INTRODUCTION:
Bone mineral and matrix tissue properties are critical to the mechanical competence of bone. Little is known about the material properties of the growing femoral head as it develops deformity following ischemic injury. The degree of bone matrix mineralization is an important determinant of the stiffness and hardness of the bone material and can be measured by quantitative backscattered electron imaging (qBEI) [1]. This established and validated method enables the description of the bone mineralization density distribution (BMDD) at the microscopic level. BMDD is the frequency distribution of the appearance of a certain mineral content found in the individual bone packets of varying age and mineral content comprising the bone structure. The purpose of this study was to determine if bone material properties are altered in necrotic bone using qBEI analysis in a well established piglet model of Legg-Calve-Perthes disease [2].

MATERIAL AND METHODS:
The study was approved by the Institutional Animal Care and Use Committee at Shriners Hospital for Children, Tampa, FL, USA. Ischemic osteonecrosis of the right femoral head was produced in six to eight-week old piglets by surgically placing a ligature tightly around the femoral neck. The contralateral, left femoral head of each animal was used as a control. Four animals were euthanized at four and two at 8 weeks following the induction of ischemia. Femoral heads were harvested, processed and embedded in PMMA as described in detail earlier (REF). BMDD parameters were determined in the subchondral, central and physeal region of the epiphysis in a blinded fashion. Paired t-test using Graph Pad Prism 4.0 (San Diego, CA, USA) was used to evaluate statistical significance. The confidence level was set at 95%.

RESULTS:
Alterations in size, bone microarchitecture and tissue mineralization were clearly seen at 4 and 8 weeks post-surgery in the necrotic epiphysis (Fig.1-2). In the necrotic subchondral region (Fig.2), areas that are normally filled with living non-mineralized chondrocytes were fully mineralized even at 4 weeks post-surgery (Fig.2). QBEI measurements revealed, that in this region the degree of mineralization (CaMean and CaPeak) was increased by 23.5% (p<0.0003) and 17.4% (p<0.004) at 4 weeks and by 28.3 and 32.2% at 8 weeks, respectively. The heterogeneity of mineralization (CaWidth) was reduced by 34.1% (p<0.001) at 4 weeks and by 32.2% at 8 weeks. The amount of low mineralized bone (CaLow) was reduced by 52.5% (p=0.0007) at 4 weeks and 57.5% at 8 weeks (Fig.3). In the central and physeal region of the osteonecrotic heads, no significant differences were found at 4 weeks. Interestingly, increases in CaMean and CaPeak (5.1% and 4.3%), a 36% reduction in CaWidth and an 11% reduction in CaLow were found at 8 weeks, indicating time dependent changes in bone material properties in the necrotic femoral head.

DISCUSSION:
Our findings indicate for the first time that the mineralization process continues in necrotic calcified cartilage and bone following infarction, leading to significant changes in BMDD and therefore material properties. Since no new calcified cartilage and bone is formed, the amount of low mineralized tissue decreases. The necrotic tissue continues to mineralize leading to hypermineralization and a more homogenous mineralization, since the tissue fully mineralizes. Previous studies have shown, that hypermineralization leads to more brittle tissue, prone to micro-fractures, which could partly explain the deformation of necrotic bone following ischemic injury. Moreover, our findings could also explain the increased radiodensity seen on plain radiographs in the early stage of Legg-Calve-Perthes disease. Our findings have also implications of potential similar alterations in adult osteonecrosis.

REFERENCES:

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