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MECHANICAL CHARACTERIZATION OF BIOLOGICAL SAMPLE SURFACES USING SCANNING ACOUSTIC MICROSCOPY (SAM)

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INTRODUCTION: Characterization of biological tissues and degradable implants like bone substitute materials based on calcium phosphates (CaP) is of great importance when considering the healing process subsequent to large bone loss. *In vivo* degradation of the CaP-cements and ingrowth of newly formed bone into the healing site ideally happen simultaneously, which is called "creeping substitution". The degrading implant material must guarantee mechanical stability of the healing site until the newly formed bone can cover functional needs. Therefore, characterization with focus on mechanical properties is desirable. Scanning acoustic microscopy (SAM) allows mechanical characterization of smooth sample surfaces with a microscopic resolution. This technique measures the alteration of acoustic waves when they are reflected at interfaces[1]. This is most easily considered in terms of acoustic impedance, which is a surface sensitive elastic property that can be correlated with an elastic modulus. It is proportional to the pressure of the acoustic wave divided by the particle displacement velocity, which is a material dependent property. The aim of this study was to evaluate the applicability of scanning acoustic microscopy for the mechanical evaluation of biological tissues and bone substitute materials in specimens prepared for microscopic evaluation.

METHODS: The custom built SAM used in this work was operated at a frequency of 50 MHz in a pulse mode. This yields to a resolution in the range of 30 µm. Specimens containing different tissues (e.g. cortical bone, tooth, CaP-cement, soft tissue) were embedded in poly methyl methacrylate (PMMA) and cut into slices of about 300 µm. The surfaces were diamond milled and polished using 1 µm diamond particles. Changes in surface roughness were measured quantitatively by laser profilometry. SAM measurements were performed to investigate the influence of surface quality on the outcome of impedance values. The effect of embedding in PMMA was assessed as well.

RESULTS: Additional polishing of samples yields to a decrease in surface roughness of 36 % and an increase in the measured impedance values of 13 to 18 % (table 1). Impedance values for different tissues, CaP-cement and two reference materials (brass and PMMA) are displayed in figure 1. The range of expected values lies between 3 and 20 MRayl. CaP-cement exhibits similar values than bone tissue. Embedding cortical bone in PMMA yields to a loss of impedance of about 10 %.

Table 1. Comparison of acoustic impedance and surface roughness parameter for a milled and an additionally polished tooth.

	Impedance [MRayl]		Roughness R_a [µm]
	enamel	dentine	tooth
milled	10.3	5.6	0.42
polished	12.2	6.3	0.27

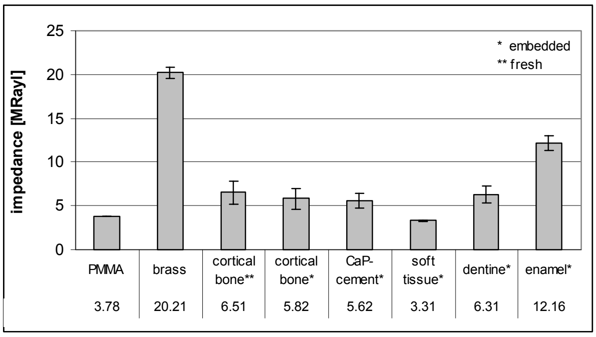

Fig. 1: Impedance measurement of different tissues, CaP-cement and reference materials.

DISCUSSION & CONCLUSIONS: The results of impedance measurements not only depend on mechanical properties of the sample, but also on its surface roughness. A rough surface yields to a bigger scattering of the wave and thus to a more attenuated signal. Therefore, sample preparation plays a crucial role. Embedding of samples in PMMA also affects the impedance values. Consequently, impedance measurements may only be used for comparative analysis. If parameters like density or an elastic modulus should be evaluated, additional knowledge of the wave velocity within a sample is required. Thus, the measurements must be calibrated with other independently obtained parameters like transmission ultrasound velocity in fresh specimens or radiological density.

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