**EFFECT OF CHOLINE STABILIZED ORTHOSILICIC ACID ON BONE DENSITY IN Ovariectomized Rats**

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**Introduction**

OVX significantly decreased bone mineral density (BMD) in the femur (-15%, SHAM vs OVX 0; p<0.0001) and spine (-14%, SHAM vs OVX 0; p=0.0005). Ch-OSA supplementation increased significantly the femoral BMD at two sites in the distal region (H2: +4.2%, H3: +7.2%, OVX 1 vs OVX 0, p<0.05) (fig 2). Lumbar BMD was marginally increased by ch-OSA supplementation in all vertebrae (fig 3).

**Results**

The serum Si concentration of supplemented rats was significantly higher compared to SHAM (p<0.0001) and OVX controls (p = 0.001). The 24 hour urinary Si excretion of supplemented OVX rats was significantly higher compared to SHAM (p=0.001) and OVX controls (p<0.0001) (table). OVX controls tended to have the lowest urinary Si excretion. The OVX rats differed from intact rats by respectively a decreased urinary excretion of Ca and P, and increased levels in serum osteocalcin and alkaline phosphatase activity. Supplementation with ch-OSA partially reversed the decrease in Ca excretion. Osteocalcin and alkaline phosphatase levels tended to be lower after ch-OSA in OVX rats compared to OVX controls.

**Methods**

Nine-months old female Wistar rats (n=58) were randomly assigned to 3 groups. One group was sham operated (SHAM, n=21) and bilateral ovariectomy (OVX) was performed in the other 2 groups: one group (OVX1, n=20) was supplemented with ch-OSA (1mg Si/kg BW/day, Bio Minerals n.v., Belgium) in the drinking water whereas rats in the second group (OVX0, n=17) were controls. Rats were pair fed (vs SHAM) a casein-based diet (0.9% Ca, 0.7% P, Altromin, Germany). Urine was collected in metabolic cages after 22 weeks of supplementation and rats were sacrificed after 30 weeks supplementation. Serum was collected after cardiac puncture in Si-free labware. Bone mineral content (BMC) and density (BMD) were analyzed by Dual Energy X-Ray Absorptiometry and were recorded for total femur, 4 regions of interest in the femur (H1: midshaft; H2, H3, H4: distal metaphysis), and lumbar vertebrae (L1-L4, see fig. 1). Si concentration was measured in serum and urine by Electrothermal Atomic Absorption Spectrometry with Zeeman background correction. Total urinary calcium and phosphor concentrations were determined by spectrophotometry. Alkaline phosphate was determined by spectrophotometry measuring the release of p-nitro phenol from p-nitro phenolphosphate. The enzyme activity was determined as the change of absorbance at 405 nm. Osteocalcin was measured using the rat osteocalcin Immuno Radiometric Assay (IRMA) kit (Immunotopics, Inc., USA).

**Table:** Serum and urine parameters (mean ± SE) measured in OVX rats supplemented with ch-OSA (OVX 1) compared to intact (SHAM) rats and OVX controls (OVX 0). a: p<0.05 vs SHAM; b: p<0.05 vs OVX control (Mann-Whitney U-test).

**Fig. 1:** Scanned areas of interest in the femur.

**Fig. 2:** Femoral BMD (mg/cm², mean ± SD) of OVX rats supplemented with ch-OSA (OVX 1) compared to intact controls (SHAM) and OVX controls (OVX 0).

**Results**

- OVX significantly decreased bone mineral density (BMD) in the femur (-15%, SHAM vs OVX 0; p=0.0001) and spine (-14%, SHAM vs OVX 0; p=0.0005).
- Ch-OSA supplementation increased significantly the femoral BMD at two sites in the distal region (H2: +4.2%, H3: +7.2%, OVX 1 vs OVX 0, p<0.05) (fig 2). Lumbar BMD was marginally increased by ch-OSA supplementation in all vertebrae (fig 3).

**Conclusion**

Long-term preventive treatment with ch-OSA prevents partially but significantly femoral bone loss in the aged ovariectomy rat model. The present results confirm earlier studies in animal and men suggesting that silicon is involved in bone metabolism and that ch-OSA (a stabilized form of orthosilicic acid) may have a beneficial action on bone health.

**References**