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ORIGINAL ARTICLE

Influence of ridge preservation procedures on extraction socket healing under antiresorptive therapy: An experimental study in rabbits

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Abstract

Background: To assess the influence of ridge preservation procedures on the healing of extraction sockets under antiresorptive therapy.

Material and Methods: A total of 10 Dutch Belted rabbits were randomly allocated to either the intravenous administration of amino-bisphosphonate (zoledronic acid) (Za) (n = 5) or a negative control group (no Za [nZa]) (n = 5). At 6 months, the mandibular and maxillary molars were extracted and the four experimental sites randomly allocated to the following subgroups: (a) socket grafting using a collagen-coated natural bone mineral (BOC) + primary wound closure, (b) coronectomy (CO), or (c) spontaneous healing + primary wound closure (SP). Za medication was continued for another 4 months. Histomorphometrical analyses considered, for example, crestal hard tissue closure of the extraction site (C) and mineralized tissue (MT) formation.

Results: Za-SP was associated with an incomplete median C (31.76% vs 100% in nZa-SP) and signs of bone arrosion along the confines of the socket. BOC had no major effects on increases in C and MT values in the Za group. CO commonly resulted in an encapsulation and partial replacement resorption of residual roots by MT without any histological signs of osteonecrosis.

Conclusions: (a) Za-SP was commonly associated with a compromised socket healing and signs of osteonecrosis, (b) BOC had no major effect on socket healing in the Za group, and (c) CO at noninfected teeth might be a feasible measure for the prevention of a Za-related osteonecrosis of the jaw.

KEYWORDS

animal experiment, antiresorptive agents, extraction socket healing, histological technic, ridge preservation

1 | INTRODUCTION

The management of patients receiving antiresorptive drugs has become a major issue in dentistry. In particular, these drugs may be associated with a medication-related osteonecrosis of the jaw (MRONJ), which is more likely to appear in the mandible 73% (22.5%) than in the maxilla.[1] Tooth extraction has been classified as a critical trigger for the occurrence of MRONJ, since the associated risk was about 16-fold increased when patients were under antiresorptive therapy.[1-3] A potent drug is the amino-bisphosphonate zoledronic-acid (Za), which is gaining importance in the therapy of osteoporosis due to its long-lasting effect on increases in bone mineral density after intravenous application.[4] Experimental animal studies provide some evidence that Za was significantly correlated with the occurrence of "exposed bone without epithelial coverage, decreases in the number of osteocytes, and increases in necrotic bone with more empty lacunae."[5]

Since the pathophysiology associated with the occurrence of MRONJ is still far from being completely understood, contemporary preventive measures focus on the atraumatic tooth extraction. In particular, a modified protocol, including coronectomy and the intentional retention of the endodontically treated root, has been proposed as a potential alternative approach to reduce the individual risk in some specific cases.[6] So far, however, there is no scientific evidence to support this clinical procedure. Another potential preventive measure may be related to the management of extraction sockets by employing alveolar ridge preservation (RP). This concept has been proven to be an effective therapeutical procedure to limit physiological remodeling processes following tooth extraction.[7] It is hypothesized that coagulum stabilization by means of a bone substitute[8] may also be considered as a reasonable preventive approach to limit the occurrence of MRONJ.

Therefore, the aim of the present study was to assess the influence of different RP procedures on the healing of extraction sockets under antiresorptive therapy in a rabbit model.

2 | MATERIAL AND METHODS

2.1 | Animals

Ten skeletally mature, female Dutch Belted rabbits (age: 6 months, mean weight 2.2 ± 0.5 kg) obtained from a certified breeder (Covance Research Products, Inc., Denver, Pennsylvania) were used in the study. All animals were housed in appropriately dimensioned cages (5400 cm^2, height 60 cm) under standard conditions of temperature in a light-controlled environment and were provided water and special diet ad libitum. The study protocol was approved by the appropriate local authority (Landesamt für Natur und Verbraucherschutz, Recklinghausen, Germany) and the current reporting followed the ARRIVE Guidelines.

2.2 | Study design

The rabbits were randomly allocated to either the intravenous application of amino-bisphosphonate Za (50 μg/kg) (Zoledronate, 4 mg/5 mL, Teva, Harlem, Netherlands) (baseline and after at least 6 months) or an untreated control group (no Za [nZa]), including five animals each (block randomization). After 6 months, the mandibular and maxillary molars were extracted and the four experimental sites randomly allocated to the following treatment procedures: (a) RP using a collagen-coated bovine bone mineral (Geistlich BioOss Collagen, Geistlich Biomaterials, Switzerland) (BOC) + primary wound closure, (b) coronectomy (CO), or (c) spontaneous healing + primary wound closure (SP) (permuted block randomization BOC, CO, 2xSP). Allocation of the treatment procedures in each animal was accomplished using a computer-generated list (Randlist, DatInf GmbH, Tübingen, Germany). All sites were left to heal for a period of 4 months.

2.3 | Anesthesia protocol

All surgical interventions followed a standardized anesthesia protocol. In particular, the premedication included 25 mg/kg ketamine and 5 mg/kg xylazine administered via the ear vein. Following orotracheal intubation, anesthesia was maintained during the operation by inhalation of an oxygen-isoflurane mixture (1-1.5 vol%). Analgesia (4 mg/kg carprofen) was provided pre- (intravenous) and postoperatively for 5 days (intramuscular).

2.4 | Surgical procedures

At BOC and SP sites, tooth extraction was gently performed without raising a flap and compromising the vestibular/oral bone walls. Subsequently, the sockets were curetted and rinsed with sterile saline. BOC was rehydrated in saline and gently packed without excess of pressure at respective sites. At SP sites, the formation of a blood clot was supported by a gentle curettage of the adjacent mucosa. At both BOC and SP sites, a primary wound closure of the socket entrance was accomplished by means of interrupted resorbable single sutures (8-0).

At CO sites, the respective crowns were carefully separated in a supracrestal position using forceps. The exposed part of the pulp was carefully removed and the entrance sealed with a temporary filling material (Cavit, 3M ESPE, Neuss, Germany). The surgical procedures were accomplished by two previously calibrated surgeons (F. S. and I. M.).

2.5 | Histological processing

The animals were euthanized with an overdose of pentobarbitone at 600 mg/kg.

The jaws were carefully dissected to obtain tissue blocks containing the respective experimental sites. All specimens were fixed in 10% neutral buffered formalin solution for 10 days.

Histological processing followed a standardized procedure established for hard tissue sectioning.[9] In brief, all retrieved tissue blocks were dehydrated using ascending grades of alcohol and xylene and subsequently infiltrated and embedded in methylmethacrylate (Technovit 9100 NEU, Heraeus Kulzer, Wehrheim, Germany) and prepared for non-decalcified sectioning. Each block was cut in the vestibulo-oral direction along with the long axis of the extraction site using a diamond wire saw (Exakt, Apparatebau, Norderstedt, Germany). The most central section of each extraction site was ground to a final thickness of approximately 40 μm and stained with toluidine blue.

2.6 | Histological and histomorphometrical analysis

Digital images (original magnification ×200) were obtained from each specimen and evaluated using a software program (Image J 1.51s, National Institutes of Health, Bethesda).

The following landmarks were identified in the stained sections (Figure 1): the width (*W*) of the alveolar crest measured between the most coronal outer aspects of both vestibular (v) and oral (o) bone walls; the bone thickness at the v and o crestal bone walls (BTv and BTo); the vertical perpendicular linear distance between BTv and BTo (BTvBTo); the closure (C) measured as the linear horizontal distance of newly formed bone spanning the width of the former extraction socket; the height (*H*) as measured perpendicularly from the width landmark to the inner confines of the cortical bone; the total area (TA) measured as the inner confines of the former extraction site (ie, noncortical bone areas). Within TA, the cross-sectional surface area of mineralized (MT) and nonmineralized tissue was assessed (Figure 1).

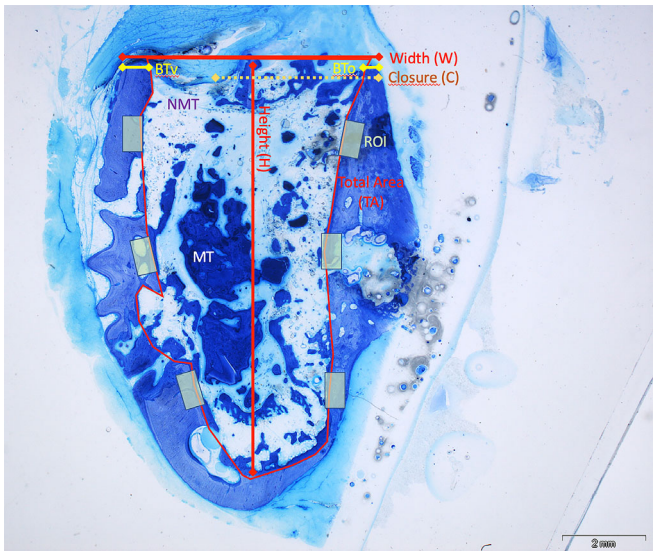

FIGURE 1 Histological landmarks defined at both vestibular and oral aspects (specimen of the Za-BOC group). BTo, bone thickness oral; BTv, bone thickness vestibular; C, linear horizontal distance of newly formed bone spanning width of the alveolar crest; *H*, height measured as the most apical extension of the former extraction site; MT, mineralized tissue within TA; NMT, nonmineralized tissue within TA, ROI's for the assessment of osteocyte and empty osteocytic lacunae; TA, surface area of the former extraction site; *W*, width of the alveolar crest

Osteocytes (OC) and empty osteocytic lacunae (EOL) were counted within six rectangles (region of interest (ROI) of 0.35 mm^2), randomly selected within the upper, middle, and lower portions of *H* at both v and o aspects. The OC and EOL density (d) was determined by calculating their ratio relative to the total number of OC + EOL in the respective ROIs (Figure 1).

All measurements were performed by two previously calibrated examiners (F. S. and G. J.). Calibration was accepted when repeated measurements of n = 5 different sections were similar at >95% level.

2.7 | Statistical analysis

The statistical analysis of the data sets was accomplished using a commercially available software program (IBM SPSS Statistics 26.0, IBM Corp., Armonk, New York).

Mean values, SDs, and medians were calculated for all parameters in different groups (ie, Za and nZa) and subgroups (ie, BOC, CO, SP) defining the animal as statistical unit. The data rows were examined with the Shapiro-Wilk test for normal distribution. Within subgroup, comparisons were accomplished using analysis of variance (ANOVA) employing Bonferroni corrections to account for multiple testing. For between group comparisons, the unpaired *t* test was used. The alpha error was set at 0.05.

3 | RESULTS

3.1 | Histomorphometrical analysis

The histomorphometrical analysis (mean ± SD) of *H*, *W*, *C*, BTv, BTo, BTv/BTo as well as TA, NMT, and MT values in different groups and subgroups are presented in Table 1.

In both Za and nZa groups, median *H* and *W* values were commonly highest at CO (Za—*H*: 6.29, *W*: 3.65; C—*H*: 6.80, *W*: 3.90 mm) and BOC (Za—*H*: 5.94, *W*: 3.44; C—*H*: 5.95, *W*: 3.46 mm) treated sites. In contrast, SP was associated with a marked reduction of both values (Za—*H*: 4.14, *W*: 2.69; nZa—*H*: 3.90, *W*: 2.27 mm), reaching statistical significance when compared with CO in the nZa group ($P = .029$) (Table 1).

Within group analyses revealed comparable median *C* values for all subgroups in both Za and nZa groups (ANOVA; $P > .05$, respectively). In particular, median *C*% values in the Za group were 39.04, 76.27, and 31.76 in subgroups BOC, CO, and SP, respectively. When compared with the Za group, these values were markedly higher in the nZa group, amounting to 84.53, 61.17, and 100.0 in subgroups BOC, CO, and SP, reaching statistical significance for BOC (unpaired *t* test; $P = .03$).

TABLE 1 Histomorphometrical analysis (mean ± SD) of *H* (mm), *W* (mm) C (% of *W*), BTv (mm), BTo (mm), BTv/BTo (mm), TA (mm^2), NMT (mm^2), and MT (mm^2) values in different groups and subgroups (n = 10 animals, n = 16 sites)

Subgroup	*H*	*W*	*C*%	BTv	BTo	BTv/BTo	TA	NMT	MT
a. Za group									
BOC (n = 7)	5.53 ± 2.15	3.58 ± 1.28	46.13 ± 28.0	0.66 ± 0.15	0.53 ± 0.13	0.27 ± 0.21	14.98 ± 9.79	5.04 ± 3.15	9.93 ± 7.53
CO (n = 5)	6.78 ± 1.78	3.76 ± 0.51	58.88 ± 40.05	0.70 ± 0.21	0.56 ± 0.12	0.77 ± 0.84	21.46 ± 3.56	10.43 ± 8.10	11.02 ± 6.81
SP (n = 4)	4.72 ± 1.65	2.82 ± 0.61	44.84 ± 37.76	0.49 ± 0.09	0.56 ± 0.10	0.25 ± 0.29	14.42 ± 12.24	8.52 ± 9.83	5.90 ± 3.38
b. nZa group									
BOC (n = 7)	5.99 ± 0.70	3.33 ± 1.00	82.10 ± 17.01	0.61 ± 0.21	0.75 ± 0.13	1.53 ± 1.26	12.55 ± 4.64	3.75 ± 1.30	8.79 ± 5.10
CO (n = 5)	6.97 ± 1.84	3.81 ± 0.52	69.37 ± 24.88	0.37 ± 0.12	0.63 ± 0.12	1.58 ± 1.13	18.43 ± 8.28	6.77 ± 3.22	11.65 ± 6.65
SP (n = 4)	4.78 ± 1.56	2.25 ± 0.15	68.42 ± 54.69	0.46 ± 0.08	0.59 ± 0.09	0.38 ± 0.38	9.57 ± 3.91	4.79 ± 1.89	4.78 ± 3.97

Abbreviations: BTo, bone thickness oral crestal bone wall; BTv, bone thickness vestibular crestal bone wall; CO, coronectomy; MT, mineralized tissue; NMT, nonmineralized tissue; SP, spontaneous healing + primary wound closure; TA, total area.

Within group comparisons revealed similar BTv, BTo, and BTv/BTo values for all subgroups investigated (ANOVA; $P > .05$, respectively). Between group comparisons, however, revealed significant differences for BTv (nZa—CO: 0.42 vs Za: 0.66 mm, unpaired *t* test; $P = .009$), BTo (nZa—BOC: 0.79 vs Za: 0.55 mm, unpaired *t* test; $P = .021$), and BTv/BTo (nZa—BOC: 2.26 vs Za: 0.29 mm, unpaired *t* test; $P = .025$) values, respectively (Table 1).

Between group comparisons revealed similar TA, NMT, and MT values in all subgroups investigated (unpaired *t* test; $P > .05$, respectively). In both Za and nZa groups, CO and BOC tended to be associated with higher TA (Za—CO: 23.12, BOC: 13.44; nZa—CO: 20.15, BOC: 13.90 mm) and MT (Za—CO: 12.21, BOC: 6.97; nZa—CO: 11.94, BOC: 9.37 mm) values when compared with SP (Za—TA: 10.29, MT: 6.06; nZa—TA: 9.49, BOC: 6.85 mm) sites. These differences, however, failed to reach statistical significance in the within group comparisons (ANOVA; $P > .05$, respectively) (Table 1).

The histomorphometrical analysis (mean ± SD) of OC, EOL, total (OC + EOL), OC density (OCd), and EOL density (EOLd) values in different groups and subgroups are presented in Table 2.

In the nZa group, median OCd and EOLd were comparable in BOC (0.87; 0.13), CO (0.87; 0.13), and SP (0.87; 0.13) subgroups (ANOVA; $P > .05$, respectively). In the Za group, SP was associated with a significant decrease in OCd and increase in EOLd values (0.80; 0.20) when compared with subgroups BOC (0.89; 0.11) and CO (0.90; 0.10) (ANOVA; $P = .001$, respectively) (Table 2). Between group comparisons revealed a significant difference for mean OCd and EOLd values in subgroups CO (unpaired *t* test; $P = .032$; $P = .032$) and SP (unpaired *t* test; $P = .04$; $P = .04$), respectively.

3.2 | Histological analysis

Representative histological views in different groups and subgroups are shown in Figures 2 to 4.

3.2.1 | Subgroup SP

In the nZa group, SP specimens were commonly associated with an undisturbed wound healing. This was evidenced by the presence of an intact, non-infiltrated, and regularly structured oral mucosa covering a crestal bone bridge spanning the former extraction site (Figure 2A). While this bone bridge had a more cortical nature in the lower jaws, its composition comprised a higher amount of woven bone in the upper jaws. The confines of the former extraction socket were partially dissolved and merged with the adjacent compartments of the alveolar ridge. The central and lateral aspects of TA underneath the bone bridge featured a non-infiltrated NMT (ie, bone marrow spaces) and MT areas exhibiting signs of an ongoing bone remodeling (Figure 2B). In the Za Group, SP specimens mainly featured an incomplete or the absence of a crestal bone bridging (Figure 2C). This was commonly associated with an ulceration of the covering oral mucosa, featuring histological signs of a mixed chronic inflammatory cell

TABLE 2 Histomorphometrical analysis (mean ± SD) of OC, EOL, total (OC + EOL), OCd, and EOLd in different groups and subgroups (n = 10 animals, n = 16 sites)

Subgroup	OC	EOL	Total	OCd	EOLd
a. Za group					
BOC (n = 7)	96.57 ± 18.11	11.63 ± 1.48	108.20 ± 19.00	0.89 ± 0.01	0.11 ± 0.01
CO (n = 5)	97.00 ± 86.00	11.01 ± 2.04	108.01 ± 22.55	0.89 ± 0.00	0.10 ± 0.00
SP (n = 4)	113.75 ± 15.321	28.75 ± 4.78	142.51 ± 19.53	0.79 ± 0.01	0.20 ± 0.01
b. nZa group					
BOC (n = 7)	124.80 ± 20.12	17.06 ± 2.45	141.86 ± 21.86	0.87 ± 0.01	0.12 ± 0.01
CO (n = 5)	114.00 ± 25.37	16.01 ± 4.13	130.01 ± 29.17	0.87 ± 0.01	0.12 ± 0.01
SP (n = 4)	125.00 ± 10.14	22.37 ± 9.64	147.37 ± 18.50	0.85 ± 0.04	0.15 ± 0.04

Abbreviations: CO, coronectomy; EOL, empty osteocytic lacunae; OC, osteocytes; SP, spontaneous healing + primary wound closure; OCd, OC density; EOLd, EOL density.

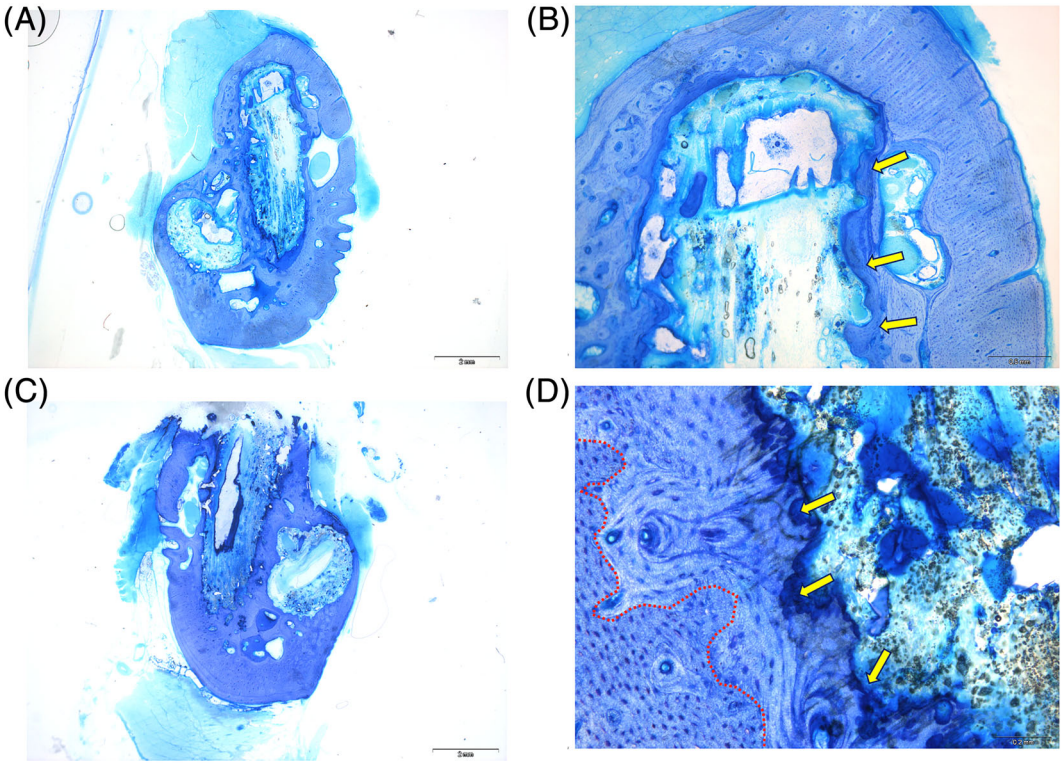

FIGURE 2 Representative histological views of wound healing in the SP subgroups. A, The formation of a crestal bone bridge spanning the former extraction socket was a common feature in the nZa group. B, Arrows depicting histological signs of an ongoing bone remodeling process at the confines of the former extraction socket (nZa group). C, An incomplete crestal bone bridge and associated ulceration of the adjacent oral mucosa was a common feature in the Za group. D, Arrows depicting obvious signs of bone arrosion, which is followed by a osteonecrosis zone (arrowheads) characterized by the reduced number of osteocytes and numerous empty lacunae (Za group). SP, spontaneous healing + primary wound closure

infiltrate in the subepithelial connective tissue compartment. The confines of the formed extraction socket mainly featured histological signs of a bone arrosion and osteonecrosis without any noticeable signs of a bone remodeling (Figure 2D).

3.2.2 | Subgroup BOC

In the nZa group, BOC specimens also commonly featured a coverage of the former extraction site by a non-infiltrated and well-organized oral mucosa. The presence of a crestal bone bridge was a main histological feature in the majority of the specimens investigated, exhibiting structural components (ie, cortical and more spongious compounds) similar to those noted in the respective SP subgroup (Figure 3A). TA areas featured varying amounts of residual BOC particles, which were commonly embedded in a newly formed network of a parallel fibered woven bone exhibiting signs of an ongoing bone remodeling. Some BOC particles were also dispersed in the adjacent subepithelial connective tissue and revealed histological signs of an encapsulation (Figure 3B). In the Za Group, BOC specimens exhibited

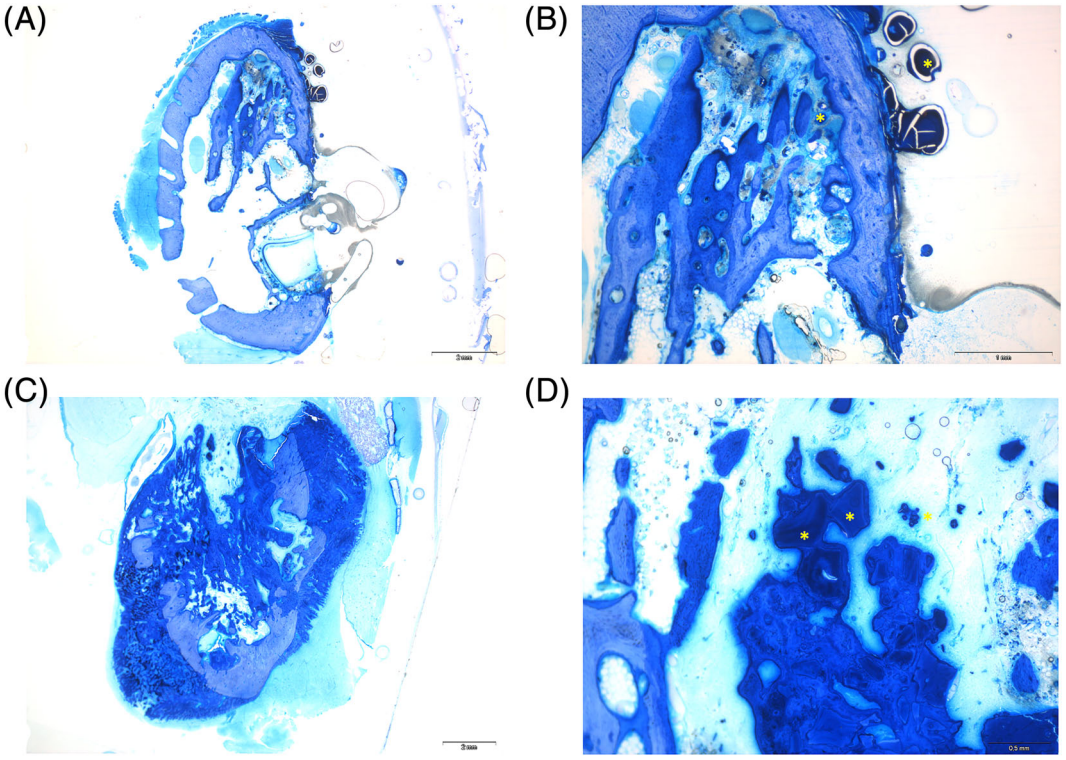

FIGURE 3 Representative histological views of wound healing in the BOC subgroups. A, The establishment of a crestal bone bridge was also a common feature in the nZa group. B, Asterisks depicting residual BOC particles which were either embedded in a network of MT or encapsulated when dispersed in the adjacent mucosa (nZa group). C, Specimen of the Za group showing an incomplete hard tissue bridge but pronounced signs of bone remodeling. D, Asterisks depicting residual BOC particles surrounded by parallel fibered woven bone (Za group). MT, mineralized tissue

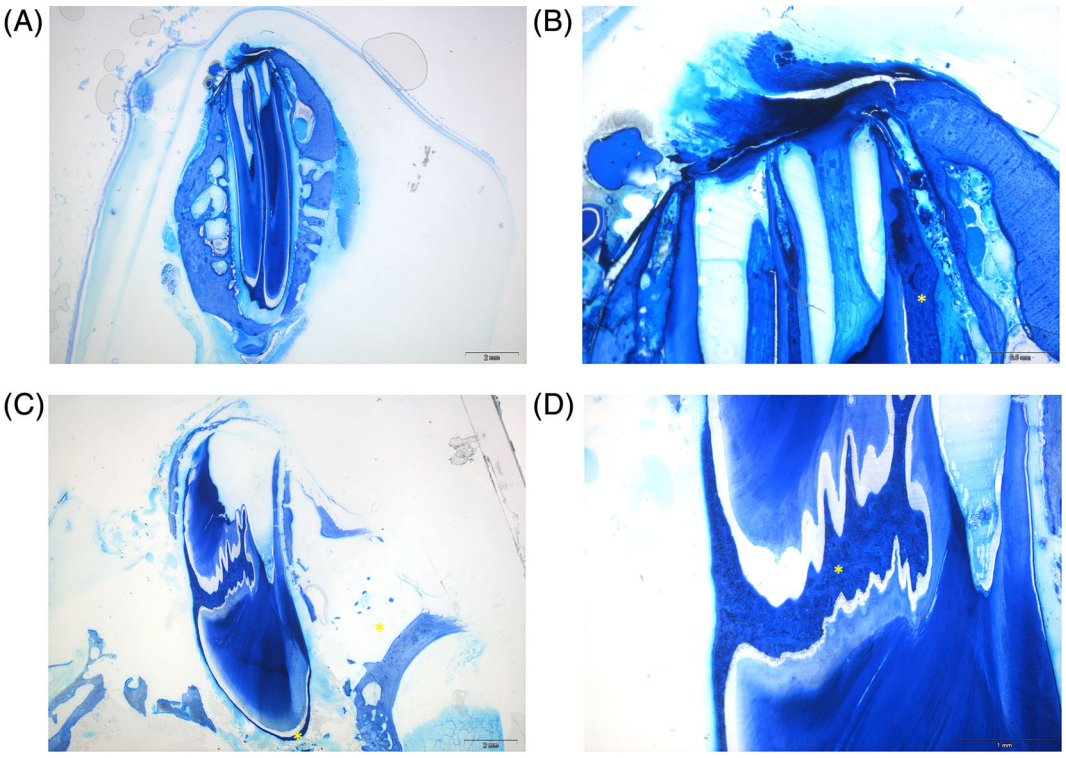

FIGURE 4 Representative histological views of wound healing in the CO subgroups. A, The establishment of a thin crestal bone bridge was merely noted at two sites (nZa group). B, Asterisks depicting the formation of a thin layer of newly formed bone along the residual root (nZa group). C, Asterisks depicting the apical extension of newly formed bone along the exposed root (Za group). D, Asterisks depicting a replacement resorption at the central aspect of the root (Za group). CO, coronectomy

major variations in the quality of the extraction socket healing. While all sites featured a complete soft tissue coverage, the formation of a hard tissue bridge was commonly incomplete and mainly based on thin areas of immature woven bone. Similar to the nZa group, the central and lateral compartments of TA featured residual BOC particles. The amount of newly formed bone surrounding and embedding these particles, however, markedly varied within this subgroup (Figures 1 and 3C). In contrast to the corresponding SP group, all specimens investigated featured an ongoing bone remodeling in the absence of any histological signs of bone arrosion or osteonecrosis (Figure 3D).

3.2.3 | Subgroup CO

In both nZa and Za groups, all CO specimens investigated revealed almost identical histological features of the associated wound healing processes. In particular, the experimental sites were associated with either a complete or an incomplete coverage of a non-infiltrated oral mucosa. The formation of a hard tissue bridge spanning the crestal aspect of the alveolar ridge was merely observed in two specimens (Figure 4A,B). At the majority of the sites, residual roots were surrounded and encapsulated by a newly formed thin layer of parallel-fibered woven bone, even extending to the most apical regions. Occasionally, this was associated with signs of a replacement resorption (Figure 4C,D). Both nZa and Za groups did not reveal any histological signs of bone arrosion or osteonecrosis along or adjacent to the confines of the socket.

4 | DISCUSSION

The present study was designed to histologically assess the influence of BOC and CO on the healing of extraction sockets in an established animal model. In fact, previous studies also employed rabbits for RP procedures at molar sites and confirmed the suitability of this model for the assessment of extraction socket healing.[10,11]

Basically, the histological analysis revealed that the administration of Za was associated with an impaired wound healing, as evidenced by an ulceration of the covering oral mucosa, an incomplete or absent crestal bone bridging, a reduced MT formation as well as signs of bone arrosion and osteonecrosis affecting the confines of the former extraction socket. This proven persistence of the extraction socket basically fulfills the current requirements for the clinical and radiographic diagnosis of MRONJ.[12,13] In this context, it must be emphasized that the applied dosage of Za has previously been proven to result in the development of MRONJ in rabbits[14] and rodents.[5,15-19] Interestingly, the first structural changes in microtomographic bone parameters were already observed at 3 weeks following administration of 66 μg/kg Za.[15] Tooth extraction under Za was commonly associated with an impaired socket healing as defined by a high incidence of osteonecrosis, inflammatory cell infiltrates, and incomplete coverage by mucosal tissue after a follow-up period of up to 8 weeks.[5,14,16-20] The findings noted in the present Za-SP group clearly corroborate the results of the latter studies, since similar histopathological features also suggested a high incidence of MRONJ at 4 months after tooth extraction. In particular, nZa-SP sites were associated with markedly higher *C*, MT, as well as significantly higher OCd and lower EOLd values, supporting the histological findings of an impaired remodeling of the "inner" socket compartment as noted at Za-SP sites. Conversely, Za-SP sites were associated with markedly lower BTv/BTo and higher *H* as well as TA values, pointing to less pronounced dimensional changes of the "outer" contour of the alveolar ridge as opposed to nZa-SP sites. In fact, previous data employing a canine model also clearly differentiated between a phase 1, mainly affecting the inner remodeling of the extraction socket and a phase 2, mainly affecting the outer surfaces of the alveolar ridge.[21] These potential effects of Za on the dynamics of extraction socket healing need to be further elucidated in future studies.

When further evaluating the present findings, it was noted that the application of BOC had no major effect on the histomorphometrical outcomes assessed within either Za or nZa groups, respectively. However, between group comparisons revealed significantly higher *C*, BTo and BTv/BTo values in the nZa-BOC as opposed to the Za-BOC group, respectively. These findings also corroborate the aforementioned assumption of less pronounced dimensional changes of the "outer" contour of the alveolar ridge under Za medication. As opposed to the corresponding Za-SP group, Za-BOC tended to be associated with higher MT values, which in turn supports the histological findings of a more pronounced bone remodeling, in the absence of osteonecrotic areas, at respective sites. One potential explanation for the slight improvement of the "inner" healing under Za medication might be due to a potential stabilization of the blood clot by the BOC related 10% types I and III collagen matrix.[22] In fact, both types of collagen were proven to play a crucial role in hemostasis due to a direct or indirect interaction with platelet receptors, thus mediating their adhesion and aggregation.[23] In a canine model without Za medication, however, BOC delayed the initial bone formation when compared with nongrafted control sites at 2 weeks.[24] Similar findings were also noted at 4 to 6 weeks after the application of BO (ie, BOC without collagen matrix) in extraction sockets of rabbits.[11] After 6 months in a canine model, BOC also failed to enhance bone formation over the control sites, but resulted in a reduction of the outer contraction of the alveolar ridge.[25] The latter finding could, however, not be observed in the present histological analysis, since BOC resulted in comparable (ie, Za group) and even significantly higher (ie, nZa group) BTv/BTo values over SP sites.

When further evaluating the results of the present study, it must be emphasized that these are the first histological data on the healing processes associated with CO under either nZa or Za conditions. CO was however proven to be a safe approach for an alternative removal of impacted teeth in either oncological patients[26] or in cases revealing a close proximity to the inferior alveolar nerve.[27] Clinical data provide some evidence that CO was associated with a similar low frequency of postoperative infections but a reduced risk for the occurrence of a dry socket when compared with a complete molar removal. Moreover, it was observed that the residual root fragments migrated by about 2 mm in a coronal direction within a period of 2 years.[27] The

avoidance of a dry socket might play a key role in the prevention of MRONJ. In fact, the present histological analysis did not reveal any characteristics of MRONJ in the Za-CO groups. The latter finding was supported by significantly higher OCd and lower EOLd values in Za treated animals when compared with the respective nZa subgroup. Moreover, between group comparisons also pointed to significant differences in mean BTv, which—along with the noted differences in OCd and EOLd values—might be attributed to anatomical variations among the animals. Interestingly, the root fragments in both Za and nZa groups were almost entirely surrounded by a thin layer of newly formed bone, occasionally resulting in a replacement resorption. The latter findings are at least in part supported by previous experimental studies, also indicating that tooth roots that were used for lateral ridge augmentation, were gradually involved in the bone remodeling process and associated with a replacement resorption.[28,29] When evaluating the results noted in the CO group, it must however be emphasized that these teeth revealed neither an endodontic nor a periodontal infection, which in turn may not reflect the known influence of oral infections on the onset and progression of MRONJ.[30,31] Another potential limitation of the present analysis is related to the lack of immunohistochemical analyses or specific staining techniques (eg, staining of tartrate-resistant acid phosphatase in osteoclasts) to further assess bone remodeling processes in different groups. This was technically limited by the non-decalcified sectioning of the tissue samples which, however, offered a better depiction of the mineralized and cellular components of the alveolar bone over decalcified paraffin-embedded sectioning.[32]

5 | CONCLUSIONS

Within its limitations, the present study has indicated that (a) Za-SP was commonly associated with a compromised socket healing and signs of osteonecrosis, (b) BOC had no major effect on socket healing in the Za group, and (c) CO at noninfected teeth might be a feasible measure for the prevention of a Za related osteonecrosis of the jaw.

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CONFLICT OF INTEREST

The authors declare no potential conflict of interest.

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