INTRODUCTION:
Biaxial crest autograft is considered the gold standard graft material despite the disadvantages of extra surgery time and complications associated with the harvesting procedure. These complications have motivated clinicians and investigators to seek alternative graft materials to extend, enhance, and/or substitute for autograft tissue. Numerous alternatives include allografts, synthetic materials, and recombinant human bone morphogenetic proteins (BMPs). Several synthetic bone graft substitutes have been developed that are designed to address these limitations associated with using human donor material. Silicate-substituted graft materials represent a modification to the structural environment of osteoconductive graft materials that have demonstrated efficacy in ovine fusion models (Wheeler et al. Spine J 2007).

Concerns over tumorigenesis have spurred investigators to consider osteoconductive materials as alternatives to osteoinductive materials. Chemotherapy models have been accepted as considered to be a significant extension of the typical testing environment since they are a more challenging fusion environment and have the potential to examine the functional limits of graft materials.

The present study used a modified chemotherapy protocol described by Morcuende (OIJ, 2004) that employs multiple treatments of cisplatin and doxorubicin to slow the bone formation rate associated with healing grafts. The objective of this work was to evaluate the performance of silicate-substituted calcium phosphate (SiCaP-30, ApaTech Ltd., UK) relative to autograft and two commercially available bone void fillers in a posterolateral fusion model with concurrent administration of chemotherapeutic drugs.

METHODS:
Skeletally mature New Zealand White rabbits weighing 4.5-5.5 kg were studied and all procedures were approved by the Institutional Animal Care Use Committee at the University of Iowa (#1003068).

The rabbits received cisplatin and doxorubicin intravenously (2.5 mg/kg) seven days postoperatively and seven and twenty-one days postoperatively. Blood was collected from each rabbit prior to administration of cisplatin and doxorubicin. Complete blood chemistry profiles were used to assess the health status of the animals throughout the study. Rabbits in poor health were given 5% dextrose-Ringer’s solution intravenously and other supplemental nourishment.

A single level posterolateral intertransverse process fusion was performed in all rabbits. A dorsal midline incision, approximately 15 cm long, was made from L1 to the sacrum and the soft tissues overlying the transverse processes were dissected via separate bilateral fascial incisions. The transverse processes of L5 and L6 were decorticated with a long, was made from L1 to L6. At no time were the vertebral bodies decorticated in the gutter of the motion segment. For animals in the autograft group, approximately 2.5 to 3.0 cc of corticocancellous bone graft was obtained bilaterally from the iliac crest. This volume of graft was taken as the maximum amount which can be harvested from the iliac crest without significant animal morbidity. Investigational implant preparation of SiCaP-30, Actifuse® ABX (ApaTech Ltd., UK), and β-TCP/Bioactive Glass/Type I Collagen (βTCP-BG) was done according to the manufacturers’ instructions for use. Approximately 3.0 cc of implant material was placed bilaterally. Animals were euthanized at 12 weeks post-surgery.

Manual Palpation and Flexibility Analysis:
Manual palpation was used to determine fusion was manual palpation and flexibility analysis. Fusion was graded by three independent blinded observers as ‘fused’ if no detectable motion was present at the treated segment when tested in flexion and extension. The fusion was graded as ‘not fused’ if motion was present. Final results were determined by agreement of at least two of the three observers.

Biomechanical, non-destructive load testing was performed in flexion/extension to a pre-determined, sub-failure load. Flexibility tests were conducted among five pure moments (0 Nm, 0.09 Nm, 0.18 Nm, 0.27 Nm, and 0.36 Nm) using an MTS 858 Biomx testing system, two MTS spine gimbals (6 degrees of freedom devices) and an Optotrak motion analysis system. Stiffness was determined and compared to normal controls (10 normal rabbit lumbar columns).

RESULTS:
Rabbits were omitted from the study (N=16) due to complications arising from the surgical and chemotherapy interventions (graft harvest, low platelets, metabolic imbalances, infection). Eleven animals were assessed for each test group except for Actifuse® ABX (N=12).

Manual Palpation: Forty-five percent (45%) of the autograft implanted rabbits were fused at 12 weeks. The SiCaP-30 implanted rabbits achieved fusion in 82% of the animals, whereas Actifuse® ABX and βTCP-BG groups had significantly lower fusion rates (33% and 0%, respectively).

Flexibility Analysis:

<table>
<thead>
<tr>
<th>ROM</th>
<th>Autograft</th>
<th>βTCP-BG</th>
<th>Actifuse® ABX</th>
<th>SiCaP-30</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>15.467 (+1.19)</td>
<td>15.467 (+1.19)</td>
<td>15.467 (+1.19)</td>
<td>15.467 (+1.19)</td>
</tr>
<tr>
<td>Prearthritis</td>
<td>9.905 (+2.23)</td>
<td>6.939 (+1.15)</td>
<td>8.931 (+4.05)</td>
<td>7.58 (+0.88)</td>
</tr>
<tr>
<td>Fusion</td>
<td>4.515 (+2.14)</td>
<td>n/a</td>
<td>4.122 (+2.63)</td>
<td>2.805 (+1.21)</td>
</tr>
</tbody>
</table>

DISCUSSION:
Chemotherapeutic agents widely used for the treatment of cancereous lesions are known to delay or decrease the rate of bone healing. Several studies in humans and experimental animals have demonstrated that chemotherapeutic drugs can have a significant negative impact on fracture healing and limb-salvage procedures.

Synthetic bone graft substitutes based on hydrated calcium phosphate hydroxyapatite (HA; CaPO4.H2O) have been used in bone repair surgery for several years. A partial substitution of phosphate with silicate, within the HA lattice, results in a significant enhancement in protein adsorption and subsequent osteoblastic cell attachment and proliferation compared to stoichiometric HA (Guth et al. Adv Eng Mater 2010). Furthermore, silicate-substituted calcium phosphate appears to direct the differentiation of mesenchymal stem cells towards an osteogenic lineage (Noble et al. Bone 2010) and to be conducive in an ovine fusion model (Wheeler et al. Spine J 2007). In this investigation, two silicate-based calcium phosphate formulations were evaluated. SiCaP-30 has increased strut porosity compared to Actifuse® ABX.

In this study, there was a diminished rate of fusion in the autograft group compared to standard non-chemotherapy controls which is consistent with prior investigations utilizing chemotherapeutic models (Tortolani et al. Spine J 2004). Results from this study showed that SiCaP-30 produced a higher fusion rate than autograft, Actifuse® ABX, and βTCP-BG.

In comparison to autograft, SiCaP-30 was associated with improved bone healing, while Actifuse® ABX had similar healing, and βTCP-BG showed no evidence of bone healing. While further investigation is appropriate in higher animal species, this study demonstrates that material modifications to silicate-substituted graft materials in the form of surface physiochemistry may have a significant bearing on their bone healing characteristics in challenging fusion models.

SIGNIFICANCE:
This investigation further confirms the negative effect of chemotherapeutic agents on bone healing in a rabbit model. Diminished healing rates have been overcome in the past with synthetic BMPs (Singh et al. Spine J 2007). This study demonstrates that SiCaP-30 has a positive effect on graft bone healing in a rabbit chemotherapy fusion model.

ACKNOWLEDGEMENTS:
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