

INTERNATIONAL
JOURNAL
of
SPINE
SURGERY

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Int J Spine Surg 2016, 10 ()

doi: <https://doi.org/10.14444/3035>

<https://www.ijssurgery.com/content/10/35>

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Outcomes of Demineralized Bone Matrix Enriched with Concentrated Bone Marrow Aspirate in Lumbar Fusion

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Abstract

Background

Multiple studies have demonstrated that a significant amount of variability exists in various demineralized bone matrix (DBM) formulations, which casts doubts on its reliability in consistently promoting fusion. Bone marrow aspirate (BMA) is a cellular based graft that contains mesenchymal stem cells (MSCs) and growth factors can confer osteogenic and osteoinductive potential to DBM. The goal of this study was to describe the outcome of DBM enriched with concentrated BMA in patients undergoing combined lumbar interbody and posterolateral fusion.

Methods

Eighty patients with a minimum of 12 months of follow-up were evaluated. Fusion and rates of complication were evaluated. Functional outcomes were assessed based on the modified Odom's criteria. Multiple logistic regression analysis was used to examine the effects of independent variables on fusion outcome.

Results

The overall rate of solid fusion (i.e patients with both solid posterolateral and interbody fusion) was 81.3% (65/80). Specifically, the radiographic evidence of solid posterolateral and interbody fusions were 81.3% (65/80) and 92.5% (74/80), respectively. Seven (8.75%) patients developed hardware-related complications, 2 (2.5%) patients developed a postoperative infection and 2 (2.5%) patients developed clinical pseudarthrosis. Charlson comorbidity index (CCI) scores of 3 and 4 were associated with non-solid unions (CCI-3, $p = 0.048$; CCI-4, $p = 0.03$). Excellent or good outcomes were achieved in 58 (72.5%) patients.

Conclusions

Patients undergoing lumbar fusion using an enriched bone graft containing concentrated BMA added to DBM can achieve successful fusion with relatively low complications and good functional outcomes. Despite these findings, more studies with higher level of evidence are needed to better understand the efficacy of this promising graft option.

KEYWORDS: LUMBAR FUSION; DEMINERALIZED BONE MATRIX; BONE MARROW ASPIRATE

VOLUME 10 ARTICLE 35 DOI: 10.14444/3035

Introduction

Transforaminal lumbar interbody fusion (TLIF) is often used in the management of spine pathologies including degenerative disk disease, deformity, trauma, severe instability, spondylolisthesis and pseudarthrosis. Several fusion aides are available to achieve a successful arthrodesis. These include autograft, allograft, bone graft enhancers, extenders and substitutes. Iliac crest bone graft (ICBG) offers good fusion success rates and remains the "gold standard". However, it is linked with certain potential morbidities (such as donor site pain, hematoma,

paresthesia and infection) that limit its use in spinal surgery.¹ Demineralized bone matrix (DBM) is an osteoinductive allograft. It has been estimated that 93% of DBM consists of a weakly osteoconductive collagen scaffold, while the remainder of its components are osteoinductive factors such as bone morphogenetic proteins (BMPs) 2 and 7.² Multiple studies have reported variation between formulations of different DBM products as well as variation between lots of the same DBM products.³⁻⁵ This variability casts doubts on the reliability and efficacy of DBM in consistently promoting fusion.

Bone marrow aspirate (BMA) is a cellular based graft that contains mesenchymal stem cells (MSC) and growth factors (such as BMPs 2 and 7) that can confer osteogenic and osteoinductive potential to osteoconductive scaffolds.⁶ BMA can be obtained via a relatively non-invasive method from the iliac crest during spine surgery and combined with DBM. The combination of BMA with DBM and allograft chips in a selective retention technology chamber has been shown to result in a graft matrix enriched with a 3- to 5-fold increase in osteoprogenitor cells.⁷⁻¹⁰ Furthermore, the addition of osteoinductive factors derived from BMA to a graft material of DBM may negate the effects of unreliable concentrations of BMPs 2 and 7 in commercially available DBM products.⁶ To date, there is a dearth of literature describing the outcomes of DBM enriched with concentrated BMA in combined PLF and TLIF. Most of the published literature on outcomes of DBM has been described in patients undergoing anterior cervical discectomy and fusion (ACDF) and posterolateral lumbar fusion only.

The aim of this study was to describe the clinical and radiographic outcomes of an enriched graft material of concentrated BMA added to DBM/allograft chips in patients undergoing combined TLIF and PLF.

Materials & Methods

After obtaining Institutional Review Board approval, a search of the medical record database for patients that underwent lumbar spine fusion by a single surgeon was performed for the period between May 2000 and January 2011. All patients in the study consented to surgery and the use of bone grafting.

Patients were included if they underwent a combined instrumented PLF and TLIF for the treatment of degenerative lumbar spine diseases with concomitant instability, if their procedure utilized concentrated BMA with DBM, and if there was at least 12 months of follow-up. Patients were excluded if they had a diagnosis of tumor, infection or fracture, underwent a posterolateral lumbar fusion alone, anterior lumbar interbody fusion, an uninstrumented fusion, or had less than 12 months of follow-up. Of the 157 patients that had combined PLF and TLIF, 73 were excluded

because their surgery involved other bone graft options such as autograft, allograft, bone morphogenetic protein-2. Of remaining 84 patients that had PLF and TLIF using BMA with DBM, 4 were excluded for less than 12 months of follow-up, leaving a total of 80 patients for inclusion in this study.

Using the properties of an affinity column, bone marrow aspirates were processed with a selective cell retention system (CELLECT, DePuy Spine, Raynham, MA).¹¹ Briefly, bone marrow aspirates were obtained from a single iliac crest using a 3-ported Jamshidi needle. Heparin was used in each syringe to inhibit clotting. In an effort to avoid dilution of the bone marrow with whole blood, 2 mL of bone marrow was obtained from each of the ports, and then the needle was advanced to new bone for additional draws of marrow. The total volume of bone marrow aspirates varied depending on the amount of graft material to be populated and was generally a ratio of 3 (aspirate volume):1 (graft material). The bone marrow was then run through the graft chamber of the selective cell retention device, which contained DBM and allograft chips, twice in accordance with the manufacturer's instructions. The enriched graft material was then placed in an interbody cage device and over the decorticated surfaces in the posterolateral gutters.

Information such as patient demographics, surgical and clinical data were obtained from the medical records. Demographic data included age and gender. Clinical data included smoking status, co-morbidities, length of hospital stay, mean follow-up time, radiographic fusion grade, complications and functional outcomes. Co-morbidities were graded based on the Charlson co-morbidity index (CCI) scores.¹² Smokers were defined as those who had continuously smoked for at least 1 year prior to surgery, as well as post-operatively. Surgical data included primary versus revision surgery, number of levels fused and estimated blood loss. Functional outcomes were assessed based on a 4-grade system: excellent, good, satisfactory and poor, in accordance with the modified Odom's criteria.¹³ Anterior-posterior (AP) and dynamic lateral radiographs were obtained in all patients as part of routine clinical follow-up. Posterolateral radiographic fusion was based on a 4-grade system, as defined by the Lenke

classification: Grade A (definitely solid), Grade B (possibly solid), Grade C (probably not solid) and Grade D (definitely not solid).¹⁴ Interbody radiographic fusion was based on a 3-grade system as defined by the Brantigan, Steffee, Fraser (BSF): BSF-1 (radiographic pseudarthrosis), BSF-2 (radiographic locked pseudarthrosis) and BSF-3 (radiographic fusion).¹⁵ A fellowship-trained and board-certified musculoskeletal radiologist graded fusion status based on radiographs. In cases of multilevel fusion with varying grades of fusion, the least fused segment was used to assign the final fusion grade. A solid fusion was defined as the concomitant presence of both Lenke A posterolateral and BSF-3 interbody fusion grades in the same patient (Figure 1). Radiographic pseudarthrosis was defined as the presence of Lenke D or BSF-1 fusion grade. The diagnosis of clinical pseudarthrosis was based on motion or non-union detected during revision surgery.

All statistical analyses were performed with R (version 3.0.2, R Foundation for Statistical Computing, Vienna, Austria). Fisher's exact test was used to assess differences in fusion outcomes based on single versus multilevel fusion. Multiple logistic regression analysis was used to examine the effects of variables such as patient age, gender, smoking, primary versus revision surgery, number of spine levels fused, length of hospital stay, length of follow-up, and Charlson comorbidity index score on fusion outcome. All analyses with a *p* value of less than 0.05 were considered statistically significant.

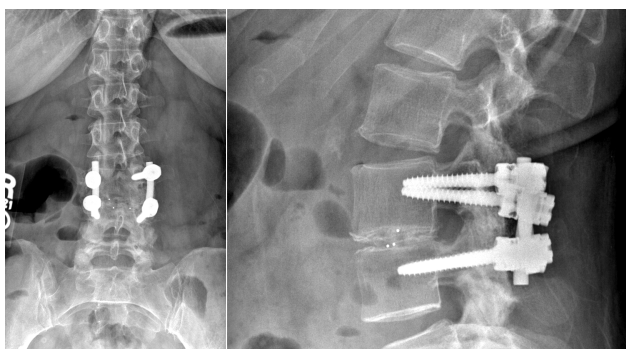


Fig. 1. Anterior-posterior and lateral radiograph showing solid posterolateral (Lenke A) and interbody (BSF-3) fusion in a 33 year-old female 1.5 years following lumbar 3-4 posterolateral and transforaminal lumbar interbody fusion using bone marrow aspirate/demineralized bone.

Results

Demographic and clinical data

The study group consisted of 80 patients (29 men and 51 women) with a mean age of 58.1 years (range = 16-80 years) and a mean follow-up of 33.2 months (range = 12-89.6 months) (Table 1). The mean CCI score was 2.1 (range = 0-9), the mean number of fused spine level was 1.3 (range = 1-3), the mean estimated blood loss was 252.2 mL (range = 20-900mL) and the mean length of hospital stay was 5.2 days (range = 4-16 days). 60 (76.3%) patients were undergoing primary surgery and there were 8 (10%) smokers (Table 1).

Radiographic outcomes

The overall rate of solid fusion (i.e patients with Lenke A and BSF-3 fusion grades) was 81.3% (65/80). Specifically, the radiographic evidence of solid posterolateral (Lenke A) and interbody fusions (BSF-3) were 81.3% (65/80) and 92.5% (74/80), respectively (Table 2). In single-level fusions, solid posterolateral and interbody fusions were 79.7% (47/59) and 93.2% (55/59), respectively. In multi-level fusions, solid posterolateral and interbody fusion were 85.7% (18/21) and 90.5% (19/21), respectively (Table 3).

Table 1. Demographic, clinical and surgical variables.

	BMA (n = 80)
Mean age, SD* (range)	58.1 +/- 1.8 (16-80)
Gender	
Male (%)	29 (36.3%)
Female (%)	51 (63.7%)
Mean Charlson co-morbidity index score (range)	2.10 (0-9)
Smoker (%)	8 (10%)
Primary surgery (%)	60 (75%)
Mean estimated blood loss in milliliters (range)	252.2 (20-900)
Mean number of fused spine levels (range)	1.3 (1-3)
Mean number of hospital stay in days (range)	5.2 (4-16)
Mean follow-up time in months (range)	33.2 (12-89.6)

*SD (standard deviation).

Using logistic regression analysis, CCI scores of 3 and 4 were associated with non-solid unions (CCI-3, CI: -7.28 to -0.03, $p = 0.048$; CCI-4, CI: -9.70 to -0.46, $p = 0.03$). None of the other variables were associated with non-solid unions (age, CI: -0.009 to 2.07, $p = 0.07$; gender, CI: -2.60 to 0.58, $p = 0.21$; smoking, CI: -2.10 to 2.97, $p = 0.73$; revision surgery, CI: -1.12 to 2.04, $p = 0.57$; multilevel fusion, CI: -1.22 to 2.56, $p = 0.48$; length of hospital stay, CI: -0.50 to 0.63, $p = 0.82$; length of follow-up time, CI: -0.02 to 0.06, $p = 0.34$).

Complications

In the study group, 7 (8.75%) patients developed hardware-related complications (in the form of hardware bursitis), 2 (2.5%) patients developed a postoperative infection, 2 (2.5%) patients developed pseudarthrosis and none of the patients developed

Table 2. Fusion grade.

	BMA (n = 80)
Overall fusion	
Both Grade A and BSF 3	65 (81.3%)
Posterolateral fusion	
Grade A (%)	65 (81.3%)
Grade B (%)	5 (6.3%)
Grade C (%)	9 (11.3%)
Grade D (%)	1 (1.3%)
Interbody fusion	
BSF-1 (%)	3 (3.8%)
BSF-2 (%)	3 (3.8%)
BSF-3 (%)	74 (92.5%)

Table 3. Solid fusion (Lenke A and BSF-3) based on the number of fused levels.

	BMA (n = 80)	p value
Posterolateral fusion		0.75
Single-level (%)	47/59 (79.7%)	
Multi-level (%)	18/21 (85.7%)	
Interbody fusion		0.65
Single-level (%)	55/59 (93.2%)	
Multi-level (%)	19/21 (90.5%)	

graft donor-site morbidities (Table 4).

Functional outcomes

Using the modified Odom's criteria, excellent or good results were achieved in 58 (72.5%) patients in the study group (Table 5). Solid fusion was achieved in 44/58 (75.9%) of patients with excellent or good outcomes and 21/22 (95.4%) of patients with satisfactory or poor compared to ($p = 0.06$).

Discussion

Combined PLF and TLIF techniques have traditionally used ICBG as the gold standard for achieving successful fusion. Due to the potential morbidities (such as donor site pain, paresthesia, hematoma, and infection) of ICBG harvest and limited graft supply in revision and multilevel fusion surgeries, there is continued interest in the spine surgery community for alternative bone graft options to aide with spine fusion.¹ In this study, we found that patients undergoing combined TLIF and PLF using an enriched graft material containing concentrated BMA and DBM can achieve successful fusion with relatively low complications and good functional outcomes. To date, this is the largest study to report on the outcome of this graft option in patients undergoing combined PLF and TLIF.

There is a dearth of evidence in the literature describing the outcomes of DBM with concentrated BMA in combined PLF and TLIF. Most of the published literature on outcomes of DBM has been de-

Table 4. Complications.

	BMA (n = 80)
Hardware-related complications	7 (8.75%)
Infection	2 (2.5%)
Clinical pseudoarthrosis	2 (2.5%)
Graft donor-site morbidity	-

Table 5. Functional outcomes by the modified Odom's criteria.

	BMA (n = 80)
Excellent or Good (%)	58 (72.5%)
Satisfactory (%)	11 (13.7%)
Poor (%)	11 (13.7%)

scribed in patients undergoing ACDF and lumbar PLF only. In addition, only one study has been published that utilized DBM enriched with concentrated BMA. In the cervical spine, DBM with autograft was shown to achieve fusion rates ranging from 88.9% - 97% in patients undergoing anterior cervical discectomy and fusion (ACDF).¹⁶⁻¹⁸ In the lumbar spine, prospective controlled trials by Cammisa et al. and Sassard et al. reported disappointing fusion rates of 52% and 60%, respectively in patients undergoing PLF alone with DBM and autograft.^{19,20} Conversely, other studies have described better success with DBM and autograft in the lumbar spine with fusion rates ranging from 70% - 91% in patients undergoing PLF alone. To the best of our knowledge, only two studies in the English literature have been published describing the outcomes of DBM in combined PLF and TLIF. In a comparative study by Ahn et al., DBM with autograft was compared with autograft alone in patients undergoing combined PLF and TLIF.²¹ At two years of follow-up, fusion success was 86% in patients in the DBM and autograft group compared to 87% in the autograft alone group. In a similar study by our group examining the outcome of DBM with BMA in combined PLF and TLIF in elderly patients (≥ 65 years of age), the fusion success rate was 83.9%.²² The fusion success rate observed in both studies is comparable to the rate of 81.3% observed in the current study.

There are several factors that are responsible for the failure to consistently achieve successful union with DBM in spinal fusions. As previously discussed, there is an unpredictable degree of variability both the concentrations of BMP among different lots of the same DBM formulation and the concentrations of BMP among different DBM formulations. Bae et al. showed that the variability of BMP concentrations among different lots of the same DBM formulation was higher than the variability of concentrations among those of different DBM formulations.⁵ In a separate in-vivo animal study by Bae et al., the variability of BMP concentrations in commercially available DBM products was noted to correlate with fusion success, which has been described to range from 0% to 75%.⁴ Additionally, the method of processing DBM and the type of carrier combined with DBM are also thought to contribute to the variability of

DBM in consistently promoting successful fusion.^{3,23,24} Lastly, the type of fusion construct may also play a role in fusion success with DBM. It is possible that the addition of an increased surface area for bone graft incorporation (in the form of an interbody fusion) contributes to a more stable construct required to promote the vascular ingrowth and cellular events needed to optimize the osteoinductive and osteogenic effects of concentrated bone marrow aspirate on bone fusion.

This current study is unique in that it utilizes a concentrated formulation of BMA to create an enriched graft material of DBM. It has been estimated that the proportion of osteogenic progenitors in bone marrow is between 1 in 5000 to 1 in 100,000 cells.²⁵ During the process of bone marrow aspiration, it has been shown that the concentration of bone-marrow derived cells (including osteoprogenitors) in an aspirate is diluted rapidly by peripheral blood.²⁶ The technique of concentrating BMA was developed as a practical means of delivering an increased number of osteogenic progenitor cells added to a graft site. The combination of BMA with DBM and allograft chips in a selective retention technology chamber has been shown to result in a graft matrix enriched with a 3- to 5-fold increase in osteoprogenitor cells.⁷⁻¹⁰ In a canine model of posterolateral lumbar fusions, Muschler et al. reported superior union rates with DBM enriched with concentrated BMA compared to DBM alone or DBM with non-concentrated BMA.^{10,27}

The only patient characteristics noted to have an association with non-solid unions in this study were CCI scores of 3 and 4. CCI scores, which take into account patient age and the presence or absence of 19 comorbidities, was initially developed in 1987 to predict 1-year mortality after an inpatient hospitalization.²⁸ It has since been validated across multiple medical and surgical sub-specialties. In the spine literature, CCI scores have been found to predict mortality, complications, and re-admissions after spine surgery.²⁹⁻³¹ It is not surprising that CCI scores are associated with decreased union rates in this study. For example, patients with multiple co-morbidities such as metabolic syndrome often have vascular diseases that may limit neo-angiogenesis and blood flow to a fusion site. In addition, advanced age, which is taken

into consideration when generating a CCI score is also associated with poor bone quality, which may hinder fusion potential.³²

Relatively low rates of complications were observed in this study. Implant-related complication in the form of hardware bursitis was the most common complication observed. Persistent low back pain despite a solid fusion in the absence of any obvious pain generator remains a challenging clinical scenario. In some of these patients, implant removal and exploration of fusion may be an appropriate option to alleviate their pain although this decision remains controversial due to the unknown mechanism of how implants generate pain.

Lastly, good or excellent results were achieved in the majority of patients in this study as measured by the modified Odom's criteria.

This study has several obvious limitations. Its retrospective design, possible selection bias associated with patient follow-up, and lack of a comparative group limits the generalizability of the results. Randomized control trials would be required to make more definitive conclusions about fusion rates, complications and functional outcomes of DBM enriched with concentrated BMA. In addition, cellular analysis of the bone marrow aspirate was not conducted on each patient. As a result, it is not possible to quantify the amount of osteoprogenitor cells in each aspirate. Another major limitation of this study is the method of radiographic assessment of fusion status. CT scan has been demonstrated to be more sensitive than radiographs for assessing fusion following instrumented lumbar fusion.^{33,34} Despite these limitations, this study is the largest of its kind and it provides valuable information and insight on a promising bone graft option for spinal fusion.

Conclusion

In summary, we found that patients undergoing combined PLF and TLIF with DBM enriched with concentrated BMA can achieve successful fusion with relatively low complications and good functional outcomes. Given the concerning potential adverse effects associated with ICBG donor-site morbidity,

concentrated BMA with DBM may be an appropriate bone graft option for patients undergoing combined posterolateral and interbody fusions for degenerative lumbar spine disease. Despite these findings, more clinical studies with higher level of evidence are needed to better understand the efficacy of this promising graft option.

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Disclosures & COI

No funds were received in support of this work. No relevant financial disclosures are associated with this work.

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Published 17 October 2016.

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