S. Petersen[1], J-E. B. Jensen[2], P. Schwarz[1,3]

[1]Research Center for Ageing and Osteoporosis, Departments of Medicine and Biochemistry, Copenhagen University Hospital Glostrup; [2]Copenhagen University Hospital Hvidovre, Denmark; [3]Faculty of Health Sciences, University of Copenhagen, Denmark.

Background

Rheumatic inflammatory autoimmune diseases are highly prevalent in western world with a life time risk of about 5-8 % and Bone loss is a frequent complication. The pathophysiology behind the bone loss is largely unknown. Bone loss correlates with inflammation and disease activity.

The P2X7 receptor, an ATP-gated ion-channel, is primarily expressed on immune and bone cells. ATP is now seen as a novel inflammatory mediator, with P2X7 as main target of the pro-inflammatory activity. The P2X7-receptor has a regulatory role in bone formation and resorption and also important regulating osteoclasts and osteoblasts.

Aim

To investigate the role of the P2X7 Receptor in a mouse model of Inflammation-Mediated Osteoposis (IMO).

Methods

Study design: 150 14-week-old male C57/B6bom and B6P2x7-/-(KO) were used, 75 of each. As shown in figure 1, 15 were sacrificed at Baseline. The remaining 60 were randomised to isotonic NaCl (vehicle) or 16 mg/g body weight Talc (Hydrous Magnesium silicate: Sigma Aldrich) injection subcutaneously on the back and sides of the animal (inducing a chronic inflammation). Mice were sacrificed, blood were drawn by cardiac puncture, Spleen was collected and weighted. Spine and hind legs were collected for further analysis.

Dual-energy X-ray Absorptiometry (DXA) were preformed on the PIXImus densitometer (Lunar) with animals in a fixed standard position and with duplicate determinations In order to evaluate BMD. Talc interferred with the DXA analysis, and the Spine were removed and rescanned.

Biomechanical testing (Lloyd instruments LR 50k, Fareham, UK) were performed on femoral midshaft (3-point-bending test) and femoral neck on the mouse femurs that were cleaned for tissue, wrapped in saline gauze, and frozen at −20°C until tested

Statistics: Standard parametric tests, T-test and ANCOVA were used as appropiate. Differences were considered statistically significant when $p < 0.05$. Simple descriptive were presented as means ± standard error of the mean (SEM).

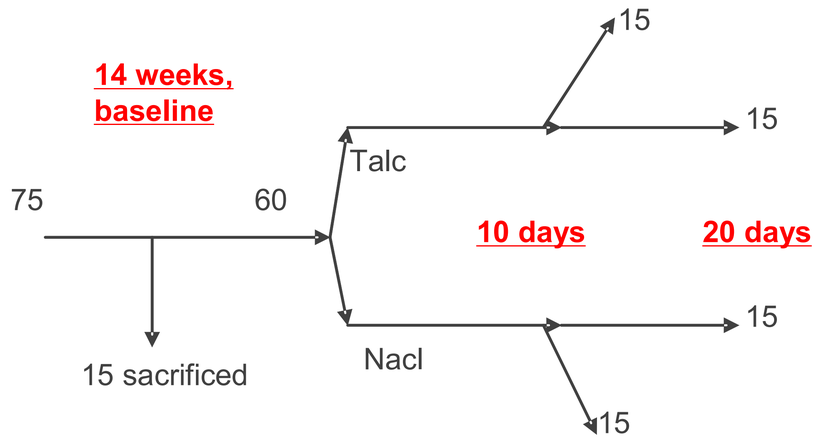


Figure 1: Study design:15 are sacrificed at baseline. 30 mice injected with Talc were sacrificed at 10 and 20 days, (15 mice at each time point). The remaining 30 mice were injected with vehicle and sacrificed at 10 and 20 days, (15 mice at each time point).

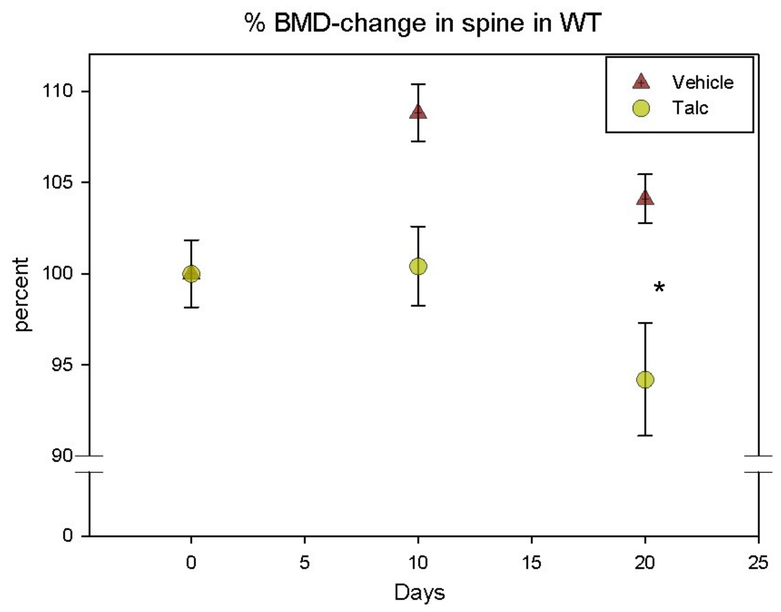


Figure 2 BMD change in lumbar spine. Data are presented as means ± SEM. * denotes a significant difference between the two groups (p=0,009)

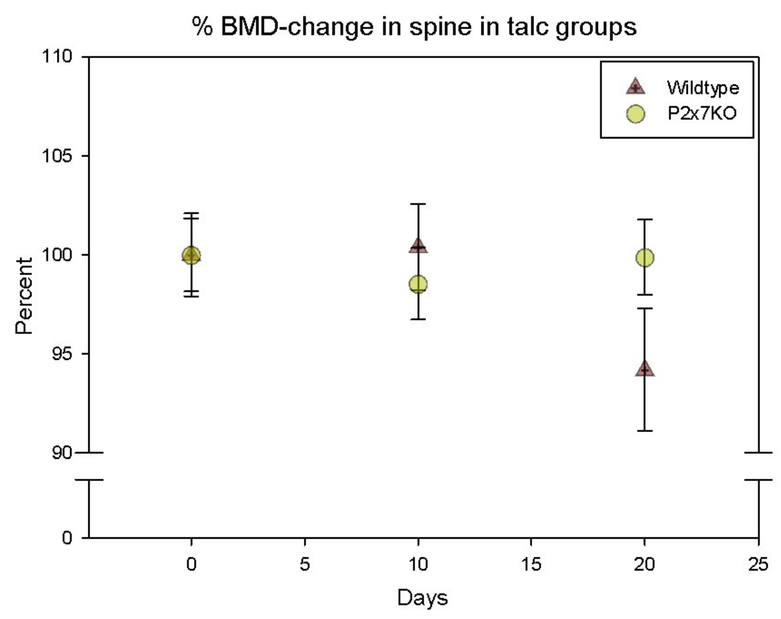


Figure 3 BMD change in lumbar spine. Data are presented as means ± SEM.

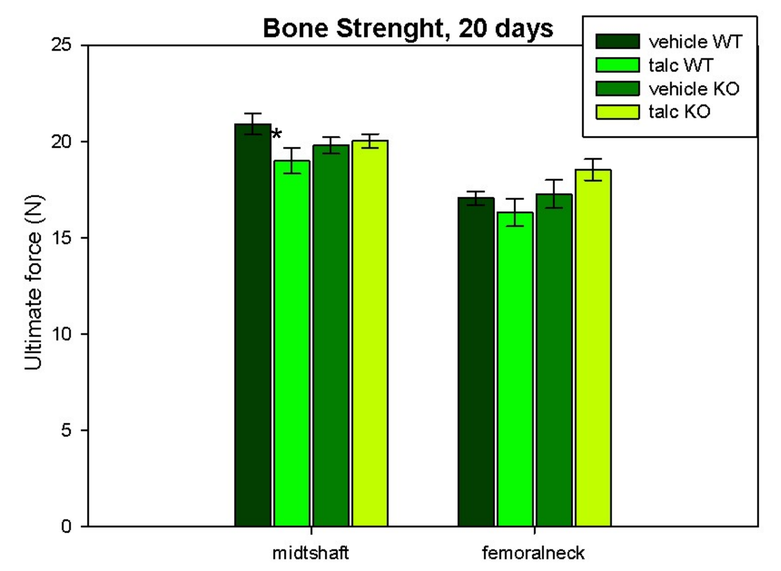
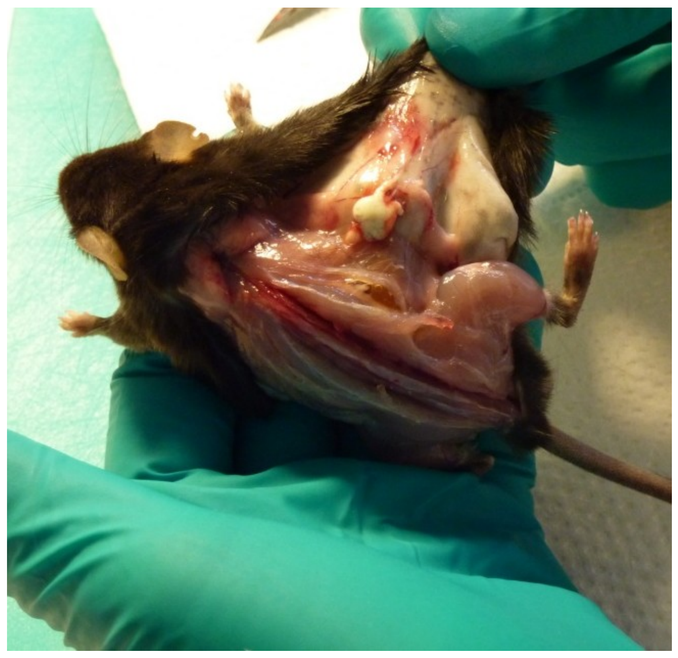


Figure 4 Ultimate force of femoral neck and shaft at 20 days. Data are presented as means ± SEM. * Talc WT midt shaft were significantly lower than vehicle (p=0.038)



	0 days	10 days	20 days
WT Vehicle	86,42 (sd 13.2)	100,24 (sd 20,8)	109,0 (sd 32,2)
Wt Talc	86,42 (sd 13.19)	130,96 (sd 59,0)	130,76 (sd 44,4)
KO Vehicle	104,27 (sd 27,0)	98,57 (sd 39,7)	101,90 (sd 19,1)
KO Talc	104,27 (sd 27,0)	112,11 (sd 37,1)	108,30 (sd 15,1)

Table 1: Spleen weight. Data as mean and SD.

Results

In WT animals, the mean spleen weight was significantly higher at 10 and 20 days compared to other groups. No significant difference was found between the KO groups. (Table 1)

At 20 days spine BMD was significantly lower in the Talc WT group compared to vehicle (0.046 vs. 0.051 g/cm: p=0.009) (fig 2). Spine BMD was significantly lower at 20 days compared to baseline in WT in the talc group (ANCOVA weight corrected: p=0.032) (fig 2)

In the KO animals no significant difference was found at 20 days between vehicle and Talc group (0.052 vs. 0.052 g/cm)(ns) (fig 3)

At 20 days a significant lower ultimate force at femoral midtshaft were found in the WT Talc group compared to Vehicle (p=0.038). Femoral neck in the talc WT was lower but not significantly. No significant differences were found in the KO groups at 20 days. (fig 4)

Conclusion

In conclusion, P2X7 might be involved in the inflammation-mediated osteoporosis – but further data are needed.

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Conflicts of interest: None