Increased Calcium Content and Inhomogeneity of Mineralization Render Bone Toughness in Osteoporosis: A Study Focusing on the Mineralization, Morphology and Biomechanics of Human Single Trabeculae

Introduction:
Numerous factors are considered to determine human bone quality. Particularly, volumetrically evaluated bone mineral density (DXA-BMD) as well as the mechanical properties of bone samples serve to assess fracture risks. However, the differentiation and degree of mineral content and/or morphology effects on bone toughness remained to a large extent unanswered due to large measuring fields containing several micro-architectural particularities (force transfer, trajectories, microcallus). Therefore, the combined analyses of three-point-bending tests and bone mineral density distribution (BMDD) were performed on single trabeculae - as the least basic component of cancellous bone - to evaluate the respective effects of mineralization and morphology in terms of bone quality.

Material and Methods:
Transiliac Border bone biopsies as well as T12 vertebrae were obtained from 18 female donors. Both iliac crest and vertebrae bone samples were prepared to ground tissue specimen. Von Kossa staining of the specimen enabled static bone histomorphometry in accordance to the ASBMR (American Society of Bone and Mineral Research) guidelines [1]. The histomorphometric assessment of vertebral BV/TV (bone volume/tissue volume), Tb.Th. (trabecular thickness), Tb.N. (trabecular number) and Tb.Sp. (trabecular separation) served for the individual classification of the donors’ bone status. Hence, the cases were divided into one osteoporosis group (n = 13, Ø 78 yrs.) under consideration of previous elevated histomorphometric limits obtained from cases with osteoporotic vertebral fractures [2] and one group with normal bone status according to its age without pathological findings (n = 5, Ø 45 yrs.). Further sections of the T12 vertebrae served for the singularization of trabeculae following soft tissue removal. The singularization of 20 trabeculae per case with a thickness < 90 μm approved 3-point-bending and the determination of mechanical properties. Moreover, in both groups single trabeculae within the range of 140-160 μm were selected to provide similar trabecular thickness for the exclusion of morphological effects in terms of material testing. Bending stiffness, Young’s modulus and work to failure were matched with mineral properties due to calcium (Ca) content and homogeneity of mineralization received by BMDD analyses of the same sample region.

Results:
The histomorphometric evaluation of osteoporosis and skeletally intact cases indicated significantly decreased values for BV/TV (5.9 ± 1.43 vs. 11.78 ± 3.13; (%), Tb.Th. (90.2 ± 12.7 vs. 106 ± 23.2; (μm)), Tb.N. (0.73 ± 0.14 vs. 1.05 ± 0.11; (mm²)) and Tb.Sp. (1270.78 ± 338.0 vs. 807.94 ± 136.35; (μm)) within the osteoporotic vertebrae. Structural parameters in the iliac crest showed same tendency, however with slightly elevated values for BV/TV (9.87 ± 3.33 vs. 16.33 ± 5.37; (%)), Tb.Th. (95.56 ± 22.03 vs. 112.2 ± 29.88; (μm)), Tb.N. (0.85 ± 0.23 vs. 1.28 ± 0.25; (mm²)) and Tb.Sp. (1071.49 ± 457.95 vs. 610.48 ± 214.11; (μm)) (Fig. A,B). The BMDD represented in skeletal intact trabeculae interstitially higher mineralized zones framed by lower mineralized trabecular surfaces (Fig. C), a combination which is considered as the toughest composition [3]. The osteoporotic trabeculae, in contrast, indicated a non-uniform alignment of various mineralized bone packets framed by cement lines with highest Ca concentrations. Occasionally, microcracks appeared in highly mineralized bone packets as well as eroded surfaces due to osteoclast activity (Fig. D).

Consequentially, the osteoporosis group showed in comparison to the skeletally intact cases significantly elevated values for the mean Ca content (22.34 ± 0.85 vs. 21.66 ± 0.29; (Wt-%)) as well as in inhomogeneity of mineralization reflected in Ca width (4.46 ± 0.3 vs. 4.22 ± 0.23; (ΔWt-%)) (Fig. C-E). According to this, the mechanical properties imposed by 3-point bending indicated a significantly decrease of bending stiffness (0.049 ± 0.014 vs. 0.03 ± 0.013; (Nm)), Young’s modulus (2.14 ± 0.51 vs. 1.21 ± 0.55; (GPa)) and work to failure (0.049 ± 0.014 vs. 0.03 ± 0.013; (mJ)) in the osteoporosis group, independently from Tb.Th.

Discussion:
The burden of osteoporotic fractures is related to various and multifactorial effects. Aging, menopause respectively estrogen withdrawal, metabolic and musculoskeletal changes may concern bone structure and/or bone mineralization. Hence, measurements focusing on bone mass and mineral density in combination, may be improved by the differentiability of architecture and elemental composition in respect of optimal targeted osteoporosis treatment. Besides the mineralization focus on the major part of inorganic bone constituents the presence of the undocumented smaller part of organic components (collagen) needs to be addressed. Nevertheless, the findings showed that beyond trabecular thickness (morphology) the degree of mineralization, particularly the homogeneity of mineralization, plays a decisive role concerning toughness and fracture risk of cancellous bone.

References: