Anterior Cervical Discectomy and Fusion Using *Escherichia coli*-Derived Recombinant Human Bone Morphogenetic Protein-2: A Pilot Study

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**Background:** Recombinant human bone morphogenetic protein-2 (BMP-2) is an osteoinductive growth factor widely used in orthopedic surgery; it is also known to be associated with postoperative airway compromise or dysphagia when applied to anterior cervical disectomy and fusion (ACDF). However, there have been no reports on ACDF using *Escherichia coli*-derived BMP-2 (E.BMP-2) with hydroxyapatite (HA). This pilot study aimed to investigate the potential efficacy and safety of E.BMP-2 using HA as a carrier in ACDF prior to designing a larger-scale prospective study.

**Methods:** Patients eligible for inclusion were those who underwent ACDF using 0.3 mg of E.BMP-2 with HA per segment for degenerative cervical disc disease between August 2019 and July 2020 and had at least 1 year of follow-up. Fusion rates were analyzed using computed tomography or flexion-extension radiographs. Visual analog scales for neck pain and arm pain and neck disability index were measured preoperatively and the final follow-up. In cases of cervical spondylotic myelopathy, modified Japanese Orthopaedic Association scores were also evaluated. Postoperative complications such as airway compromise, dysphagia, wound infection, neurologic deficit, hoarseness, heterotopic ossification, seroma, and malignancy were investigated.

**Results:** A total of 11 patients and 21 segments were analyzed. All clinical outcomes significantly improved at the final follow-up compared with the preoperative indices (p < 0.05). Only 1 case of dysphagia and no cases of airway compromise, wound infection, neurologic deficit, hoarseness, heterotopic ossification, seroma, or malignancy were observed during the follow-up period. Of the 21 segments, 15 segments showed solid fusion at 3 months after surgery, 4 segments at 6 months, and 1 segment at 12 months. Only 1 segment showed pseudoarthrosis, resulting in a fusion rate of 95.2%.

**Conclusions:** The outcomes of ACDF could be enhanced using 0.3 mg of E.BMP-2 with HA per segment. Based on this study, larger-scale prospective studies can be conducted to evaluate the efficacy and safety of E.BMP-2 in ACDF.

**Keywords:** Degenerative cervical disc disease, Anterior cervical disectomy and fusion, *Escherichia coli*-derived bone morphogenetic protein-2, Prevertebral soft tissue swelling
Anterior cervical discectomy and fusion (ACDF) is a highly effective surgical technique for degenerative cervical disc diseases, such as cervical spondylotic myelopathy (CSM) or radiculopathy.\(^1,2\) Although autologous iliac bone grafts (IBG) is the gold standard for ACDF, there are several limitations regarding donor site morbidity. To replace autologous IBG, different types of bone substitutes or growth factors have been introduced.\(^3,4\) Although several bone graft materials have been used, fusion rates in multi-level ACDF is still not satisfactory.\(^5\)

Recombinant human bone morphogenetic protein-2 (BMP-2) is an osteoinductive growth factor widely used in orthopedic surgery. The efficacy of BMP-2 in ACDF has been reported in several studies; however, it is also known to be associated with prevertebral soft-tissue swelling after surgery, resulting in airway compromise or dysphagia.\(^3,6\) In 2008, the U.S. Food and Drug Administration (FDA) issued a public health notification of the life-threatening complications associated with BMP-2 in ACDF.\(^7\) Nevertheless, BMP-2 has been used off-label to promote fusion in patients who underwent ACDF in the United States: 16.73% in 2007 and 12.01% in 2011.\(^8\)

Initially, Chinese hamster ovary cell-derived BMP-2 (CHO–BMP-2) was commonly used. Recently, Escherichia coli-derived BMP-2 (E.BMP-2) has been introduced to overcome the high cost and low yield of CHO–BMP-2.\(^9\) Several studies have reported the efficacy and appropriate dose of E.BMP-2 in lumbar spinal surgery to increase fusion rates.\(^10-12\) It can be speculated that a risk of prevertebral soft-tissue swelling after ACDF using E.BMP-2 with hydroxyapatite (HA) as a carrier may be lower than with a collagen carrier. Due to its high affinity for E.BMP-2, HA may be able to prevent the initial burst release of E.BMP-2, which can lead to prevertebral swelling.\(^13\) However, the results of using E.BMP-2 with HA in ACDF have not yet been reported. This pilot study aimed to investigate the efficacy and safety of E.BMP-2 using HA as a carrier in ACDF prior to designing a larger-scale prospective study.

### METHODS

**Patient Enrollment**

The Institutional Review Board of Seoul National University Hospital approved this pilot study (No. H-1510-127-715) and informed consent was obtained from the enrolled patients. Patients who underwent ACDF using 0.3 mg of E.BMP-2 with HA per segment for degenerative cervical disc disease from August 2019 to July 2020 and had at least 1 year of follow-up were eligible for inclusion. Patients who underwent surgery for trauma, spinal tumors, or infectious diseases or had a history of cervical fusion were excluded. In this study, 11 patents and 21 segments were enrolled.

**Surgical Procedures**

Under general anesthesia, the Smith-Robinson technique was utilized for an anterior approach.\(^14\) When the anterior aspect of the vertebral column was exposed, the appropriate level(s) was confirmed by simple radiography. Following discectomy, cartilaginous endplates were removed by using a high-speed burr to promote fusion. Uncinectomy or removal of the ossification of the longitudinal ligament was performed, if needed. A tricortical bone block with a series of drill holes or a polyetheretherketone cage (Wave Cage; CGBio Inc., Seongnam, Korea) was inserted for interbody fusion. The bone block and cage were each filled with morselized bone obtained through a decompression procedure and 0.3 mg of E.BMP-2 (Novosis, CGBio Inc.) with HA per segment (Fig. 1). Additionally, morselized bone was grafted around the inserted bone block or cage. The anterior cervical plate was then fixed to increase stability (Fig. 2). To minimize postoperative neck swelling, oxidized cellulose (Surgicel; Ethicon, Somerville, NJ, USA) and fibrin sealants (Greenplast Q; GC Pharma, Yongin, Korea or Tisseel; Baxter, Deerfield, IL, USA) were applied over the plate.\(^15,16\) A drain was placed in all patients. In addition, 5 mg of dexamethasone was administered intraoperatively at the discretion of the surgeon immediately after surgery, considering the risk factors for neck swelling.\(^17,18\) Postoperatively, a rigid cervical orthosis was applied to each patient for 6 weeks. A senior spine surgeon (HK) at a single center performed all surgical procedures under intraoperative neuromonitoring.

**Outcome Measures**

The visual analog scale for neck pain and arm pain and neck disability index were scored preoperatively and at

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**Fig. 1.** (A) Hydroxyapatite sufficiently absorbing *Escherichia coli*-derived bone morphogenetic protein-2 (E.BMP-2) solution for 10 minutes. (B) Polyetheretherketone cage filled with morselized bone and 0.3 mg of E.BMP-2.
the final follow-up. In the case of CSM, modified Japanese Orthopaedic Association scores were also evaluated. Postoperative complications such as airway compromise, dysphagia, wound infection, neurologic deficit, hoarseness, heterotopic ossification, seroma, and malignancy were investigated.

Fusion rates were evaluated using computed tomography (CT) or flexion-extension radiographs at 3 months, 6 months, or 1 year after surgery. If bone union was confirmed at one time point, no additional CT or flexion-extension radiography procedures were performed. Union was defined as the presence of bridging trabecular bone between the endplates in CT or the absence of a maximum of > 2 mm of motion between the spinous processes at the operated level(s) in flexion-extension radiographs.\(^{19}\)

Cage subsidence was defined as a loss of anterior disc height (ADH) or posterior disc height (PDH) of at least 2 mm between the immediate postoperative and final follow-up lateral radiographs (Fig. 3).\(^{20}\) Cervical alignment was measured using Cobb’s angle between C2 and C7 and segmental angle between the superior endplate of the upper vertebra and the inferior endplate of the lower vertebra at an operated level(s) in the postoperative and final follow-up lateral radiographs. A spine fellow (HJS), who did not participate in the surgery, analyzed all radiological data.

Statistical Analysis
IBM SPSS ver. 25.0 (IBM Corp., Armonk, NY, USA) was used for the statistical analysis. The Wilcoxon signed-rank test was used to assess clinical and radiological outcomes. Statistical significance was set at \(p < 0.05\).

RESULTS
The patients’ demographic data and baseline characteristics are presented in Table 1. The average patient age was 57.7 years, and the mean follow-up period was 15.0 months. All clinical outcomes significantly improved at the final follow-up compared with the preoperative indices (Table 2). Although several cases showed moderate to severe airway swelling in simple radiographs taken 1 to 3 days after surgery (Fig. 4), there was only 1 case of dysphagia at the time point of discharge (patient 11), 1 case of cerebrospinal fluid leakage (patient 11), and 1 case of pneumomediastinum (patient 4), which improved with
conservative treatment without readmission or reoperation. However, no cases of airway compromise, wound infection, neurologic deficit, hoarseness, heterotopic ossification, seroma, or malignancy were observed during the follow-up period.

Of the 21 segments, 15 segments showed solid fusion at 3 months after surgery, 4 segments at 6 months, and 1 segment at 12 months. Only 1 segment showed pseudoarthrosis, resulting in a fusion rate of 95.2%. Although ADH and PDH significantly decreased postoperatively and at the final follow-up, the differences in ADH and PDH were 0.7 ± 0.7 and 0.4 ± 0.6, respectively; thus, no cases of cage subsidence were found. No significant differences were observed in postoperative and final cervical alignments and segmental angles (Table 3).

**DISCUSSION**

BMP-2, which has a striking osteoinduction effect, is widely used in spine fusion surgery, including ACDF, to overcome the shortcomings of autologous IBG. Frenkel et al. reported that the overall fusion rate for the patients who underwent multilevel ACDF without CHO–BMP-2 was 82.6% compared with a 100% fusion rate in the group with 0.2–0.4 mg of CHO–BMP-2 per level. In a prospective study by Buttermann, ACDF grafting of 0.9 mg of CHO–BMP-2 per segment with allograft was as effective as autologous IBG in terms of clinical outcomes and fusion rates.

However, there have been several reports regarding the life-threatening complications of BMP-2 in ACDF resulting from postoperative neck swelling. Shields et al. reported complications of 23.2% after the use of high-dose CHO–BMP-2 (2.1 mg/level) and 8.6% of prolonged hospital stay or readmission due to dysphagia or airway compromise. A meta-analysis showed that the use of BMP-2 is

| Table 1. Demographics and Baseline Characteristics of the Patients |
| --- |
| No. | Sex | Age (yr) | BMI (kg/m²) | BMD (g/cm²) | Diagnosis | Fusion level | Fusion time (mo) | Risk factor of pseudoarthrosis | Additional procedure | Use of dexamethasone | Follow-up period (mo) |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| 1 | Female | 78 | 21.8 | −2.0 | CSM | C3–5 | 3 (C3–4), nonunion (C4–5) | - | - | 5 mg once | 18 |
| 2 | Male | 71 | 22.0 | −0.8 | CSM | C4–6 | 3 | DM, CKD | - | 5 mg once | 20 |
| 3 | Male | 45 | 32.9 | 0.6 | CSM | C4–7 | 3 | - | Uncinctectomy | - | 21 |
| 4 | Female | 56 | 25.9 | −0.1 | CSMR | C5–7 | 3 | - | Uncinctectomy | - | 20 |
| 5 | Male | 41 | 37.5 | 0.4 | CSMR | C6–7 | 6 | Smoking | Uncinctectomy | 5 mg once | 14 |
| 6 | Male | 47 | 21.4 | - | CSR | C5–7 | 3 | - | Uncinctectomy | 5 mg once | 12 |
| 7 | Female | 48 | 32.5 | - | CSM | C5–6 | 6 | DM | - | - | 12 |
| 8 | Female | 62 | 27.8 | 0.0 | CSMR, OPLL* | C5–6 | 12 | - | Uncinctectomy, removal of OPLL | 5 mg once | 12 |
| 9 | Female | 73 | 30.0 | −2.0 | CSM | C5–7 | 3 | - | - | 5 mg once | 12 |
| 10 | Male | 54 | 24.7 | 0.6 | CSMR | C4–6 | 3 | DM, smoking | Uncinctectomy | - | 12 |
| 11 | Male | 60 | 23.5 | 0.4 | CSM, OPLL* | C3–6, 3 (C3–4), 6 (C4–6) | Smoking | Floating of OPLL | 5 mg once | 12 |

BMI: body mass index, BMD: bone mineral density, CSM: cervical spondylotic myelopathy, DM: diabetes mellitus, CKD: chronic kidney disease (estimated glomerular filtration rate < 60 mL/min/1.73 m²), CSMR: cervical spondylotic myeloradiculopathy, CSR: cervical spondylotic radiculopathy, OPLL: ossification of the posterior longitudinal ligament.

*Segmental OPLL.

| Table 2. Clinical Outcomes |
| --- |
| Variable | Preoperative | Final follow-up | p-value |
| --- | --- | --- | --- |
| VAS-NP | 6.8 ± 2.0 | 3.4 ± 1.6 | 0.005* |
| VAS-AP | 5.9 ± 2.5 | 2.0 ± 1.9 | 0.005* |
| NDI | 29.5 ± 5.9 | 22.4 ± 5.5 | 0.033* |
| mJOA score | 12.8 ± 1.1 | 15.4 ± 3.0 | 0.049* |

Values are presented as mean ± standard deviation. VAS-NP: visual analog scale-neck pain, VAS-AP: visual analog scale-arm pain, NDI: neck disability index, mJOA: modified Japanese Orthopaedic Association.

*Significant difference.
a risk factor for dysphagia after ACDF, with an odds ratio of 5.52.\(^1\) However, the incidence of dysphagia or hoarseness was not statistically different in 215,047 patients who used BMP or not in a U.S. database review. Despite reports from the U.S. FDA considering these complication rates, BMP-2 is still used off-label in ACDF.\(^8\)

Recently, E.BMP-2 has been used to overcome the limitations of the high cost and low yield rate of CHO–BMP-2. In a clinical study by Cho et al.,\(^1\) lumbar posterolateral fusion (PLF) using 6 mg of E.BMP-2 per level bilaterally showed a 100% fusion rate 24 weeks after surgery. Choi et al.\(^1\) reported a 100% PLF rate using 2.5 mg of E.BMP-2 per level unilaterally in addition to lumbar interbody fusion, and a solid fusion was observed at a mean of 4.5 months after surgery. Son et al.\(^1\) compared grafting autologous IBG and E.BMP-2 when performing lumbar interbody fusion and an additional one-sided PLF. They concluded that 1.0 mg of E.BMP-2 was effective and safe in short-level lumbar PLF, reporting a 100% fusion rate. Wang et al.\(^2\) reported a union rate of 82.9% at 6 months postoperatively and 100% at 12 months postoperatively when grafting E.BMP-2 with β-tricalcium phosphate granules in ACDF. In previous studies using BMP-2 in spine fusion surgery, a solid union was observed within a relatively short follow-up period after surgery.\(^10-12,22\) Likewise, 15 of 21 segments (71.4%) showed solid fusion at 3 months after surgery and 19 of 21 segments (90.4%) at 6 months after surgery in the present study. This is considered to be a great advantage of BMP-2. And the final fusion rate of 95.2% using E.BMP-2 was not lower than that of 84.8% to 100% using autologous IBG in previous studies.\(^23\)

Although the exact cause of postoperative prevertebral soft-tissue swelling when using BMP-2 in ACDF is not known, it is related to a robust inflammatory reaction and an excessive dose of BMP-2.\(^16\) In a meta-analysis, complication rates including reoperation, readmissions, reintubations, or tracheostomies were higher in cohorts with higher BMP doses per level and were significantly positively correlated with the dose of BMP used per level. Moreover, 0.2–0.6 mg of BMP-2 per level was enough to gain good fusion in the multilevel fusion of the cervical spine.\(^24\)

In the present study, only 1 case of dysphagia and no case of airway compromise were observed. We attributed these favorable findings to the fact that we performed the following perioperative procedures. First of all, a low

| Table 3. Radiological Outcomes |
|-----------------------------|
| Variable                  | Immediate postoperative | Final follow-up | p-value |
| Anterior disc height (mm)  | 7.9 ± 1.5                | 7.2 ± 1.4       | 0.001* |
| Posterior disc height (mm) | 6.6 ± 1.5                | 6.2 ± 1.3       | 0.005* |
| Cervical alignment (°)     | –11.3 ± 10.8             | –12.0 ± 12.7    | 0.859  |
| Segmental angle (°)        | –4.8 ± 6.0               | –4.5 ± 5.8      | 0.168  |

Values are presented as mean ± standard deviation. *Significant difference. It was measured using Cobb’s angle between C2 and C7; positive value indicates kyphosis; negative value indicates lordosis. \(^\dagger\) It was measured using Cobb’s angle between superior endplate of upper and inferior endplate of lower vertebra at the operated level(s); positive value indicates kyphosis; negative value indicates lordosis.

Fig. 4. A 54-year-old male patient diagnosed with cervical spondylotic myeloradiculopathy underwent anterior cervical discectomy and fusion with bilateral uncineectomy and received 0.3 mg of Escherichia coli-derived bone morphogenetic protein-2 per segment. Lateral radiographs of the cervical spine taken at postoperative day 1 (A), postoperative day 2 (B), postoperative day 3 (C), and 3-month follow-up (D). There were no symptoms such as dyspnea or dysphagia, and prevertebral soft-tissue swelling improved without dexamethasone administration.
A dose of 0.3 mg of E.BMP-2 per level was used. Secondly, E.BMP-2 was used with HA as a carrier. BMP-2 should be used with a carrier, such as collagen sponge, for the best outcomes. Because of the poor mechanical strength of the collagen sponge matrix, BMP-2 is easily released under pressure, which can lead to an ectopic bone formation and robust inflammation, followed by neck swelling. In contrast, the BMP-2 solution is not bound to the surface of HA but is physically entrapped in the highly porous structure of HA granules. After implantation, E.BMP-2 is expected to be slowly and continuously released from HA through diffusion, desorption, or degradation with lower risk of early leakage causing adjacent soft-tissue inflammation such as severe neck swelling. Third, oxidized cellulose was used for hemostasis, and fibrin sealants were used to prevent the spread of E.BMP-2. Finally, dexamethasone was administered intravenously, if needed.

Of the 11 patients, 1 patient (9.1%) had dysphagia, which was operated at the C3/4 level (patient 11). Although the sample size was small, it was within the incidence range of previous studies that reported the incidence of dysphagia from 1% to 79% in ACDF with or without BMP-2. It is difficult to conclude that there is a definite correlation between E.BMP-2 use and dysphagia in this patient because the upper surgical level at C3/4 is one of the risk factors for dysphagia after ACDF. However, as several cases showed significant airway swelling, the following prospective large study should address the soft-tissue swelling and dysphagia in a quantitative way, using more systematic and objective tools. It was supposed that cerebrospinal fluid leakage (patient 11) and pneumomediastinum (patient 4) were not related to E.BMP-2 use.

To the best of our knowledge, this is the first study to report the clinical results of E.BMP-2 using HA as a carrier in ACDF. However, this study has several limitations, including the small sample size without a control group, the short-term follow-up period, and the inability to establish an appropriate dose of E.BMP-2 for a solid and safe union. In addition, the use of oxidized cellulose, fibrin sealants, and dexamethasone might have helped to lower the risk of E.BMP-2 causing severe neck swelling. However, omitting these perioperative procedures poses an ethical challenge because the safety of E.BMP-2 in ACDF has not yet been established. It will be necessary to establish the efficacy and safety of E.BMP-2 with HA in ACDF compared to IBG through longer follow-up studies with higher levels of evidence in larger patient groups.

**CONFLICT OF INTEREST**

No potential conflict of interest relevant to this article was reported.

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**REFERENCES**

1. Choi SH, Kang CN. Degenerative cervical myelopathy: pathophysiology and current treatment strategies. Asian Spine J. 2020;14(5):710-20.
2. Kang KC, Lee HS, Lee JH. Cervical radiculopathy focus on characteristics and differential diagnosis. Asian Spine J. 2020;14(6):921-30.
3. Buttermann GR. Prospective nonrandomized comparison of an allograft with bone morphogenic protein versus an iliac-crest autograft in anterior cervical discectomy and fusion. Spine J. 2008;8(3):426-35.
4. Pisano AJ, Short TK, Formby PM, Helgeson MD. Anterior cervical discectomy and fusion techniques: bone graft, biologics, interbody spacers, and plating options. Semin Spine Surg. 2016;28(2):84-9.
5. Frenkel MB, Cahill KS, Jawahary RJ, Zacur G, Green BA, Levi AD. Fusion rates in multilevel, instrumented anterior cervical fusion for degenerative disease with and without the use of bone morphogenetic protein. J Neurosurg Spine. 2013;18(3):269-73.
6. Carragee EJ, Hurwitz EL, Weiner BK. A critical review of recombinant human bone morphogenetic protein-2 trials in spinal surgery: emerging safety concerns and lessons learned. Spine J. 2011;11(6):471-91.
7. U.S. Food and Drug Administration. FDA public health notification: life-threatening complications associated with recombinant human bone morphogenetic protein in cervical spine fusion. Silver Spring: U.S. Food and Drug Administration; 2008.
8. Lord EL, Cohen JR, Buser Z, et al. Trends, costs, and complications of anterior cervical discectomy and fusion with and without bone morphogenetic protein in the United States Medicare population. Global Spine J. 2017;7(7):603-8.
9. Jin YZ, Zheng GB, Lee JH. Escherichia coli BMP-2 showed comparable osteoinductivity with Chinese hamster ovary derived BMP-2 with demineralized bone matrix as carrier. Growth Factors. 2019;37(1-2):85-94.

10. Choi SH, Koo JW, Choe D, Hur JM, Kim DH, Kang CN. Interbody fusion in degenerative lumbar spinal stenosis with additional posterolateral fusion using Escherichia coli-derived bone morphogenetic protein-2: a pilot study. Medicine (Baltimore). 2020;99(24):e20477.

11. Cho JH, Lee JH, Yeom JS, et al. Efficacy of Escherichia coli-derived recombinant human bone morphogenetic protein-2 in posterolateral lumbar fusion: an open, active-controlled, randomized, multicenter trial. Spine J. 2017;17(12):1866-74.

12. Son HJ, Choi SH, Lee MK, Kang CN. Efficacy and safety of Escherichia coli-derived recombinant human bone morphogenetic protein-2 in additional lumbar posterolateral fusion: minimum 1-year follow-up. Spine J. 2021;21(8):1340-6.

13. Lee JH, Hwang CJ, Song BW, Koo KH, Chang BS, Lee CK. A prospective consecutive study of instrumented posterolateral lumbar fusion using synthetic hydroxyapatite (Bongros-HA) as a bone graft extender. J Biomed Mater Res A. 2009;90(3):804-10.

14. Smith GW, Robinson RA. The treatment of certain cervical-spine disorders by anterior removal of the intervertebral disc and interbody fusion. J Bone Joint Surg Am. 1958;40(3):607-24.

15. Sabel M, Stummer W. The use of local agents: Surgicel and Surgifoam. Eur Spine J. 2004;13(Suppl 1):S97-101.

16. Bridwell KH, Anderson PA, Boden SD, Vaccaro AR, Wang JC. What’s new in spine surgery. J Bone Joint Surg Am. 2005;87(8):1892-901.

17. Jenkins TJ, Nair R, Bhatt S, et al. The effect of local versus intravenous corticosteroids on the likelihood of dysphagia and dysphonia following anterior cervical discectomy and fusion: a single-blinded, prospective, randomized controlled trial. J Bone Joint Surg Am. 2018;100(17):1461-72.

18. Liu FY, Yang DL, Huang WZ, et al. Risk factors for dysphagia after anterior cervical spine surgery: a meta-analysis. Medicine (Baltimore). 2017;96(10):e6267.

19. Oshina M, Oshima Y, Tanaka S, Riew KD. Radiological fusion criteria of postoperative anterior cervical disectomy and fusion: a systematic review. Global Spine J. 2018;8(7):739-50.

20. Zajonz D, Franke AC, von der Hoh N, et al. Is the radiographic subsidence of stand-alone cages associated with adverse clinical outcomes after cervical spine fusion?: an observational cohort study with 2-year follow-up outcome scoring. Patient Saf Surg. 2014;8(1):43.

21. Shields LB, Raque GH, Glassman SD, et al. Adverse effects associated with high-dose recombinant human bone morphogenetic protein-2 use in anterior cervical spine fusion. Spine (Phila Pa 1976). 2006;31(5):542-7.

22. Wang Z, Lee S, Li Z, et al. Anterior cervical disectomy and fusion with recombinant human bone morphogenetic protein-2-adsorbed β-tricalcium phosphate granules: a preliminary report. J Orthop Surg Res. 2020;15(1):262.

23. Shriver MF, Lewis DJ, Kshettry VR, Rosenbaum BP, Benzel EC, Mroz TE. Pseudoarthrosis rates in anterior cervical discectomy and fusion: a meta-analysis. Spine J. 2015;15(9):2016-27.

24. Hofstetter CP, Hofer AS, Levi AD. Exploratory meta-analysis on dose-related efficacy and morbidity of bone morphogenetic protein in spinal arthrodesis surgery. J Neurosurg Spine. 2016;24(3):457-75.

25. Boerckel JD, Kolambkar YM, Dupont KM, et al. Effects of protein dose and delivery system on BMP-mediated bone regeneration. Biomaterials. 2011;32(22):5241-51.

26. King WJ, Krebsbach PH. Growth factor delivery: how surface interactions modulate release in vitro and in vivo. Adv Drug Deliv Rev. 2012;64(12):1239-56.