Electromechanical Properties Of Cortical Bone Across Hydration States: A Proxy For Bone Quality

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INTRODUCTION: Bone quality is a collection of attributes and characteristics that determine bone strength and integrity. It encompasses bone mineral density but extends beyond it to include other structural and compositional parameters. From a material perspective, bone can be regarded as a hierarchical composite in which water is a structurally significant phase, distributed across different levels, and it contributes in distinct ways to the behavior of the bone[1]. Variations in water content are closely linked to porosity and ductility, parameters that strongly influence bone fragility. These diverse roles of water affect both the mechanical and electrical properties of bone. Advanced imaging methods, such as short echo-time MRI, can quantify bone water, but their application has largely remained limited to research due to cost and accessibility. In this study, we investigate how bone water content influences both electrical and mechanical properties, with the goal of evaluating its feasibility as a proxy for bone quality and supporting the development of non-invasive, cost-effective diagnostic techniques.

METHODS: Cortical bone samples were harvested from the diaphysis of fresh-frozen bovine femurs (N=12). After marrow removal, rectangular prism and dog-bone-shaped specimens were prepared using a band saw and CNC machining. Samples from femurs were distributed across six regions of the diaphysis to minimize location-dependent bias, and the impedance data were analyzed by modeling bone behavior as a circuit with resistive and capacitive components. Water content was systematically altered through cycles of vacuum drying (≤60 °C to minimize collagen matrix compromise), rehydration with phosphate-buffered saline (PBS), and heat drying up to 110 °C. Drying and rehydration cycles were used to quantify water loss and assess the reversibility of hydration, and weight changes were recorded to monitor variation in water content. Although artificially removing water from bone deviates from in vivo conditions, it has been used in the literature to develop in vitro testing of bone properties. Mechanical properties were evaluated through standardized tensile testing of dog-bone specimens, while electrical properties were assessed using electrical impedance spectroscopy (EIS). For EIS, rectangular prism samples were prepared with their ends coated in silver conductive paint to ensure uniform electrical contact. Measurements were performed along the longitudinal axis of the bone to align with its primary load-bearing orientation. Each sample was tested across a frequency range of 20 Hz-10 MHz, later refined to 1 kHz-1 MHz to account for setup limitations, with 1,600 frequency points collected three times per run.

RESULTS SECTION: Vacuum drying (16 h, ≤60 °C) reduced the weight of bone samples by 5%, while heat drying resulted in a 13% reduction. EIS showed that within the experimental frequency range, bone exhibited electric impedance behavior similar to a circuit with a parallel resistor and capacitor (RC). Compared to hydrated samples, vacuum-dried specimens demonstrated significant increases in real and imaginary impedance, respectively (Fig. A & B). When comparing the phase angle vs. frequency behavior of hydrated vs. dried bone samples, both were consistent with parallel RC circuit behavior, but a clear shift was present, demonstrating the change in resistive and capacitive values between the hydrated and dry states of bone (Fig. B). Tensile testing further demonstrated that reduced water content resulted in an altered stress-strain curve compared to the hydrated state, underscoring the structural role of water in maintaining load-bearing function.

DISCUSSION: These findings demonstrate that water content is a critical structural factor influencing cortical bone's electrical and mechanical properties. While the degree of water removal exceeded in vivo changes, the experiments provide valuable insight into hydration-electromechanical relationships in cortical bone. A key limitation is that both electrical and mechanical testing were confined to the longitudinal axis, reflecting the primary load-bearing orientation of cortical bone. While appropriate for evaluating structural strength, future investigations should also explore other directions, especially the radial direction, where transport pathways, porosity, and water distribution may reveal additional insights. Furthermore, the use of bovine cortical bone and restriction to a single anatomical site highlights the need for physiologically relevant studies in human bone. Importantly, the strong link between impedance-derived parameters and mechanical performance underscores the potential of EIS as a non-invasive, water-sensitive tool for bone quality assessment.

SIGNIFICANCE/CLINICAL RELEVANCE: As the global population ages, the incidence of skeletal diseases such as osteoporosis—characterized by impaired bone quality and elevated fracture risk—is expected to rise. Early diagnosis and routine monitoring of bone health are critical for effective prevention and management. This study provides foundational evidence that electrical impedance spectroscopy may serve as a non-invasive method to evaluate water-related changes in bone quality.

REFERENCES: (1)Surowiec, R. K., Allen, M. R., & Wallace, J. M. (2022). Bone hydration: How we can evaluate it, what can it tell us, and is it an effective therapeutic target?. *Bone reports*, *16*, 101161.

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