Evaluation of a hydrogel membrane on bone regeneration in furcation periodontal defects in dogs

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The aim of the study was to evaluate bone regeneration using a canine model with surgically created periodontal defects filled for 12 weeks using a stratified biomaterial consisting in a biphasic calcium phosphate (BCP) covered with a crosslinking hydrogel acting as polymer membrane of silated hydroxypropyl methylcellulose (Si-HPMC) as the tested new concept. Bilateral, critical-sized, defects were surgically created at the mandibular premolar teeth of six adult beagle dogs. The defects were randomly allocated and: (i) left empty for spontaneous healing or filled with: (ii) BCP and a collagen membrane; (iii) BCP and hydrogel Si-HPMC membrane. At 12 weeks, the experimental conditions resulted in significantly enhanced bone regeneration in the test BCP/Si-HPMC group. Within the limits of this study, we suggest that the hydrogel Si-HPMC may act as an occlusive barrier to protect bone area from soft connective tissue invasion and then effectively contribute to enhance bone regeneration.

Keywords: Periodontal diseases, Furcation defect, Calcium phosphate, Hydrogel

INTRODUCTION

Many bone graft materials (e.g., autogenous bone, allografts, deproteinized bovine bone xenografts, and bone alloplastic grafts) have been used to treat periodontal intrabony defects1-3. Alloplastic biomaterials are of synthetic origin and provide an alternative to allografts or xenografts. Various bone alloplastic graft materials are available, including bioactive glass4, hydroxyapatite (HA), beta-tricalcium phosphate (β-TCP), and biphasic calcium phosphate ceramic (BCP)5. Histological evidence indicates that these synthetic biomaterials act as biological bone fillers. Their use mainly enhances regeneration along the junctional epithelium, providing very limited new connective tissue attachment6. However, potential of periodontal regeneration can be enhanced when grafting materials are combined with barrier membranes7. BCP is a combination of HA and β-TCP in a variable weight ratio (60/40 or 80/20) and particle size (mainly 80–200 μm or 0.5–1 mm). It presents advantages in that it combines short-term (β-TCP) and long-term (HA) absorbable phases, allowing for a controlled bioactivity4. BCP has been evaluated in various preclinical and clinical studies in periodontology8-11 and implantology12-14. Thus, it could be considered as a relevant osteoconductive scaffold for bone regeneration5. Nevertheless, results have indicated that bone graft materials remain unpredictable in terms of bone filling and periodontal regeneration. In addition, the morphology of intrabony periodontal defects may be unfavorable for regenerative procedures in wide lesions or when lesions are associated with limited residual bone walls15. Furthermore, maintaining wound and blood clot stability during the initial healing phases is essential for enhancing bone regeneration and final clinical outcomes16,17.

In addition, in intrabony or class II furcation defects, ad integrum periodontal regeneration may be obtained through the use of cohesive barrier membranes. The goal of guided tissue regeneration (GTR) is to prevent the migration of epithelial or connective tissue cells into the blood clot and along the instrumented root surface18. Cells from the periodontal ligament and bone need to repopulate the root surface and regenerate cement19, thus limiting apical migration of the long junctional epithelium along the root surface. Barriers used in GTR can be either non-resorbable (expanded polytetrafluoroethylene [PTFE]) or resorbable (polymer or collagen) membranes20. In unfavorable defects (wide or with limited bone walls), the membranes can be used in association with graft materials. Combined therapies have been shown to enhance clinical results compared to the use of bone graft materials alone21,22. However, the use of regenerative membranes is quite difficult to use and can lead to a significant risk for complications, such as infection or early exposure7.

Due to the non-easy handling biomaterial with the current membranes, a lot of generalist dental surgeons
do not use GTR for periodontal treatment. Majority of patients has no adequate treatment, resulting in high morbidity of periodontal diseases. Recently, new biomaterials have been proposed as potential regenerative membranes, including chitosan23,24 or hydrogels25,26. Additionally, silated hydroxypropyl methylcellulose (Si-HPMC), which is a self-crosslinking absorbable hydrogel, has shown promising biocompatibility as well as characteristics that might facilitate periodontal regenerative therapies27,28. Due to its crosslinking and elastic properties, Si-HPMC could serve as an effective GTR barrier for enhancing bone-filling material stability during the healing phase. The gelation process delays cell and tissue colonization by slowly degrading the Si-HPMC29,30, potentially preventing the migration of epithelia and connective tissue31.

Dogs are the main animal model employed in periodontal regeneration research32. In fact, various preclinical systems have been developed in dogs, such as surgically induced furcation defects in maxillary premolars33 and supraalveolar periodontal defects in mandibular premolars34. These experimentally created, critical-sized defects do not regenerate spontaneously and can be used to evaluate the efficacy of biomaterials in bone regeneration. The surgical creation of supraalveolar periodontal defects is the most commonly used preclinical research model in dogs.

Objectives of the study: In the present study, the main objective was to evaluate new bone (NB) formation in experimentally surgically created supraalveolar periodontal defects that were either (i) left empty for spontaneous healing (control condition) or filled with the following materials: (ii) BCP and a collagen membrane (clinical reference treatment); (iii) BCP and a hydrogel Si-HPMC reticulating membrane (test). The second objective was to evaluate the potential of the Si-HPMC hydrogel to be used as a periodontal membrane. The study was designed according to the ARRIVE guidelines35, which were adapted for in vivo preclinical animal research36,37.

**MATERIALS AND METHODS**

**Animals**

Six female adult beagle dogs were purchased from a professional stockbreeder (mean age 48±2 months, mean weight 16±1 kg). We preferred to include dogs of the same sex (female) for practical reasons of stabling, who developed spontaneous/natural periodontitis and whose age (less than 4 years) excludes alterations of the bone remodeling due to aging. During the experimental period, the dogs were housed in a collective kennel and put on a soft food diet, with water available ad libitum. Animal handling and surgical procedures were conducted according to the guidelines set by the European community for the care and use of laboratory animals (2010/63/UE) and were approved by the Animal Welfare Committee at the ONIRIS College of Veterinary Medicine in Nantes.

**Surgical procedure**

One week prior to the surgical procedure, the animals underwent professional scaling under general anesthesia. The surgery was also performed under general anesthesia using an intravenous injection of diazepam (0.25 mg/kg; Valium®, Roche, Boulogne Billancourt, France) and propofol (4 mg/kg; Rapinovet®, Schering Plough, Hertfordshire, UK). A single dose of morphine (0.1 mg/kg; Morphine, Cooper, Melun, France) was injected subcutaneously during surgery as an analgesic. Local anesthesia with 0.50% bupivacaine (Bupivacaïne®, Agettatt, Lyon, France) was also administered in mandibular blocks. After general anesthesia, the gingiva was disinfected using an iodine solution, and the animal was covered with a sterile drape.

A muco-periostal flap was reflected at the level of
The third and fourth mandibular premolar teeth (P3 and P4) and critical-sized supraalveolar periodontal defects were created as described previously. In brief, the alveolar bone was surgically reduced circumferentially around P3 and P4 to a level of 6 mm from the cement–enamel junction. The preparation of the defect sites was performed with a motor-driven drill (Aesculap, Tuttlingen, Germany) and a fissure carbide bur under constant saline irrigation. A manual root-planning of the root surface was performed in order to eliminate totally the periodontal ligament. Thereafter, each side of the mandible was randomly assigned to one of the three conditions, then for each side of the mandibles, there was more than one defect. The negative control sites were left empty for spontaneous healing and all others sites were filled with one of the biomaterials (Fig. 1). The flaps were repositioned coronally and sutured with absorbable sutures (Vicryl® 3.0, Ethicon, Issy les Moulineaux, France). The transgingival wound closure left the tooth structure intact in order to permit a primary intention healing. The same operator (X.S.) performed all of the surgical procedures. Altogether, the experiment involved 18 defects. Prophylactic antibacterial treatment was administrated after surgery (Stomorgyl® [spiramycin and metronidazole], Merial, Lyon, France).

**Animal sacrifice**

After a healing period of 12 weeks, animals were euthanized by an intravenous overdose of sodium pentobarbital (Dolethal®, Vetaquinol, Lure, France). Both mandibles were immediately dissected, placed in a formaline solution, and stored at 4°C.

**Histological preparation**

The non-decalcified bone samples were dehydrated in an ascending series of ethanol solutions (70–100%) and then in pure acetone for 24 h. The samples were impregnated in methyl methacrylate (Prolabo, Briare, France) for four days and then embedded in a polymethyl methacrylate resin before sectioning. Blocks were cut into 100 μm slices with a circular diamond saw (Microtome 1600, Leica, Frankfurt am Main, Germany) for hematoxylin-eosin or into 7 μm slices with a hard tissue microtome (Leica SM 2500, Leica) for Goldner’s staining. The axis of the sectioned slices was primarily vertical and bucco-lingual, located in the center of the furcation defect. A second cutting axis, which was perpendicular to the first one, was also made in order to investigate bone and periodontal regeneration in a mesio-distal axis. The slices were examined by polarized and light microscopy after hematoxylin-eosin or Goldner’s staining.

**Histomorphometric analysis**

Histomorphometric and histological analysis were performed by the same investigator (A.F.), who was blinded to the specific experimental conditions and validated by a second examiner (X.S). Also, a calibration procedure was performed before starting the analysis. Prior to histological preparation, three-dimensional reconstructions were made using a micro-CT scanner (SkyScan 1172, Bruker microCT, Kontich, Belgium). Images were obtained using a mode source at 80 kV/124 μA and a signal threshold set at 20. The rotation angle was 180°. NRecon software (SkyScan) was used for 3D reconstructions. Three levels of horizontal cuts were evaluated (apical/medium/coronal) for each furcation defect. We used a custom-made program, which was developed using image analysis software (QWin, Leica). The BCP particles, bone, and non-mineralized tissues were easily discriminated on micro-CT images based on their respective grey levels. The area of interest was manually defined based on the bottom of the defect apically, the two roots laterally, and the bottom of the furcation coronally (Fig. 2). Micro-CT reconstructions not only allowed for qualitative and quantitative analysis of NB formation, but also permitted qualitative evaluation of ceramic degradation. The respective surface of non-mineralized tissue (NM), biomaterial (B), and NB formation were calculated (in μm²). The bone regeneration ratio (BR) represents the relative ratio of mineralized tissue (B+NB) to the total surface area of interest (B+NB+NM), according to the following formula:  

\[
BR = \frac{(B+NB)}{(B+NB+NM)}
\]

For scanning electron microscopy (SEM), samples were sputtered with a thin layer of gold-palladium alloy (EM Scope, Laughton, England). SEM micrographs were obtained using the backscattered electron mode (BSEM) at 15 kV (SEM, LEO 1450 VP). SEM analysis through secondary electrons and BSEM allowed a qualitative investigation of NB formation and ceramic degradation following a vertical, bucco-lingual axis.

**Statistical analysis**

All data were expressed as mean and standard deviation (SD) or median and confidence interval (95% CI).
Nonparametric statistical tests were applied due to the relatively small sample size. Intragroup comparisons were made using the Friedman test for repeated measurements, whereas the Kruskal-Wallis test was used for relative ratio. Thereafter, the Mann-Whitney test and Bonferroni correction was used to evaluate the differences in the measured parameters among the groups. In some cases the “exact” option was performed to compensate the limited sample size. All statistical analyses were performed using commercial software (SPSS 18.0; SPSS, Chicago, IL, USA). The p-values<0.05 were considered as a strong tendency because of the small number of samples in each group and p-values<0.016 were considered to be statistically significant.

RESULTS

No complications were observed during the surgeries, and postoperative healing was uneventful in all of the dogs. Moreover, no allergic reactions, infectious complications, or premature exposure of membranes were observed throughout the entire study period.

Histological observations
The two conditions using BCP exhibited NB formation
in direct contact with the BCP granules, which were surrounded by osteoid tissue and osteoblast-like cells (Figs. 3 and 4). In the test group, less BCP granules were present in the center of the defect and only few were visible in the connective tissue in the buccal part of the defect (Fig. 5). In the test and in the reference treatment group, when the NB formation extended to the top of the furcation defect, a non-mineralized space was systematically observed; this separated the bone and the root surface (Fig. 6). The width of this non-mineralized space differed depending on the location (i.e., from about 50 μm along the root to around 200 μm at the roof level of the furcation). No signs of ankylosis or root resorption were observed.

Qualitative 3D CT and SEM analysis

Wound healing in the three conditions was characterized by variable bone regeneration in the defect area. 3D CT reconstructions revealed that NB formation was clearly less extensive in the control and the reference groups compared to the test group (BCP/Si-HPMC). In this test group, the extent of NB formation appeared to be quite homogeneous in the vertical and horizontal directions (Fig. 7). In the control group, NB was mainly located in the apical part and was in direct contact with the existing bone surrounding the defects. No complete healing or regeneration of these surgically created furcation defects were obtained in any specimen within the negative control group. In the reference group, NB was predominantly located in the apical or medial parts of the defects. In contrast, in the BCP/Si-HPMC group, NB was more extensive, reaching the top of the furcation and may completely filled the defect (Fig. 2).

The qualitative SEM study was correlated to the histological examination (Fig. 8). In the control group, the newly formed bone was mainly located in the apical part, decreasing progressively to the top of the defects; however, the NB never reached the top of the furcation. In the reference group (BCP/collagen membrane), a complete healing of the defect was never achieved, with the NB mainly located in the apical/medial parts of the defects. BCP acted as an osteoconductive material because newly formed mineralized bone could be observed in direct contact with the BCP granules. In the BCP/Si-HPMC test group, the NB reached the top of the defects; its density appeared to be greater in the apical/medial areas compared to the coronal areas of the defects. In addition, the degradation/resorption of the BCP appeared to be well advanced compared to the reference group, with only a few granules remaining in
Fig. 7 3D CT reconstructions of the three conditions after three months of healing. Note that NB formation is very limited in the negative control group (a) compared to the reference BCP/collagen membrane group (b) or the test group: BCP/hydrogel Si-HPMC membrane (c). In the BCP/Si-HPMC group, the bone regeneration occurred until the roof of the furcation defect.

Fig. 8 BSEM pictures of surgically created supra-alveolar periodontal defects: (a) negative control group, (b) reference BCP/collagen membrane group, (c) BCP/hydrogel Si-HPMC membrane group. From the base (blue line) of these surgically created critical-size defects, bone regeneration is limited in the control group (a). In the two conditions using BCP, the biomaterials exhibit osteoconductive properties, with NB surrounding the BCP. However, the kinetics of BCP resorption appears to be quite different in the test group BCP/hydrogel Si-HPMC membrane (c) compared to the reference BCP/membrane group (b) where BCP particles are more visible. In the test group, the BCP particles still visible are mainly located at the buccal face of the defects.

Fig. 9 Inter-group comparison of the total bone regeneration relative ratio (BR) while the graphs are plotted on the median value. The BCP/hydrogel Si-HPMC showed significantly increased levels of BR when compared to the negative control; while positive control demonstrated higher level of BR compared to the negative control and lower level when compared to the experimental material but following Bonferroni correction this was below statistical significance. The red lines indicate median value; *p<0.05; **p<0.016.

Histomorphometric analysis

The mean BR values based on the CT horizontal reconstructions are presented in Fig. 9, Tables 1 and 2. In the horizontal reconstructions, the BR was evaluated at three levels: apical, medial, and coronal areas of the defects. Also, the BCP granules were mainly concentrated at the external areas of the defects, predominantly at the level of the buccal and lingual walls. The NB was clearly more extensive in this test group and the reconstruction of the buccal osseous wall appeared more regular compared to that obtained with BCP and collagen membrane.

In conclusion, the inter-group comparison of the total bone regeneration relative ratio (BR) indicated that the BCP/hydrogel Si-HPMC showed significantly increased levels of BR when compared to the negative control; while positive control demonstrated higher level of BR compared to the negative control and lower level when compared to the experimental material but following Bonferroni correction this was below statistical significance.
Table 1  Descriptive statistics of histomorphometric parameters by the groups

<table>
<thead>
<tr>
<th></th>
<th>BCP/Collagen membrane</th>
<th>BCP/hydrogel Si-HPMC</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Coronal</td>
<td>Medial</td>
<td>Apical</td>
</tr>
<tr>
<td>Surface of BCP</td>
<td>12.61 (25.78)</td>
<td>42.79 (65.11)</td>
<td>12.61 (25.78)</td>
</tr>
<tr>
<td>Surface of new</td>
<td>103.28 (155.33)</td>
<td>170.98 (247.63)</td>
<td>915.89 (347.27)</td>
</tr>
<tr>
<td>bone</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Surface of non-</td>
<td>54.06 (75.62)</td>
<td>702.46 (203.90)</td>
<td>525.26 (349.07)</td>
</tr>
<tr>
<td>mineralized</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ratio of bone</td>
<td>0.01 (0.19)</td>
<td>0.18 (0.25)</td>
<td>0.68 (0.11)</td>
</tr>
<tr>
<td>regeneration</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

All values are expressed as mean values and (SD) of pixels×104.

Table 2  Intra and inter-group comparison of the bone regeneration relative ratio (BR)

<table>
<thead>
<tr>
<th></th>
<th>1. BCP/Collagen membrane</th>
<th>2. BCP/hydrogel Si-HPMC</th>
<th>3. Control</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.327 (0.133)</td>
<td>0.524 (0.104)</td>
<td>0.134 (0.095)</td>
</tr>
<tr>
<td></td>
<td>1&lt;3 p=0.025*</td>
<td>2&lt;3 p=0.004**</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Coronal</td>
<td>Medial</td>
<td>Apical</td>
</tr>
<tr>
<td></td>
<td>0.009 (0.191)</td>
<td>0.368 (0.106)</td>
<td>0.061 (0.136)</td>
</tr>
<tr>
<td></td>
<td>1/3 p=0.262</td>
<td>2&lt;3 p=0.010**</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Medial</td>
<td>Apical</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.176 (0.248)</td>
<td>0.579 (0.095)</td>
<td>0.097 (0.140)</td>
</tr>
<tr>
<td></td>
<td>1/3 p=0.522</td>
<td>2&lt;3 p=0.004**</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Apical</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.676 (0.106)</td>
<td>0.624 (0.071)</td>
<td>0.243 (0.169)</td>
</tr>
<tr>
<td></td>
<td>1&lt;3 p=0.004**</td>
<td>2&lt;3 p=0.004**</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Intra-group</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Apical&gt;Coronal p=0.006*</td>
<td>Apical&gt;Coronal p=0.006*</td>
<td>Apical&gt;Coronal p=0.037*</td>
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<tr>
<td></td>
<td>Apical&gt;Medial p=0.004*</td>
<td>Apical/Medial p=0.722</td>
<td>Apical/Medial p=0.286</td>
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<tr>
<td></td>
<td>p-value</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Medial/Coronal p=0.855</td>
<td>Medial/Coronal p=0.004*</td>
<td>Medial/Coronal p=0.355</td>
</tr>
</tbody>
</table>

NS—not significant, *p<0.05; **p<0.016

furcation defects. The BR was 0.134±0.095 (n=6) in the control group, 0.327±0.133 (n=6) in the reference group and 0.524±0.104 (n=6) in the BCP/Si-HPMC test group. Notably, a significant increase in BR was observed in the two groups using BCP (i.e., reference BCP/collagen membrane and BCP/ hydrogel membrane Si-HPMC) in comparison to the control group (p=0.025 and p=0.004, respectively). The total BR was more important in the test group BCP/Si-HPMC, compared to the reference treatment (BCP/collagen membrane) and the results were significant (p=0.025).

In all conditions, a decrease in NB formation was found from the base to the top of the surgically created furcation defects. In fact, the BR appeared to be quite similar in the apical part of the defects for the two groups using BCP (BCP/membrane, BCP/HPMC, BCP/Si-HPMC), and the results were significant when compared to the control group (p=0.004 and p=0.004, respectively). At the apical level, the reference group showed the best results, but the BR decreased dramatically in the medial level (BR decreased from 0.676 [apical] to 0.176 [medial] (p=0.004). At the medial level of the furcation defect, a significant difference in BR was observed for the test group BCP/Si-HPMC, in comparison to the control group (p=0.004) and the reference group (p=0.010). For the test group, NB formation was less important in the medial part compared to the apical part of the defect, but the difference was not significant. At the coronal level, the NB formation appeared very limited in the control and reference groups compared to the test group. At the coronal level, NB formation was the most intensive in the BCP/Si-HPMC group. The increase in BR was significant compared to the control group (BR: 0.368 vs. 0.061; p=0.010) and the reference group (BR: 0.368 vs. 0.009; p=0.037).
DISCUSSION

In the present study, our aim was to evaluate bone regeneration in supraalveolar periodontal defects on mandibular premolars, filled with BCP granules and a collagen membrane or with BCP granules and an hydrogel of Si-HPMC injected over the graft material, acting after reticulation as a barrier membrane. The supraalveolar periodontal defect in dogs represents a well-documented critical-size model with incomplete spontaneous healing. It is a valuable model for evaluating bone-graft materials alone or in combination with bioactive agents\(^3\). In these surgically created, critical sized defects, the retention of the bone graft material and the stability of the clot are crucial parameters. Therefore, this preclinical model is appropriate for evaluating new generations of biomaterials designed to facilitate the surgical handling and initial wound stability\(^4\).

In the control group, spontaneous bone regeneration was very limited and mainly occurred in the apical part of the defects, which was in direct contact with the basal osseous area. More coronally, NB formation decreased significantly and soft tissues collapsed into the basal osseous area. More coronally, NB formation never reached the roof of the defects. The NB formation never reached the roof of the defect. The BR was significantly lower compared to the reference group. The BR was significantly higher compared to the control group leading to a more intensive intergranular colonization during the initial stages of healing. At 3 months, a stronger bone remodeling was observed in the inner part of the defect in the test group (BCP+Si-HPMC membrane) compared to the positive control group (collagen membrane+BCP), in this kind of very complex and unfavorable surgically created periodontal defects.

No adverse inflammation, foreign body reactions, ankylosis, or root resorption were noted in any specimen. Such a large difference in BR between the reference and BCP/Si-HPMC groups was unexpected, as these two conditions were quite similar. They both utilized the same BCP and a resorbable membrane (i.e., collagen or hydrogel) to cover the bone graft. It is possible that the difference in BR can be attributed to the difference in the kinetics of degradation displayed by collagen and hydrogel membranes. The gelation process of the Si-HPMC may delay cell and tissue colonization\(^29,30\), Slow degradation of the Si-HPMC\(^46\) may prevent invagination of connective tissue into the defect, which could maintain cohesion of BCP granules and the blood clot for a longer period. Moreover, the resorption of the BCP granules was completely different between the two conditions. In the reference group, a limited resorption of the BCP was observed, whereas a more pronounced resorption was seen in the BCP/Si-HPMC group. These results seem to be in accordance with those published in recent research\(^47,48\) where the cells recruited into the membrane expressed signals for bone regeneration (BMP-2, FGF-2, TGF-b1 and VEGF). The resorbable membrane could act...
as a bioactive compartment rather than a passive barrier. In another recent study (49), the authors observed a foreign body giant cells reaction at the level of 2 new porcine dermis-derived collagen membranes which could affect bone regeneration. In a previous study, we used smaller sized BCP particles and an Si-HPMC membrane to treat maxillary furcation defects (32). However, the viscosity of the hydrogel used in the study did not efficiently maintain the BCP in the defect area during the surgical phase. Therefore, in the present study, we increased the concentration of the hydrogel from 3 to 4% with an increased viscosity (38). Thus, the Si-HPMC polymer was easier to handle during the surgical phase and may have acted as an occlusive membrane after gelation. The significant difference in bone regeneration between the BCP/Si-HPMC test group and the control or the reference groups indicates that Si-HPMC hydrogel may represent a valuable regenerative membrane for covering bone-graft material and enhancing bone regeneration. The hydrogel Si-HPMC may also act as a carrier for stem cells (30). Even though it shows good biocompatibility and soft tissue tolerance (28,38), the mechanical properties of Si-HPMC after reticulation can still be optimized in order to enhance resistance to stresses.

Our methodology focused on qualitative and quantitative evaluations of newly formed bone in a critical-sized model. Following non-decalcified inclusion, the histological examinations could not precisely evaluate the nature of the periodontal attachment. In the two conditions, no signs of ankylosis or root resorption were observed, and non-mineralized tissue was systematically present between the root surface and the newly formed bone. Further pre-clinical studies will be needed to investigate the occurrence of true periodontal regeneration.

In conclusion, the results of this study in dogs has suggested that Si-HPMC hydrogel may act as an occlusive barrier that supports bone regeneration. Future clinical trials in humans will be needed to validate the results of this study.

CONFLICTS OF INTEREST AND SOURCE OF FUNDING STATEMENT

The authors declare that they have no conflicts of interest. The study biomaterials were kindly provided by Biomatlante, Vigneux de Bretagne, France.

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