THE REPURCUSSIONS OF POLYDEOXYRIBONUCLEOTIDE ON THE INITIAL PHASES OF BONE GROWTH DURING LATERAL WINDOW SINUS FLOOR A RISE AND SIMULTANEOUSLY PLACEMENT OF IMPLANTS

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Abstract

Purpose: Modern orthopaedic and maxillofacial procedures always call for a quick bone tissue regeneration to fill in surgically generated gaps or quickly boost bone implant fixation.

Methodology: 2 months prior to lateral sinus floor elevation, 3 bimaxillary premolars (P2, P3, and P4) were retrieved from 4 individuals. Each sinus was assigned either the test group or the control group after the lateral elevation of the sinus membrane. Upon lateral sinus floor elevation, the choice of a collagenated synthetic bone graft with PDRN (test group) or a collagenated synthetic bone graft lacking PDRN (control group) was performed on the sinuses.

Results: There were no appreciable differences in AH, PH, BICp, BICa total, BICa coronal, and BICa middle values amongst sinuses in the control and test groups (all P>0.05). In comparison to the sinuses in the control group (55.6%22.1%), the BICa apical of the test group's sinuses (76.7%9.3%) displayed statistically higher values (P=0.038). Statistics revealed that there were differences in pNB, pRBP, and pFVT between the two groups in AOI_A (P=0.038, P=0.028, and P=0.007, respectively). The samples from the control and test groups did not substantially differ in terms of pNB, pRBP, and pFVT in AOI_C and AOI_M (all P>0.05).

Conclusions: According to the histologic results, lateral sinus floor elevation with PDRN may promote early new bone development and increase bone-to-implant interaction.

Keywords: Sinus Floor Elevation, Bone Graft, Collagenated Synthetic Bone Graft, PDRN

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1. Introduction

Modern surgical techniques in orthopaedic and maxillo-facial surgery always request a fast bone tissue regeneration to fill gaps created by surgery or rapidly increase fixation of bones implants. Currently, the autogenous bone graft is the most used device, a kind of golden standard (Lorenzetti et al., 1998; Jensen et al., 1998), mainly for two reasons; it is a sort of structure which can be integrated into the receiving site, thus filling the existing gap, but it also provides an osteoinductive signal for the mesenchymal precursor cells, that are induced to proliferate and to differentiate into active osteoblasts, thereby accelerating the healing process (Lane et al., 1999). Thus far, bone is the only true osteoinductive material able to promote and induce new bone tissue formation from osteoblast precursors, stimulating stem cell differentiation into mature osteoblasts.

The osteoinductive properties of natural bone are due to the presence of Bone Morphogenetic Proteins (BMP) in the matrix (Harakas, 1984; Reddi, 1992; Urist, 1965; Service, 2000). This protein has been first isolated and then produced as a cloned series of molecules from the BMP protein family (Urist and Strates, 1971; Service, 2000). Above all Bmp-2 and Bmp-7 are extremely powerful mitogenic and differentiating agents (Boden, 1999). However their effect is not as long standing as wished and, in order to work properly, they have to be utilized at very high, non-physiological doses (even 6 orders of magnitude higher) whose long-term effects have not been thoroughly yet understood (Service, 2000). Growth factors, like PDGF, FGF, IGF, and TGF-h are important molecules implicated in the bone regeneration processes (Canalis et al., 1988, Joyce et al., 1990, 1991; Bourque et al., 1993; Einhorn, 1996) and are currently under examination as therapeutic agents. Their use, already largely experimented on rodents has proven to be difficult to translate to humans, and there are still many doubts about the proper doses that should be used (Bruder et al., 1999).

Maxillary molar tooth extraction has been shown to make implant implantation more challenging since it results in sinus pneumatization and alveolar bone loss [1]. Certain surgical techniques have been suggested to improve the available bone height in the posterior maxilla. Boyne and James originally suggested a lateral sinus floor elevation method in specific. [2] The lateral sinus floor elevation treatment demonstrated predictable outcomes and good survival rates in ongoing follow-up investigations [3]. In terms of endo-sinus bone gain, sinus floor elevation via the lateral method performed better than transalveolar sinus floor elevation[4]. Despite the fact that lateral sinus floor raising previously exhibited great prediction and achievement rates, the introduction of growth hormones was recommended to encourage bone development and minimise duration of therapy [5]. Bone morphogenetic protein-2 (BMP-2) and other osteoinductive growth factors have been used in sinus floor elevation techniques in numerous research [6, 7]. Nevertheless, due to adverse reactions such face swelling, BMP-2 has not been utilised frequently in therapeutic settings. Research claim that many growth factors found in platelet-rich fibrin (PRF) may enhance the regeneration of hard tissues, and PRF has been explored for its potential to speed up the production of new bone.[8,9] A recently published meta-analysis, nevertheless, revealed that there is scant evidence to argue for the use of PRF for sinus floor elevation [10]. The purpose of the research was to assess, using histomorphometric evaluation, the

effectiveness of PDRN in lateral sinus floor elevation with a synthetic bone substitute materials.

2. Methodology

There were four male adult patients who ranged in age from 28 to 38 years old and weight from 55 to 64 kg. The University gave its approval for this project. The nature of this pilot study precluded the determination of a sample size. Each individual underwent a sinus elevation treatment (lateral approach) implant placement at the location of their excised premolars in the maxilla. This was done in accordance with a split-mouth design. The test cohort received collagenated synthetic bone that was grafted with PDRN onto an elevated sinus floor, while the control group received collagenated synthetic bone that was not grafted with PDRN. The research's procedure for surgery was carried out in light of earlier research. The lateral approach to sinus floor elevation and the architecture of the sinuses. In a nutshell, intravenous injections of a mixture of tiletamine/zolazepam (0.1 mg/kg), xylazine (2.3 mg/kg), and atropine sulphate hydrate (0.05 mg/kg) were used to execute surgical operations underneath general anaesthesia. Iodine solution was used to clean the surgical site before applying 2% lidocaine to the surgical site's buccal and palatal surfaces for infiltration anaesthesia. Surgery's first stage (tooth extraction). The P2, P3, and P4 premolars of the maxilla were all extracted. The primary closure was then finished using resorbable sutures. The alveolar ridges were given two months to heal. Phase 2 of the procedure (implant implantation with sinus floor elevation). Mid-crestal incisions were made in the P2, P3, and P4 molar regions, and vertical releasing incisions were made in the distobuccal and mesiobuccal sides of P1 and M1. Elevating the full-thickness flap revealed the infraorbital tube's buccal lateral wall. With the aid of a round bur, the lateral wall was eliminated and the infraorbital tube was subsequently separated and protected. A rectangular osteotomy measuring around 126 mm was then performed to prepare the buccal lateral wall of the sinus chamber.
Utilising a simple X-ray, the osteotomy's inferior boundary was placed 1-2 millimetres above the sinus cavity's inferior border. Taking into account residual bone height (RBH; approximately 1.5 mm) and implant length (8 mm), the superior border was set to be elevated by around 6 mm vertically. The osteotomy line's distal edge was placed close to the mesial side of M1. The mesial border of the osteotomy was widened enough to safely remove the Schneiderian membrane because the P3 site was close to the septum seen in the majority of sinus cavities. A sinus curette was used to carefully separate the Schneiderian membrane in order to create room for bone augmentation. Every sinus received the same amount of collagenated synthetic bone (OSTEON 3 collagen, 6–10 mm), which was then placed there and allowed to absorb liquid-form PDRN (5.625 mg/3 mL) for ten minutes, as in previous research [25]. Then, 2 implants were placed in each enhanced sinus at the same time. Two implants were positioned in each hemimaxilla, 10 mm apart. In order to shield the grafted materials, a resorbable collagen membrane (20 mm x 30 mm; GENOSS) was lastly applied to the bony window. A 5/0 Monosyn suture was used to close the flap. The subjects were closely watched, and within the first several days after every surgical technique, antibiotics and analgesics were administered intravenously. During the healing times, there weren't no negative incidents.

Histologic processing

To achieve slices that were not calcified, the samples were dried and implanted in a

block of methyl-methacrylate glue. The bucco-palatal orientation was used to cut histological segments. A diamond cutter was used to cut each block. To create samples that were 100 m thick, portions were sawed. Then, using a diamond grinder, these specimens were ground and polished to a thickness of 30 m. All histologic pictures were captured as digital photographs and saved for histomorphometric assessment after the specimens had been stained with Goldner's trichrome.

Linear measurements

- Residual bone height (RBH, mm): distance from the most coronal to the apical point of pristine bone.
- Augmented height (AH, mm): distance from the lowest to the highest point of augmented bone.
- Protruding height (PH, mm): distance from the lowest point of the exposed fixture surrounded by the sinus membrane to the implant apex.
- Bone-to-implant contact in pristine bone (BICp, %): The percentage of pristine bone in contact with the implant surface.
- Bone-to-implant contact in augmented bone (BICa, %): The percentage of newly formed bone in contact with the implant surface. BICa was divided into 3 parts (coronal, middle and apical) depending on the implant's vertical position in the sinus.

Composition of the augmented area

Three rectangular regions of interest, designated as the most coronal region (AOI_C), middle region (AOI_M), and most apical region (AOI_A), were set within the augmented sinus zone to measure the depositing of new bone and remaining bone graft particulates. The AOI was placed 0.5mm lateral to the implant threads pitch.

- New bone area percentage (pNB, %)
- Residual bone graft particle area percentage (pRBP, %)
- Fibrovascular connective tissue area percentage (pFVT, %)

Statistical analysis

The mean, standard deviation (SD), and median values were derived to reflect the information gathered from the linear regression analysis and the make-up of the enhanced region in each AOI. The Mann-Whitney U test was used in inter-group studies to assess the variation between the control and test groups. The Kruskal-Wallis test and post-hoc (Bonferroni correction) were used for intra-group analyses. SPSS version 23 was used for all statistical calculations. For both the linear and surface analyses, the threshold for statistical significance was established at P0.05.

3. Results

Table 1. The linear measurements of the histomorphometric analysis

Groups	RBH (mm)	AH (mm)		PH (mm)		BIC_p (%)		BIC_a total (%)		BIC_a coronal (%)		BIC_a middle (%)		BIC_a apical (%)	
	Mean±SD	Mean±SD	Median	Mean± SD	Median	Mean±SD	Median	Mean± SD	Median	Mean±SD	Median	Mean±SD	Median	Mean± SD	Median
Control gro	1.3 ±0.3	6.5± 1.5	6.8	0.4 ±0.6	0.1	84.1 ±9.0	83.2	70.7 ±8.5	73.4	89.7±8.0	90.9	70.4 ±11.4	71.2	55.6 ±22.1	64.2

up (n=8)														
PDRN group (n=8)	1.6±0.5	7.1±1.1	6.9	0.1±0.2	0	80.7±8.4	82.3	78.3±11.1	79.6	85.4±10.7	86.4	73.3±22.9	70.6	76.7±9.3	77.4
P value	0.236	0.574		0.279		0.534		0.161		0.505		1.000		0.038 a)	

The Mann–Whitney U test was performed. A statistically significant difference was found in BIC_a apical values between the samples in the PDRN-treated and control groups. RBH: residual bone height, AH: augmented height, PH: protruding height, BIC_p: bone implant contact on pristine bone, BIC_a: bone implant contact on augmented area, SD: standard deviation, PDRN: polydeoxyribonucleotide

Table 2. The surface histomorphometric measurements of each AOI in the augmented area

Location of the AOI	**Control group (n=8)**		**PDRN group (n=8)**		***P* value**
	Mean±SD (%)	**Median**	**Mean±SD (%)**	**Median**	
Coronal (AOI_C)					
pNB	49.9±16.7	51.4	40.5±16.5	42.3	0.328
pRBP	13.3±12.6	13.4	21.4±7.9	21.7	0.161
pFVT	36.8±15.9	40.1	38.2±16.9	31.3	0.798
Middle (AOI_M)					
pNB	27.4±15.0 b)	25.1	25.4±16.4	28.1	0.721
pRBP	23.6±13.5	25.3	23.0±6.8	25.0	0.721
pFVT	49.1±19.2	50.1	51.6±18.1	50.8	0.878
Apical (AOI_A)					
pNB	12.1±10.0 b)	12.9	26.3±12.3	29.9	0.038 a)
pRBP	14.8±9.9	18.6	24.5±4.3	17.5	0.028 a)
pFVT	73.1±17.1 b,c)	68.6	49.2±11.1	50.3	0.007 a)

The surface areas of all selected regions (coronal, middle and apical) were 1 mm^3. AOI: area of interest, pNB: percentage of new bone, pRBP: percentage of residual bone graft particle, pFVT: percentage of fibro vascular connective tissue. a)Statistically significant difference between the control and test groups (P<0.05). b)Statistically significant difference from AOI_C within the control group (P<0.05). c)Statistically significant difference from AOI_M within the control group (P<0.05)

4. Discussion

The purpose of the research was to determine how PDRN affected lateral-window sinus floor elevation's early bone development. At two months, samples from the PDRN group had significantly higher new bone development nearby the Schneiderian membrane area than samples from the control group (P=0.038) (Table 2). The BIC value in the increased region of the adjacent Schneiderian membrane was also considerably greater in the PDRN group's samples than in the other groups'

samples (P=0.038) (Table 1). These findings verified the osteoinductive ability of PDRN in an unfavourable region for the creation of new bone—a region that was adjacent to the Schneiderian membrane and far from pristine bone. Osteoprogenitor cells typically come from healthy bone, particularly the sinus floor or lateral window [11]. In simple terms, as the distance from the measured pure bone grows, fresh bone growth diminishes. As this is going on, some studies have hypothesised that the Schneiderian membrane might encourage the production of new bone by supplying more osteogenic cells [12,13]. However, numerous studies have shown that, in comparison to the bone wall, its influence on the production of new bones is incidental [14,15]. Our investigation found a reduction in new bone production in the apical direction, which is consistent with earlier research, indicating that the Schneiderian membrane may not contribute to sinus floor elevation by acting as an osteoinductive factor.

No discernible variations in pNB, pRBP, or pFVT were discovered between samples from the test and control groups in the coronal and middle regions. However, there were statistically significant variations in the pNB, pRBP, and pFVT values across samples in the 2 groups in the apical region next to the Schneiderian membrane. This pattern resembles the findings of an earlier investigation that used BMP-2 to detect new bone growth after two weeks [16]. The distance from the bone wall, which has the capacity to promote osteogenesis, may be responsible for these outcomes. The bone wall, which has a high osteogenic potential, appears to be disguised by BMP-2 or PDRN's osteoinductive impact; however, the effect extends below the Schneiderian membrane, in an area with a low osteogenic potential. Adenosine A2a receptor-mediated angiogenesis, which might aid in bone repair, is known to be controlled by PDRN. In a prior study, the PDRN therapy group demonstrated increased angiogenesis, which caused human bone marrow mesenchymal stem cells to migrate [17]. Adenosine A2a receptor agonists may also hasten the production of new bone. In a study subjects with a calvaria lesion, an adenosine A2a receptor agonist demonstrated greater bone repair. By enhancing osteogenic potential in the middle and apical sections in comparison to the control group, rapid angiogenesis may enhance new bone regeneration [18]. In this sense, PDRN might be used as a technique to encourage bone regeneration in patients or in settings with complicated osteogenic potential. The most successful biomolecule for promoting bone repair is thought to be BMP-2 [20]. A sinus floor raising model has been used to study the administration of BMP-2, and the osteoinductive effects were confirmed [16,19]. Clinical applications of BMP-2 have not yet been widely adopted, despite evidence of their usefulness.

When BMP-2 was used at high doses, this may have resulted in side effects such inflammatory responses, seroma, or oedema [21,22]. The Schneiderian membrane, lateral window, and enhanced area were all characterized and scrutinized in histologic findings in this investigation to check the safety of PDRN. Both groups' animals recovered without experiencing any significant inflammation. First, both sets of subjects displayed a typical Schneiderian membrane made up of periosteum, lamina propria, and epithelial lining. Although bone graft particles were integrated into the lamina propria, neither samples from the test group nor those from the control group showed any signs of inflammation around the fragments. In the entire Schneiderian membrane, the mean thickness of the epithelium and lamina propria were 45 and 354 m, respectively, according to a prior study

[21]. The epithelium and lamina propria thicknesses in this investigation were measured to be 30–40 m and 0.7–1.3 mm, respectively. Following raising the sinus floor for two months, the epithelium thicknesses was standard. The lamina propria was found to be somewhat thicker than usual, nevertheless. It is assumed that during the recuperation period following surgery, the amount of blood vessels and mucous glands may have grown, leading to thickening of the lamina propria layer. Future research should assess the Schneiderian membrane's histological and structural alterations, particularly those in the lamina propria layer, 4–6 months following sinus floor elevation.

Secondly, a resorbable collagen membrane was typically placed across the area of the lateral window. Despite bone bridging, incomplete corticalization was evident since both groups saw bony wall renewal around the lateral window. This may have been impacted by the lateral window's size [23] and a lack of healing time during the preliminary assessment of PDRN's osteogenic potential. Third, neither group saw any particular negative consequences, but both groups did have new bone production and bone remodelling in the augmented area. In this work, an osteoblast rim was seen surrounding osteoids that were resting on newly formed bone.

5. Conclusion

In conclusion, it was shown that the adjacent Schneiderian membrane (apical area) of PDRN has an osteoinductive impact for sinus floor elevation. To encourage bone regeneration, PDRN could be utilized in regions with low osteogenic potential. However, because this was a pilot study, a small sample size was used in this investigation. The outcomes should also be evaluated cautiously because of confounding variables like different sinus cavity diameters and shapes. To determine the ideal clinical application of PDRN, additional study must be conducted using a larger sample size and a range of PDRN dosages.

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