Introduction: The morbidity associated with harvesting autograft for spine arthrodesis is well-recognized, and it would be desirable if skeletal substitute materials could be used instead of autograft to promote spine fusion. Criteria for recognizing successful fusion are controversial. Radiographic studies of the fusion mass are important, but it may be difficult to distinguish bone mineral from residual synthetic calcium phosphate radiographically. The purpose of this study is to describe histological findings of biopsies obtained from patients who participated in a randomized clinical study that compared Osteogenetic Protein-1 (OP-1) with local autograft mixed with HA/TCP granules for spine fusion.

Methods: A total of 20 patients having an L3/4 or L4/5 degenerative spondylolisthesis with spinal stenosis were included in this study. Each patient underwent single level posterolateral fusion using pedicle screw instrumentation. Patients were randomly assigned to receive either OP-1 Putty (Stryker Biotech, Hopkinton, MA) alone, or a combination of local autograft with hydroxyapatite/tricalcium phosphate (HA/TCP) granules (Ceratite®, 70% HA and 30% β-TCP, Nippon Tokushu Togyo, Nagoya, Japan). One vial of the OP-1 Putty (3.5 mg OP-1/1g of collagen matrix) was used on each side of the posterolateral fusion in the OP-1 group, and a mixture of 5 grams of HA/TCP granules mixed with local autograft used on each side in the other group. Patients who were determined to have fused by radiographic studies underwent removal of pedicle screws, and with informed consent, a small biopsy was obtained from the fusion mass. Patients who did not show radiographic evidence of fusion did not undergo biopsy of fusion mass. Each specimen was fixed in formalin, decalcified, embedded in paraffin, sectioned, and stained with hematoxylin and eosin. Sections were viewed with a light microscope using transmitted and polarized light. The study protocol was fully approved by the institutional review board.

Results: As of June 2003, a total of 23 specimens obtained from sixteen patients (n=7 in OP-1 group, n=9 in the control group) were available for evaluation. There were 9 males and 7 females, with an average age of 65 years (49 to 77 years). Although the study was randomized, mean age was significantly higher in the OP -1 group (mean 70, range 51 -77) than the control group (mean 58, range 43-71, p=0.02). One patient refused to complete the study. Surgical exploration: At least one year postoperatively (12-32 months), seven patients of the OP -1 group (78%) and nine patients (90%) of the control group showed radiographic evidence of fusion (less than 5 degrees of anular motion and less than 2mm of translation). Instrumentation removal and surgical exploration were performed and a completely solid arthrodesis was observed in four cases of the OP-1 group and seven cases of the HA/TCP control group.

HA/TCP control group (9 patients, 10 specimens): Residual biomaterial was identified in each of the cases treated with HA/TCP mixed with autograft. Although HA and TCP cannot be distinguished by light microscopy alone, we would anticipate that the TCP had dissolved (or had been resorbed), and the HA was still present. Because it was present after one year, the HA was probably of high crystallinity. The pieces of HA ranged up to 1 mm in diameter, while the individual crystals of HA within each granule appeared to have ranged up to approximately 10 µm in diameter. In some areas the residual HA showed extensive bone apposition, but in other areas the granules were surrounded by fibrous tissue (Fig. 1). Giant cells were also present around the HA granules in some areas. There was no acute or chronic inflammation associated with the calcium phosphate granules. Each of the biopsies in this group also contained pieces of necrotic bone, sometimes associated with new bone in a pattern that suggests gradually incorporating bone graft.

OP-1 group (7 patients, 13 specimens): Histologically viable bone was identified in each of the specimens from the OP-1 group (Fig. 2). The bone is histologically unremarkable and, based on its microscopic appearance, is expected to have normal mechanical properties. Residual OP-1 carrier (bovine collagen) was recognized by light microscopy in only one case. One of the biopsies in the OP-1 group contained slight chronic inflammation of uncertain significance, but no acute inflammation was present in any biopsy. Necrotic bone was minimal in the OP-1 treated group. One of the OP-1 specimens contained evidence of hyaline cartilage surrounded by bone in a pattern that suggested a late stage of remodeling of bone of endochondral origin.

Discussion: It may be difficult to distinguish bone from residual synthetic calcium phosphates when radiographs are used as an outcome measure in fusion studies. The removal of hardware one year after surgery in this prospective, randomized study, provides an opportunity to document the histological findings in each group. Granules of residual hydroxyapatite were still present even at one year in all nine patients who had been treated with the combination of HA/TCP granules and autograft. Some of the granules were surrounded by bone; others were surrounded by soft tissue, as might be expected when osteoconductive granules are used in this clinical application. To our knowledge, the current investigation serves as the first report of surgical exploration and histological assessment of a human instrumented posterolateral lumbar fusion using bone morphogenetic protein. Biopsies from patients treated with the OP-1 showed evidence of viable bone, with rare recognizable residual OP-1 and no significant inflammatory reaction. The safety and efficacy of each treatment will require completion of the prospective clinical study, but the histologic demonstration of essentially normal bone in the OP-1 group is encouraging.