치의학 석사학위논문

Mechanical Evaluation of bone formation by Escherichia coli-derived rhBMP-2/ACS in beagle dogs

비글에서 Escherichia coli-derived rhBMP-2/ACS에 의한 골 형성의 Mechanical Evaluation

2017년 2월

서울대학교 대학원
치의과학과 치주과학 전공
강 민 정

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지도교수 구 기 태

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치의과학과 치주과학 전공

강 민 정

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위 원 장 _____ (인)

부 위 원 장 _____ (인)

위 원 _____ (인)

-Abstract-

Mechanical Evaluation of bone formation by Escherichia coli-derived rhBMP-2/ACS in beagle dogs

Minjeong Kang

program in periodontology, Department of Dental Science Graduate School, Seoul National University
(Directed by professor Ki-Tae Koo, D.D.S., Ph.D.)

Objective

In the present study, we performed a mechanical evaluation to determine whether new bone similar to the host bone can be formed following guided bone regeneration (GBR) with O-BMP(Escherichia coli-derived rhBMP-2/ACS).

Materials and methods

Saddle type defects, each having a diameter of 10 mm x 6 mm, were formed on 4 locations on the mandibles of 8 male beagle dogs. For each dog, a bone graft was placed on each site at random using O-BMP, Infuse, A-OSS, or ACS only. After 3 months(12 weeks), bone formation near the areas where the procedure was performed

was inspected visually and radiographically, after which the implants were placed (TSII SA Φ 5.0x6mm (Osstem)). During the implantation, a trephine drill (Φ3.7/Φ4.5) was used to obtain bone specimens for compression testing to be performed later, while insertion torque and ISQ were measured. The upper prosthesis was inserted at 3 months after the implantation (cylindrical resin block as a single unit), at which time ISQ was measured again. The bone specimens obtained during the implantation were compressed by 1 mm/min to measure the compression yield strength (0.2% offset).

Result

In all experimental groups, definitive new bone formation noticeable by visual inspection or radiography occurred at 12 weeks post-GBR. The experimental results were analyzed using a nonparametric test (Kruskal-Wallis test). During implantation, at 3-months post-GBR, the torque values for O-BMP, Infuse, A-OSS, and ACS were all within the recommended implantation value range (10 - 40 N). Most of the experimental groups, including O-BMP, showed mean ISQ values ≥ 70 during the implantation, and outstanding initial fixation power of the fixture was observed in all groups. At 3 months post-implantation, most of the experimental groups showed mean ISQ values ≥ 70 during the insertion of the upper prosthesis. Moreover, the highest compression yield strength value in the bone specimen was found in the group administered O-BMP.

Conclusion

O-BMP exhibited the capability to form new bone similar to the host bone. It was equivalent to other currently used methods.

Key word : Bone morphogenic protein, E-coli derived rhBMP-2, osteoinduction, new bone formation.

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Introduction

The loss of a permanent tooth results in atrophy and loss of nearby periodontal tissues used to support that tooth, especially the alveolar bone. Such atrophy and loss of the alveolar bone can cause an anatomical problem that increases the difficulty of implantation for adequate recovery of the functions compromised by the loss of the tooth. Adequate implantation on the atrophied and lost alveolar bone requires a certain level of recovery in the amount and shape of the bone. Bone quality equivalent to that of the host bone level is especially important; therefore, various bone graft procedures were performed.

For the bone graft procedures, autogenous, allogenic, xenogenic, and synthetic bones were used alone or in combination, and each bone graft material achieved new bone formation through osteogenesis, osteoinduction, and osteoconduction. Various growth factors are known to play a role during such a bone formation process.

Among them, bone morphogenetic proteins (BMPs) were first described in 1965 by Dr. Marshall Urist, an orthopedic surgeon, who discovered a protein extracted from a demineralized bone matrix that had an osteoinductive capability.(1,2,3,4) Since then, many studies in various fields have been conducted for the treatment of traumatic, degenerative, and necrotic bone lesions with the use of BMPs. Even in the field of dentistry, the efficacies of few materials, including osteoinductive BMPs, had been proven in animal experiments or clinical trials.

There are approximately 20 members that belong to the family of human BMPs, and among them, BMP-2, BMP-4, and BMP-7 are considered to play an important role in bone formation.(5,8,9) BMP-2

was the first growth factor found to induce cartilage and bone formation, followed by the discovery of the important roles played by other members in osteoinduction.(11) BMPs are involved in endochondral bone formation and direct(intramembranous) bone formation through complex cellular events, such as recruitment and proliferation of monocytes and mesenchymal cells, cartilage formation, vascularization, bone formation, and bone remodeling and maturation.(6,7,18) In addition, BMPs are also known to have inductive effects on cartilage formation(19) and have been reported to improve the recovery of soft tissues.(20) In other words, BMPs play several important roles in bone induction, such as chemotaxis, mitosis, and differentiation into cartilage, and they are considered to be pleiotropic morphogens. In 2001, the Food and Drug Administration(FDA) approved the use of rhBMP-7(aka OP-1; Stryker Biotech) in orthopedic surgery as a substitute for autogenous bone for the non-union of long bone.(12) In 2004, rhBMP-7 was also used in posterolateral fusion.(12) Moreover, in 2002, rhBMP-2(Infuse; Medtronic 社) was approved for use as a bone graft material in anterior lumbar interbody fusions(ALIFs) with a lumbar fusion device, an orthopedic procedure, and thereafter, the range of use for rhBMPs has gradually expanded to maxillofacial procedures. Currently, various commercial rhBMPs are used in dental implants for alveolar bone regeneration.

Accordingly because the amount of native BMP that can be obtained from bone is very little, rhBMPs were developed and are being mass-produced for clinical use. Compared to rhBMP-2 obtained from mammalian(Chinese hamster ovary [CHO]) cells, rhBMP-2 obtained from a bacterial expression system(Escherichia coli) has a higher level of expression and is more useful for mass production; it is

therefore developed and used more often.

Furthermore, carrier materials also play an important role. They enable growth factors to be better maintained for application after rhBMP-2 is delivered to a defect site. Among these carrier materials, absorbable collagen sponges (ACSs) have been evaluated and used often in in vivo and clinical settings due to their ease of operation and rapid biodegradation.

The objective of the present study was to investigate whether O-BMP(rhBMP-2/ACS) can facilitate the formation of a new bone similar to the host bone in dogs that have experienced loss of alveolar bone and whether the proper amount and shape of bone can be recovered. In addition, the present study also aimed to determine whether O-BMP could be an effective bone graft material by comparing its post-implantation conditions to those from using autogenous bone and various other commercially available materials.

Materials and Methods

Experimental animals

Eight male beagle dogs with a mean age of 30 weeks and weight of 9.5 - 10 kg were used. Defects were formed on 4 locations on the mandible of each beagle, followed by implantation. The collected data included procedure date, amount of anesthesia administered during the procedure, visual observation and radiography results, follow-up dates, and specific findings for each individual.

Surgical procedure

1. Extraction

Tooth extraction was performed in the surgery room under sterile conditions using general anesthesia that was administered intravenously with a 1:1 ratio of ZLT (zoletile) and ZLZ (rumpun, xylezine) and maintained during the procedure through a ventilator with 1:1.5 ratio of ISO (isoflurane) and O_2. The 1_{st}, 2_{nd}, and 3_{rd} mandibular premolars and the 1_{st} and 2_{nd} mandibular molars were extracted.

2. Defect formation and GBR

- saddle type defect (10 mm x 6 mm) (Fig 1)

A saddle type defect (10 mm x 6 mm) was formed on 4 separate locations on both sides of the mandible (2 on the left and 2 on the right) after 2 months of healing from tooth extraction. In addition, a bone screw was fixed on two locations between the defect sites on

both sides to surmise the position of the defect sites after new bone formation. For this procedure, local anesthesia with lidocaine HCl containing 1:100,000 epinephrine was administered to the gum area where the procedure was being performed.

Thereafter, GBR was performed for new bone formation in the defect sites. The bone graft materials used were O-BMP(rhBMP-2/ACS), Infuse, A-OSS, and ACS only. Here, to minimize any deviation stemming from differences in the location, 4 types of materials were applied in a random order at the implantation sites. An absorbable collagen membrane(bio-guide) was used.

At 3 months post-GBR, observations were made visually and photographically to look for dehiscence, inflammation, and erythema in the surgical sites, while the degree of bone regeneration was observed via radiographic analysis. X-rays were taken in close contact from both sides of each site, and the exposure time was maintained at 0.05 sec.

3. Implantation

The implant used for implantation was TSII SA Φ5.0x6mm (Osstem), which is a straight implant with an SA(sand blasted with alumina and acid-etched) surface. A trephine drill (Φ3.7/Φ4.5) was used during the implantation and the bone obtained at this time was used for compression strength measurements. Here, bone specimens were obtained randomly, 2 each from the host bone and each experimental group (a total of 10 bone specimens), and were compressed by 1 mm/min to measure the bone strength as compression yield strength(0.2% offset). Moreover, the insertion torque during implantation was measured with a KAVO monitoring system and ISQ was measured by verifying the initial fixation power

after the implantation.

4. prosthesis insertion

First, at 3 months of healing after implantation, the ISQ value was measured and compared to the ISQ value from the implantation stage to determine implant stability.

Thereafter, the prosthesis was inserted. The prosthesis was a single unit in the form of a cylindrical resin block and was attached only to the frontal area of the implantation site. Impressions of the original teeth were not taken prior to the fabrication of the prostheses; therefore, difficulties were experienced with normal occlusal fit between the maxillary and mandibular teeth. Using the anatomy of the maxillary teeth, the anatomy of the mandibular teeth was surmised and prostheses were immediately fabricated on site.

5. Sacrifice

The dogs were sacrificed after maintaining the prostheses for 3 months.

Results

Among the 8 dogs, 1 dog died following GBR. In the groups that were evaluated, satisfactory new bone formation was observed by visual inspection or radiography during the 3 months of healing following GBR. An incision on the gums for visual inspection confirmed new bone formation, and adequate bone width and height were maintained. In addition, at 3 months post-GBR, distinct new bone formation was identified in the defect site by radiographic observation. (Fig 2) Following GBR and subsequent new bone formation, implantation was performed successfully and healing progressed well. In all experimental groups, synostosis was successful and peri-implantitis did not occur.

Trephine drilling was performed to obtain bone specimens for compression strength measurements. The insertion torque values measured during TSll implantation are shown in Table 1. Assuming a normal recommended insertion torque of 10-50 N, all experimental groups, including O-BMP, showed insertion torque values within the normal range.

In addition, ISQ values measured from 7 dogs during the implantation stage are shown in Figure 3-(a).
As shown in the graph, in most of the experimental groups, including O-BMP, the mean ISQ value of $\geq$ 70 was observed, demonstrating excellent initial fixture fixation power in all groups.

At 3 months post-implantation, the upper prosthesis was inserted as a resin block in 4 dogs, and the ISQ value was measured at this

stage.(Fig 3-(b)) Again, all experimental groups except for 1 dog showed a mean value of ≥ 70, demonstrating that the excellent fixation power of the implant fixture was maintained in comparison with the post-implantation levels. ISQ values measured after implantation at 3 months post-GBR and before prosthesis installation at 6 months post-GBR were analyzed using a nonparametric test (Kruskal-Wallis test), the results of which showed no significant differences in ISQ values among the experimental groups ($p>0.05$)(Table 2). Therefore, it was determined that the capability of O-BMP to form new bone is similar to that of other existing bone graft materials. This resin block was maintained and verified until 3 months after prosthesis installation, and until that time, bone absorption near the fixture was not observed. However, all experimental groups did show a slight decrease in bone level after prosthesis insertion due to insufficient oral hygiene.

A total of 10 bone specimens obtained from trephine drilling and 2 specimens selected arbitrarily from each experimental group after sacrifice were measured for compression yield strength with 0.2% offset.(Fig 4) However, 2 A-OSS specimens broke into 2 pieces during bone biopsy, and the compression yield strength was not measured due to insufficient length. Consequently, compression yield strength results measured from the remaining specimens, including the host bone, are shown in Table 3. The mean compression yield strength of O-BMP was 74.1, which was found to be excellent in comparison to that of other materials. These values were also analyzed using a nonparametric test(Kruskal-Wallis test), and no significant differences among the experimental groups were found. ($P=0.212(P>0.05)$)

Discussion

Many studies have already provided clinical proof of BMPs' ability to induce effective bone formation through osteoinduction. Moreover, BMPs are being used clinically in orthopedic surgery, maxillofacial surgery, and dentistry in new bone-forming graft materials that can be substituted for autogenous bone. The present study aimed to investigate whether rhBMP-2(1.05mg) obtained from E. coli with ACS as the carrier(size: 5 cm*1.25 cm* 0.5 cm) can facilitate the formation of a new bone similar to the host bone as ably as previously developed rhBMP materials. Increasing the concentration of rhBMP is thought to lead to more extensive and aggressive bone remodeling. However, it has been reported that the concentration of rhBMP-2 generally does not have an influence on the amount of bone formation. Wikesjö et al. observed new bone formation with varying concentrations of rhBMP-2 and found that the amount of bone formation was unrelated to rhBMP-2 concentration.(13) Moreover, Boyne et al. also reported that when collagen soaked with rhBMP-2 was applied during maxillary sinus floor augmentation, the amount of bone formation and concentration were found to be unrelated to each other.(23)

rhBMP-2 in an absorbable collagen sponge(ACS) carrier has been used in increasing alveolar bone mass prior to the intraosseous placement of dental implants.(14,15) Moreover, rhBMP-2 has been reported to significantly increase the amount of new bone area and bone-to-implant contact, while also increasing bone growth in peri-implant osseous defects and the area around the surface of intraosseous dental implants during the placement of a dental

implant.(16) Furthermore, in a study that compared the group that received rhBMP-2-absorbed ACS and the group grafted with autogenous bone for maxillary sinus floor elevation, the results showed that there was no difference in the rate at which bone formation occurred, while the rhBMP group showed new bone formation with higher bone density.(21.22)

Infuse(rhBMP-2/ACS at a 1.5-mg/cc concentration) received approval from the FDA in 2002 as a graft material for certain interbody spinal fusion procedures, and as its applicable areas expanded, it was approved for use as a graft material in sinus augmentation and alveolar ridge augmentation for filling defects in tooth extraction sites in March 2007.(17)

A-oss is a product that is similar to Bio-Oss, which is a product from the world-renowned pharmaceutical company Geistlich Pharma. It is a low-temperature type xenogenic bone graft material, which has a structure and properties similar to those of human bones. This means that it has a similar pore structure as human cancellous bone, and its main component is hydroxyapatite(HA), which contains carbonates. In other words, it is an osteoconductive bovine bone substitute that plays the supporting role of maintaining the volume of the graft site until it is replaced by new bone.

O-BMP is a product similar to Infuse, which is an Escherichia coli-derived rhBMP-2(rhBMP-2/ACS) in a freeze-dried white powder form 1.05mg (size: 5 cm*1.25cm*0.5 cm).

The ACS (absorbable collagen sponge) is an absorbent, moldable, cohesive carrier for rhBMP-2. The ACS carrier matrix locally maintains the concentration of rhBMP-2 to induce bone formation in the applied area and acts as a temporary scaffold for new bone formation.

First, satisfactory new bone formation was observed, visually and radiographically, in all experimental groups during the 3 months of healing following GBR. Upon visual inspection, no dehiscence, erythema, or inflammatory response was observed in the gums. On radiography, the defect boundaries disappeared at 6 weeks post-GBR and new bone formation was verified at 3 months post-GBR. In conclusion, distinct new bone formation near the defect area was observed in all experimental groups. (Fig 2)

In these experiments, the maximum implant insertion torque values in all groups were confirmed to range within the recommended insertion value range.(Table 1) A previous study that implanted various types of implants on healthy jawbones of beagles also found that insertion torque values ranged between 20 and 50 N. (24) Accordingly, it was confirmed that the adequate formation of new bone similar to healthy host bone had occurred in the defects in all experimental groups.

In the experimental groups, the ISQ value at implantation was ≥ 70, which was appropriate to proceed to the prosthesis stage. As a result, the mean ISQ value from the experimental groups who progressed to the prosthesis stage was ≥ 75. This demonstrated that the experimental groups showed excellent values for initial fixture fixation power as well as the fixture fixation power before prosthesis insertion. Moreover, the ISQ value was mostly higher in these experimental groups from measurements taken before prosthesis insertion than during implantation. In other words, it was determined that following implantation, new bone formation and osseointegration between the implant and nearby bone were successful. Moreover,

because there were no significant differences in the ISQ values among the experimental groups (P>0.05), O-BMP can be considered to possess similar new bone-forming capabilities as other existing graft materials. (Table 2)

Afrer implantation at 3 months post-GBR, ACS only and Infuse (rhBMP-2/ACS) initially showed higher values, while O-BMP and A-OSS showed relatively lower values. However, O-BMP and A-OSS showed a noticeable increase in the ISQ value at 6 months post-GBR (3 months following implantation). ACS and Infuse (rhBMP-2/ACS) had high ISQ values during implantation and showed a low increasing trend, while O-BMP and A-OSS showed a high increasing trend, which ultimately resulted in no major differences in the ISQ value among the experimental groups during prosthesis placement.(Graph 1) In the present study, it was found that ACS and Infuse(rhBMP-2/ACS) showed most of the new bone formation during the first 3 months after implantation, while O-BMP showed a slightly different pattern of gradual new bone formation over 6 months. Such gradual bone formation was especially prominent in A-OSS, which initially showed the lowest ISQ value after implantation. It is believed that the osteoconductive properties of A-OSS had an influence on the observation of these values. However, additional studies are necessary to support the experimental results that showed the highest ISQ in the experimental group that only received ACS.

In the experiment that measured compression yield strength (excluding A-OSS, in which the specimen broke into 2 pieces during the bone biopsy procedure), O-BMP showed a higher strength value

(74.1) than other materials.(Table 3) Based on this, it was determined that newly formed bone after the placement of an O-BMP graft had sufficiently high strength relative to other materials or the host bone. This also did not show significant differences versus the other test group, including the host bone. ($p>0.05$) Therefore, it was confirmed that similar to other graft materials, O-BMP was able to form new bone with hardness values similar to those of the host bone. In a study by Dennis R et al., as well as in other studies, compression yield strength and bone density exhibited a proportional relationship to each other with the same elongation value.(25) Such bone density represents the mineral content in the bone, which is related to bone quality. Later, additional studies about bone quality will be necessary.

In the present study, in view of the maximum torque value, ISQ value, and compression yield strength during implantation, it was shown that the capability of O-BMP was similar to that of currently used graft materials in forming new bone with a hardness comparable to that of host bone.

Reference

1. Urist MR, Bone formation by autoinduction. Science 1965;150:893-899.
2. Urist MR, Strates BS, Bone morphogenetic protein. J Dent Res 1971;50(6):1392-1406.
3. Urist MR, Iwata H, Ceccotti PL, et al. Bone morphogenesis in implants of insoluble bone gelatin. Proc Natl Acad Sci USA 1973;70:3511-15.
4. Urist MR, Mikulski A, Lietze A. Solubilized and insolubilized bone morphogenetic protein. Proc Natl Acad Sci U S A 1979;76:1828-32.
5. Carreira AC, Lojudice FH, Halcsik E, Navarro RD, Sogayar MC, Granjeiro JM: Bone morphogenetic proteins: facts, challenges, and future perspectives. J Dent Res 2014;93:335-345.
6. Cook SD, Rueger DC. Osteogenic protein-1: biology and applications. Clin Orthop Relat Res 1996;324:29-38.
7. Reddi AH. Initiation of fracture repair by bone morphogenetic proteins. Clin Orthop Relat Res 1998;355(Suppl S):66 - 72.
8. Sakou T. Bone morphogenetic proteins: from basic studies to clinical approaches. Bone 1998;22:591 - 603.
9. Schmitt JM, Hwang K, Winn SR et al. Bone morphogenetic proteins: an update on basic biology and clinical relevance. J Orthop Res 1999;17:269 - 278.
10. wozney JM, Rosen V, Celeste AJ, et al. Novel regulators of bone formation: Molecular clones and activity. Science 1988;242:1528-1534.
11. Wang EA, Rosen V, DJ, et al. Novel regulators of bone formation: Molecular clones and activity. Science 1988l relevance. JAcad Sci USA 1990;87:2220-4.
12. Ong KL, Villarraga ML, Lau E, Carreon LY, Kurtz SM, Glassman

SD. "Off-label use of bone morphogenetic proteins in the United States using administrative data". Spine 2010;35(19): 1794 - 800.
13. Ulf M. E. Wikesjö, Paola Guglielmoni, Ananya Promsudthi, Kyoo-Sung Cho, Leonardo Trombelli, Knut A. Selvig, Lisa Jin, John M. Wozney. Periodontal repair in dogs: effect of rhBMP-2 concentration on regeneration of alveolar bone and periodontal attachment. Journal of Clinical periodontology 1999;26(6):392-400.
14. Hanisch, O., Tatakis, D. N., Boskovic, M. M., Rohrer, M. D. ;Wikesjö. Bone formation and reosseointegration in peri-implantitis defects following surgical implantation of rhBMP-2. The International Journal of Oral and Maxillofacial Implants 1997;12:604 - 610.
15. Jovanovic, S. A., Hunt, D. R., Bernard, G. W., Spiekermann, H., Nishimura, R., Wozney, J. M. & Wikesjö, U. M. E. Long-term functional loading of dental implants in rhBMP-2 induced bone. A histologic study in the canine ridge augmentation model. Clinical Oral Implants Research 2003;14:793 - 803.
16. David L. Cochran, Robert Schenk, Daniel Buser, John M. Wozney, and Archie A. Jones. Recombinant human bone morphogenic protein-2 stimulation of bone formation around endosseous dental implants. journal of periodontal 1999;70:139-150
17. William F. McKay, Steven M. Peckham, and Jeffrey M. Badura. A comprehensive clinical review of recombinant human bone morphogenetic protein-2 (INFUSE® Bone Graft). Int Orthop. 2007;31(6):729 - 734.
18. Canalis E, Economides AN, Gazerro E. Bone morphogenetic proteins, their antagonists and the skeleton. Endocr Rev 2003;24:218 - 235.
19. Reddi AH. Role of morphogenetic proteins in skeletal tissue

engineering and regeneration. Nat Biotechnol 1998;16:247 - 252.
20. Herford AS, Boyne PJ, Williams RP. Clinical applications of rhBMP-2 in maxillofacial surgery. J Calif Dent Assoc 2007;35:335-341.
21. Boyne PJ, Marx RE, Nevins M, Triplett G, Lazaro E, Lilly LC, Alder M, Nummikoski P. A feasibility study evaluating rhBMP-2/absorbable collagen sponge for maxillary sinus floor augmentation. Int J Periodont Rest Dent 1997;17:11-25.
22. Triplett GR, Nevins M, Marx RE, Spagnoli DB, Oates TW, Moy PK, Boyne PJ. Pivotal, Randomized, parallel evaluation of recombinant human bone morphogenetic protein-2/absorbable collagen sponge and autogenous bone graft for maxillary sinus floor augmentation. J Oral maxillofacial surg 2009;66:1947-1970.
23. Boyne PJ, Lilly LC, Marx RE, Moy PK, Nevins M, Spagnoli DB, Triplett RG. De novo bone induction by recombinant human bone morphogenetic protein-2(rhBMP-2) in maxillary sinus floor augmentation. J Oral Maxillofacial surg 2005;63:1693-1707.
24. Manresa C1, Bosch M, Echeverría JJ. The comparison between implant stability quotient and bone-implant contact revisited: an experiment in Beagle dog. Clin Oral Implants Res. 2014;25(11):1213-21
25. Dennis R. Carter and Wilson C. Hayes. Bone Compressive Strength: The Influence of Density and Strain Rate. Science 1976:194(4270):1174-1176.

TABLES

Table 1) Torque values measured during implantation.
(the mean ± SD values.)

Test Group	Maximum torque (Ncm)
O-BMP	36.80 ± 8.70
Infuse	35.21 ± 16.19
A-OSS	32.37 ± 0.00
ACS	42.18 ± 11.41

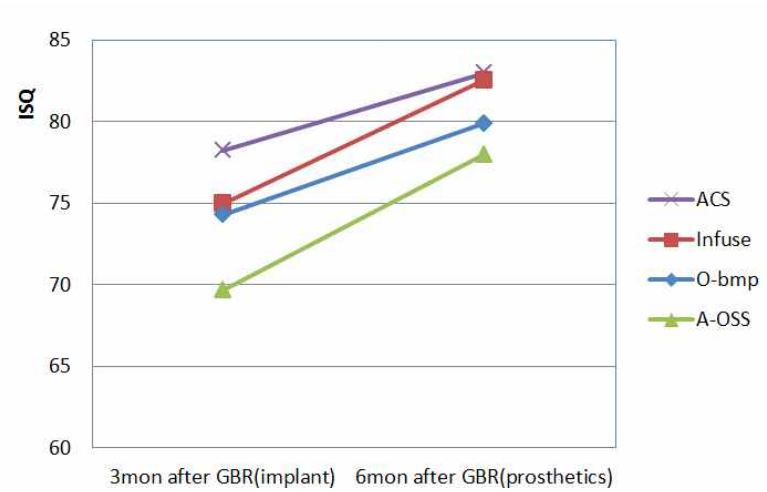
Table 2) Statistical summary of ISQ (3 and 6 months after GBR) between experimental groups(Kruskal-Wallis test)

parameter	Test group	mean	standard deviation	p value
ISQ (3mon after GBR)	O-BMP	74.29	7.27	.472
	Infuse	74.99	8.20	
	A-OSS	69.68	8.64	
	ACS	78.25	3.20	
ISQ (6mon after GBR)	O-BMP	79.90	2.41	.241
	Infuse	82.56	4.08	
	A-OSS	78.00	0.71	
	ACS(2)	83.00	2.83	

Table 3) the mean value of Compressive yield strenght(MPa)
(the mean ± SD values.)

Test group	specimen	Compressive yield strength(MPa)	Mean ± S.D (MPa)
Host bone	specimen #1	22.6	35.6 ± 18.4
	specimen #2	48.6	
O-BMP	specimen #1	75.3	74.1 ± 1.78
	specimen #2	72.8	
infuse	specimen #1	56.1	51.6 ± 6.39
	specimen #2	47.1	
ACS	specimen #1	31.1	44.5 ± 19.1
	specimen #2	58.0	

GRAPH



Graph 1) Pattern of change in ISQ immediately after implantation and after prosthesis placement. Despite having a low initial ISQ value, O-BMP showed gradual bone formation for 3 months, reaching an ISQ value similar to that of other materials during the prosthesis procedure at 6 months post-GBR.

FIGURES

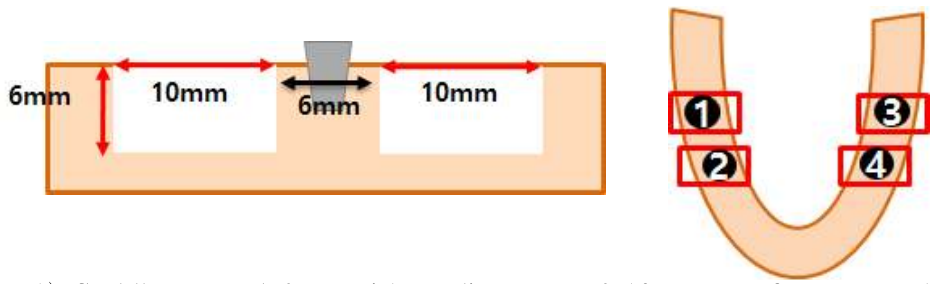


Fig 1) Saddle type defect with a diameter of 10 mm x 6 mm on the mandibles of 8 beagles. A bone screw was fixed on the host bone between the defect sites, which was intended to surmise the position of the defect sites after new bone formation. Sites 1, 2, 3 and 4 were designated on the left and right sides of the mandible.

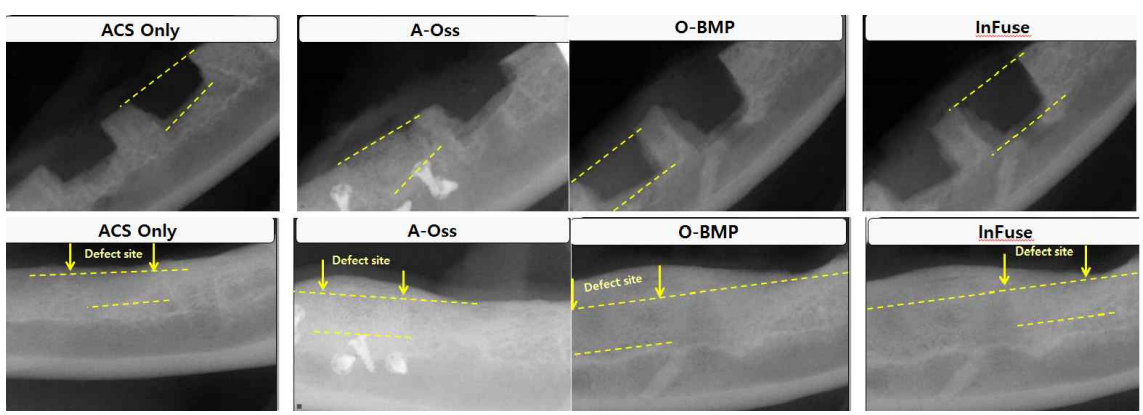
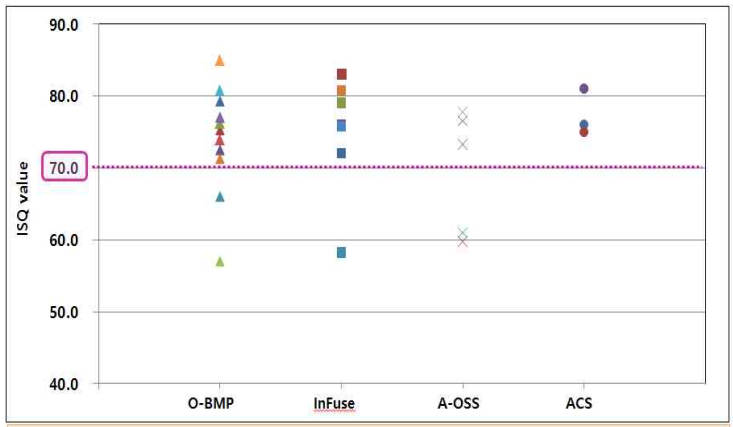


Fig 2) Radiographic images at 12 weeks post-GBR. Distinct new bone formation was identified in the defect sites.

(a)



(b)

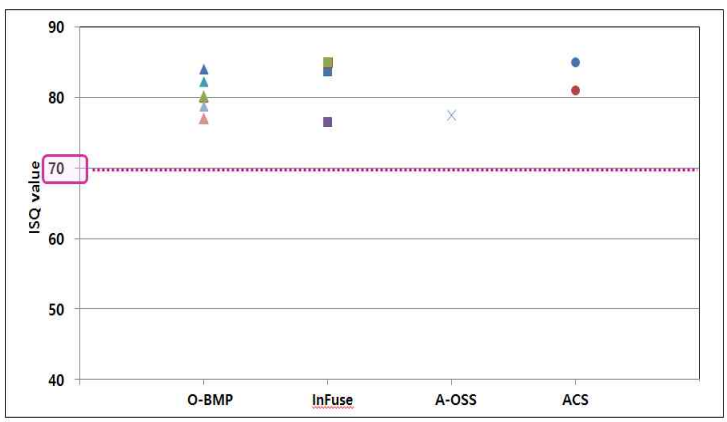


Fig 3) ISQ value range after implantation (3 months after GBR) (a) and prosthesis insertion (6 months after GBR) (b). Most experimental groups, including O-BMP, maintained a value ≥ 70.

Fig 4) Compression yield strength measurements (0.2% offset), 1 mm/min compression.

-국문초록-

비글에서 Escherichia coli-derived rhBMP-2/ACS에 의한 골형성의 mechanical evaluation

강 민 정
서울대학교 대학원 치의과학과 치주과학 전공
(지도교수 구 기 태)

1. 목 적
O-BMP(Escherichia coli-derived rhBMP-2/ACS)를 적용한 골재생술 이후에 host bone 수준의 new bone이 형성되는지 mechanical evaluation를 시행하였다.
2. 방 법
수컷의 8마리 beagle dog의 하악 4군데에 각각 직경 10mm x 6mm의 saddle type defect를 형성하였다. 각각의 개체, 각각의 site에 임으로 ① O-BMP ② Infuse ③ A-OSS ④ ACS only로 골이식을 시행하였다. 그 후 3개월(12주) 후 육안과 X-ray로 시술부위의 골생성을 확인하였고 implant를 식립하였다(TSII SA Φ5.0x6mm(osstem)).
implant 식립시 trephine dril(Φ3.7/Φ4.5)로 추후 압축강도 측정을 위한 골시편을 채취하였고, insertion torque과 ISQ를 측정하였다.
implant 식립 3개월 후 상부보철을 체결하였으며(single unit로 원통형

레진 블록) 이때에도 ISQ를 측정하였다. 임플란트 식립시 채취한 골시편은 1mm/min로 압축하여 압축항복 강도를 측정하였다(0.2% offset)

3. 결 과

모든 실험군에서 GBR 12주 이후에 육안으로나 x-ray 상으로 현저한 new bone formation이 일어났다. 실험 결과는 비모수적 검정을 사용하여 분석하였다(Kruskal-Wallis 검정)

GBR 3개월 후 임플란트 식립시 토크 값은 O-BMP, Infuse, A-OSS, ACS 모두 임플란트 권장식립 값(10-40N) 내에 분포한다.

O-BMP를 포함한 대부분의 실험군에서 임플란트 식립시 ISQ value도 평균 70이상의 값을 가지며 모든 실험군에서 초기 fixture 고정력이 우수함을 확인할 수 있었다. 임플란트 식립 3개월 후 임플란트 상부 보철 체결 단계에서의 ISQ value값도 대부분의 실험군에서 평균 70이상의 값을 나타내었다. 또한 골시편의 압축항복 강도에서는 O-BMP를 이식한 실험군에서 가장 높은 값을 나타내었다.

4. 결론

O-BMP가 현재 사용되는 다른 재료들과 동등한 수준으로 host bone 수준의 골형성능력을 보여준다.

주요어: Bone morphogenic protein, E-coli derived rhBMP-2, 골유도, 새로운 골 형성.

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