An evaluation of the effectiveness of connective tissue grafts as a barrier with bioresorbable collagen membrane in the treatment of mandibular Class II furcation defects in humans: a case report series of 20 cases

KEY WORDS Class II furcation, collagen membrane, subepithelial connective tissue graft

Background: Class II furcation defects indicate a specific common clinical problem and the need for effective therapy. Connective tissue grafts have been used as a barrier in a limited number of previous reports for protecting and stabilising wounds of furcation sites during the healing period, so as to support bone regeneration when closing mandibular Class II furcation defects. The present case report series was carried out to compare the effectiveness of connective tissue grafts (CTGs) as a barrier with bioresorbable collagen membrane in the treatment of mandibular Class II furcation defects in humans.

Study design: Twenty chronic periodontitis patients with a single Class II furcation defect on a buccal or lingual surface of mandibular teeth were included in the study. Vertical probing pocket depth (V-PPD), vertical relative attachment level (V-RAL) and relative gingival margin level (RGML) were recorded at baseline and 6 months post-surgery.

Results: After 6 months, the mean V-PPD reductions observed in the test group (2.43±0.46mm) and control group (2.21±0.61mm) were statistically significant. However, the difference was not statistically significant between the test group and the control group. In both the test and the control group, the mean RAL gains at 6 months were statistically significant, compared with the baseline data. At 6 months, the mean V-RAL gains in the test group (2.08±0.67) were greater than the control group (1.73±1.15), but the difference was not statistically significant (P=0.06). Complete closure of furcation was achieved at one site in the test group compared with no sites in the control group.

Conclusion: It can be concluded that 6 months after surgery, both connective tissue graft as a barrier, and resorbable collagen membrane resulted in improvement in terms of V-RAL gains and reduction in V-PPD and horizontal probing depth.

Introduction

The ideal aim of furcation therapy is to retain the tooth and completely close the furcation area, thereby regenerating normal local conditions and improving the likelihood of tooth retention\(^1,2\). Class II furcation defects indicate a specific common clinical problem and a need for effective therapy\(^3,4\). Several techniques have been proposed and promoted to treat and improve the clinical condition of mandibular Class II furcation involved molars, and varying results have been found\(^2,5,6\). Excluding isolated reports of favourable furcation fill with autografts and demineralised freeze-dried bone allo-
grafts, bone replacement grafts have had limited success in managing Class II furcation defects. Problems associated with bone replacement grafts have included graft containment, epithelial exclusion, microbial contamination and variable inductivity of the graft. Extensive comparative studies using bioresorbable and non-absorbable barriers for guided tissue regeneration (GTR) furcation therapy have been performed. Although some reports have demonstrated significant clinical attachment gain, other studies have shown that no significant clinical improvement occurs. Regardless of the barrier membrane used, the results obtained from most of the GTR procedures clearly show the limitations of this therapy in this type of defect.

It has been suggested that combination treatment, including both a bone replacement graft and GTR barrier would provide the most beneficial regeneration therapy for Class II furcations. The advantage of using a bone graft with a membrane is that an initial blood clot stabilisation may be established and the potential for having dead space under the membrane is decreased. However, the results obtained in controlled clinical studies have demonstrated that the use of bone replacement grafts together with barrier membranes is of limited significance in providing additional benefits compared with the use of the membrane alone. Therefore, the use of replacement grafts to improve the results of guided tissue regenerative therapy is not clearly justified.

Recently, connective tissue grafts (CTGs) have been reportedly used as a barrier for protecting and stabilising wounds at furcation sites during the healing period and to support bone regeneration for closing mandibular Class II furcation defects in humans. Various histological studies of healing of subepithelial connective tissue grafts to the root surface revealed areas of regeneration, with new bone, cementum and connective tissue attachment coronal to the original gingival margin. Bellal et al. reported more beneficial effects with the connective tissue graft as a barrier in the treatment of mandibular Class II furcation defects compared with GTR therapy. Therefore, the present case report series was undertaken to compare the effectiveness of CTG as a barrier with bioresorbable collagen membrane in the treatment of mandibular Class II furcation defects in humans.

### Study design

Twenty chronic periodontitis patients (11 males and 9 females; mean age 35.90±7.32 years) with a single Class II furcation defect on the buccal or lingual surface of mandibular teeth were included in the study. All of the patients were free of systemic diseases and were not receiving any medication. Patients fulfilling the following criteria were included in the study:

- the presence of at least one Class II furcation defect affecting buccal/lingual surfaces of mandibular molars as determined by clinical and radiographic evaluation
- the presence of ≥3mm horizontal furcation probing depth, but not entirely through furcation
- a normal response to the electric pulp test
- the gingival margin positioned coronal to the furcation fornix.

Patients with the following criteria were excluded from the study:

- patients non-compliant with the periodontal maintenance program
- patients who were smokers or used any other tobacco products
- patients exhibiting tooth mobility in the treatment area.

If the patients met the above selection criteria, possible types of treatments and their associated risks and benefits were explained to the patients and an informed consent form was issued, which the patients signed and agreed to the treatment. The study was approved by the ethical committee of Jawaharlal Nehru Medical College, Sawangi (Meghe), Wardha.

### Initial therapy

After proper examination and diagnosis, initial therapy consisted of oral hygiene instructions, full mouth supra- and subgingival scaling and root planing under local anaesthesia. If necessary, occlusal adjustment was performed. Plaque control instructions were repeated until patients achieved 80 to 85% plaque control. A re-evaluation exami-
nation was performed 6 weeks following completion of initial therapy to determine the patient's response to the therapy and to confirm the need for periodontal surgery.

Clinical measurements

On the day of the surgical procedure, a full-mouth plaque score was assessed by the plaque index (PI) (Turesky–Gilmor–Glickman modification of Quigley–Hein)\(^{18}\). Gingival inflammation was assessed by papillary bleeding index (PBI)\(^{19}\). The probing measurements recorded for assessment of results were vertical probing pocket depth (V-PPD), vertical relative attachment level (V-RAL) and relative gingival margin level (RGML). The measurements were recorded at three sites of each furcation surface for each tooth: the mesial line angle, distal line angle and midbuccal/midlingual. For later calculations, the mean of the three sites were taken into consideration. These measurements were recorded with a computerised constant force probe (Florida Disk Probe, Florida Probe Corporation, Gainesville, FL, USA) with a constant probing force of 15gm (a pressure of 154N/cm\(^2\)), tip diameter of 0.40mm, precision of 0.1mm and a probe length of 20mm. Horizontal probing depth (HPD) was recorded using a colour-coded curved furcation probe with 0 to 3, 3 to 6 and 6 to 9mm markings (Nabers probe PQ2N, Hu-Friedy, IL, USA). The operator recorded all measurements at baseline and 6 months postsurgically. Complete closure of the defect was assessed at the end of the 6-month period using the bone sounding procedure.

Radiographic measurements

Intraoral periapical radiographs using the long cone paralleling technique were taken at baseline and at 6 months (Figs 1 to 4).

The surgical procedure

Prior to the surgical procedure, the patient was instructed to rinse with 0.2% chlorhexidine gluconate (Hexidine\(^{\text{R}}\), ICPA Health Products Ltd, Mumbai, India) for 1 minute after induction of local anaesthesia (2% lidocaine, adrenaline 1:100,000). Intraseptal incisions were given using Bard-Parker number 12 and 15 surgical blades (Wincut, Goldwin Medicare Ltd, Mumbai, India) on the buccal and lingual aspects, with the blade directed towards the crest of the alveolar bone followed by vertical releasing incisions, if necessary, extending into the alveolar mucosa. This had the aim of achieving proper access to the defect, as well as to obtain coronal displacement of the flap. A tooth mesial and a tooth distal to the experimental site were included in the flap. The full-thickness (mucoperiosteal) flap was reflected facially and linguually using a periosteal elevator (24G, Hu-Friedy) to expose the alveolar bone margin (Fig 5). The subsurface of the flaps were carefully curetted to remove any possible pocket epithelium. The granulation tissue in the furcation defect was removed and the exposed root surface, including the roof of the furcation, was scaled and planed using hand instruments (Gracey curettes, Hu-Friedy) and ultrasonic instruments (Piezon\(^{\text{R}}\) Master 400, Electro Medical Systems, Nyon, Switzerland). After haemostasis and irrigation with physiological saline solution, intrasurgical measurements were recorded. The osseous defects were measured at their deepest point both vertically and horizontally after flap elevation and debridement. Horizontal bone depth of the furcation defect at its deepest location was measured using a William’s graduated probe and an additional reference probe placed across the most prominent root surface to bridge the first probe. Vertical bone depth of the furcation defect at its deepest location was measured using the fornix of the furcation as the fixed reference point. After assessment of the intrasurgical parameters, the sites were selected for treatment with either GTR or CTG by tossing a coin.

Surgical procedure for the test group

The surgical procedure for the test group is shown in Figures 5 to 7. In cases of CTG, donor palatal tissue was harvested by a ‘trap door approach’ from the area between the maxillary first molar and maxillary cuspid. A template was placed at the recipient site to harvest a graft of the correct dimensions. An incision was made in the palate that was parallel to the maxillary premolar and first molar at a distance of approximately 3mm apical to the gingival margin.
Perpendicular incisions were made to establish the width of the graft for covering the entire area of furcation. A flap was reflected to expose the underlying connective tissue. The connective tissue graft was removed with a periosteal elevator. The graft was then placed on a sterile gauze pad and irrigated with saline. It was modified as per the required dimensions after removal of excess fatty and glandular tissue. The primary flap was then returned to its original position and sutured with 4-0/5-0 Mersilk (Ethicon, Johnson & Johnson, Somerville, NJ, USA) sutures to obtain primary closure. This was done using the interrupted suturing technique or crossed horizontal suspension suturing technique. A template was prepared to completely cover the exposed furcation, so that it covered at least 3 mm of alveolar bone apical to the furca and extended coronally up to the cementoenamel junction (CEJ) (Fig 6). The harvested connective tissue graft was trimmed accordingly. The connective tissue graft was then secured to cover the furcation defect by facing the periosteal surface of the graft to the furca (Fig 7). Finally the flap was repositioned to completely cover the connective tissue graft and sutured with 4-0/5-0 Mersilk sutures to obtain closure, using vertical mattress or the interrupted suturing technique.

### Surgical procedure for the control group

The control sites were treated according to the principles of guided tissue regeneration, with the application of Bio-Gide®, resorbable bilayer membrane (Geistlich Pharma, Wolhusen, Switzerland). A template was prepared to completely cover the defect, extending from at least 3 mm apical to the adjacent alveolar bone crest to a level approaching the CEJ. The resorbable collagen membrane was trimmed accordingly. The membrane was secured over the defect and sutured with a sling suture using 5-0 vicryl resorbable sutures. Finally, the flap was coronally repositioned to completely cover the GTR membrane and sutured with 4-0/5-0 Mersilk sutures to obtain closure, using vertical mattress or the interrupted suturing technique. Immediately after surgery, patients were prescribed a non-steroidal anti-inflammatory (Ibugesic Plus, which is a combination of ibuprofen + paracetamol) t.d.s for 5 days and systemic antibiotics (amoxycillin, 500mg, t.d.s for 5 days). Patients were instructed not to brush the teeth in the treated area. All patients were placed on 0.2% chlorhexidine glucuronate (Hexidine, ICPA) twice daily, for 1 minute, for 4 to 6 weeks. Patients were instructed not to disturb the pack and to avoid undue trauma to the treated site. The patients were recalled at 1 month, 3 months and 6 months following surgical treatment. Clinical measurements recorded preoperatively were repeated at 3 and 6 months post-operatively.

### Statistical analysis

The means and standard deviations (mean±SD) values were calculated for all clinical parameters. Student paired t test was used to compare data from baseline with those at 6 months for each treatment group. Comparisons between treatment groups at baseline and 6 months were carried out using Student unpaired t test. During the 6-month study period, the wound healing occurred without any problems. None of the cases showed either exposure of membrane or necrosis of CTG. None of the selected patients dropped out before the termination of the study. All of the patients were satisfied with the results. The mean PI and PBI scores at baseline and 6 months for both test and control group remained low (<1).

### Baseline characteristics for test and control groups

At baseline, mean V-PPD was 4.07±0.86mm in the test group and 3.74±0.52mm in the control group (Tables 1 and 2). Similarly, mean V-RAL was 12.68±1.27mm in the test group and 12.97±1.16mm in the control group. Mean RGML was 8.56±1.28mm in the test group and 9.22±1.10mm in the control group. Horizontal probing depth was >3mm in all of the patients in the test and control group at baseline. Intrasurgical hard tissue measurements recorded for mean open horizontal defect depths were 4.20±0.73mm for the test group and 4.20±0.94mm for the control group. Mean open vertical defect depth
Fig 1  Preoperative intraoral periapical radiographs (test group).

Fig 2  Post-operative intraoral periapical radiographs (test group).

Fig 3  Preoperative intraoral periapical radiographs (control group).

Fig 4  Post-operative intraoral periapical radiographs (control group).

Fig 5  Reflection of the flap (test group).

Fig 6  Measurement with tin foil at the recipient site (test group).

Fig 7  Graft placed at recipient site (test group).

Fig 8  Placement of periodontal pack (control group).
was 2.80±0.56mm for the test group and 2.60±0.73mm for the control group. At baseline no statistically significant differences were observed between the test and control groups.

### Clinical outcomes at 6 months

#### Vertical probing pocket depth

In the test group, the mean probing pocket depth decreased from 4.07±0.86mm at baseline to 1.64±0.61mm at 6 months. The mean V-PPD reductions observed at 6 months (2.43±0.46mm) were statistically significant (P=0.000). In the control group, the mean PPD was decreased from 3.74±0.52mm at baseline to 1.53±0.52mm at 6 months.

There were statistically significant reductions in the mean vertical probing pocket depth at 6 months (P=0.000) compared with the baseline. At 6 months, the mean PPD reduction in the test group (2.43±0.46mm) when compared with the control group (2.21±0.61mm), did not show a significant difference (P=0.36).
**Vertical relative attachment level**

In the test group, the mean V-RAL decreased from 12.68±1.27 mm at baseline to 10.60±1.43 mm at 6 months. Student paired t test indicated that the mean RAL gain at 6 months was statistically significant ($P=0.000$), compared with the baseline data. In the control group, the mean vertical relative attachment level decreased from 12.97±1.16 mm at baseline to 11.24±0.84 mm at 6 months. The mean V-RAL gain observed at 6 months was significantly greater ($P=0.000$), compared to the baseline data. At 6 months, the mean V-RAL gain in the test group (2.08±0.67) was greater than the control group (1.73±1.15) but the difference was not statistically significant ($P=0.06$).

**Relative gingival marginal level**

In the test group, the mean relative gingival marginal level increased from 8.56±1.28 mm at baseline to 8.98±1.30 mm at 6 months, with a mean gingival recession of 0.41±0.61 mm. However, Student paired t test results indicated that the mean gingival recession observed at 6 months was statistically non-significant ($P=0.060$) compared with the baseline data. In the control group, the mean relative gingival marginal level increased from 9.22±1.10 mm at baseline to 10.02±1.08 mm at 6 months with a mean gingival recession of 0.79±0.41 mm. The Student paired t test indicated that the mean gingival recession observed at 6 months was statistically significant ($P=0.000$) compared with the baseline data.

At 6 months, the mean gingival recession in the test group was 0.41±0.61 mm and when compared with the control group (0.79±0.41), the difference of 0.37±0.23 was not statistically significant ($P=0.12$).

**The frequency distribution**

Complete closure of the furcation was achieved at one site in the test group, compared with no sites in the control group. The improvement in horizontal classification from Class II to Class I was seen at three sites in the test group and at three sites in the control group. However, one site in the test group and two sites in the control group remained unchanged.

**Discussion**

At baseline, none of the investigated parameters in both of the experimental groups showed any statistical difference, thus, ensuring the same starting point for both of the procedures tested. Bioresorbable bilayer collagen membrane (Bio-Gide®) was used as an active control in the present study. The placement of a GTR barrier membrane represents the recommended and most well-documented regenerative treatment modality for mandibular Class II furcation defects. In particular, the type of bioresorbable collagen membrane used in the present study had previously shown promising results. All patients participating in the study showed good oral hygiene levels and a healthy gingival condition throughout the study period, as indicated by the PI and PBI scores.

The primary efficacy parameter for validation of clinical regeneration after GTR procedure is the gain in clinical attachment level (CAL). For outcomes relating to clinical regeneration of furcation treated sites, it is becoming increasingly common to measure attachment gain in both the vertical and horizontal directions. This is because a primary goal of regenerative furcation therapy is to reduce the magnitude of the furcation defect to a size that can be maintained by routine hygiene methods and mechanical instrumentation. In the present study, significant gains in the mean vertical RAL were observed in both of the groups at 6 months; 2.08±0.67 mm in the CTG group, and 1.73±1.15 mm in the GTR group. When the difference in V-RAL gain was compared, a greater but not statistically significant amount of RAL gain was observed in the CTG group. In a recent publication, Becker et al reported the results of bioresorbable membrane therapy for the treatment of Class II furcations. The results showed that the 31 Class II furcations that were treated resulted in 2.1 mm of gain in clinical attachment levels. The mean gain in RAL observed in the present study is in agreement with those results published by Becker et al. Furthermore, the present findings agree with previous reports in the literature that indicate...
favourable results with bioresorbable membrane as a barrier when used in the treatment of Class II furcations. In terms of RAL gain, the results of the present study are comparable to those reported for other studies using CTG as a barrier for the treatment of mandibular molar Class II furcation defects. Bouchard et al. compared connective tissue grafts as a barrier with expanded polytetrafluoroethylene (ePTFE) membranes for closing 24 mandibular buccal Class II furcation lesions, and reported comparable potential for both types of graft in supporting bone regeneration in mandibular Class II furcation lesions. Recently, Belal et al. reported the results of a study that evaluated the response of connective tissue grafts as a barrier therapy for 10 Class II mandibular furcation defects, and 2.83 mm of V-CAL gain was observed. A gain in CAL using CTG as a barrier (as observed in the present study) also supports the favourable clinical and histological results of Bruno and Bowers, who used connective tissue grafts in their case report study. The good results for CAL improvement obtained by using a connective tissue graft could be explained by the fact that in addition to providing a space for regenerating cells, the connective tissue graft may have better supported and protected the wound of the furcation site during the healing period (similar to or perhaps better than the GTR barrier membrane).

A primary goal of periodontal therapy is to reduce PPD so as to limit the risk of local re-infection. Shallow pockets have a strong, negative predictive value for future disease progression, whereas deep pockets in treated areas are a risk indicator for periodontal disease progression. In the present study, statistically significant reductions in mean vertical probing pocket depth were observed in both the test (2.43 ± 0.46 mm) and control (2.21 ± 0.61 mm) groups during the study period, with an initial pocket range of 3 to 6 mm. The CTG group showed a greater mean V-PPD reduction compared with the GTR group during the 6-month study period. However, the difference was not found to be significant. The mean V-PPD reductions observed in the present study for both groups are comparable to the results reported by Bouchard et al. and Belal et al. Bouchard et al. compared periodontal regeneration in mandibular buccal Class II furcation defects between an ePTFE membrane and a connective tissue graft and observed a mean V-PPD reduction of 1.9 mm in the CTG group and 2.2 mm in the GTR membrane group. Belal et al. also observed mean V-PPD reduction of 2.43 mm with GTR membrane and 2.5 mm with the CTG group, when comparison was made between the two groups for the treatment of Class II furcation defects. However, Lekovic et al. reported a higher PPD reduction of 4.14 mm (6.47 mm to 2.33 mm) with CTG when it was used as a barrier for the treatment of mandibular Class II furcation defects. The higher PPD reduction reported by Lekovic et al. with the CTG group could be because of the inclusion of periodontal pockets of variable depth, which may have influenced the treatment output.

The results of the present study showed comparable clinical improvement between the two procedures. However, the number of Class II furcation defects that closed or were converted to Class I was higher in the connective tissue graft group; 20% (1) of the initial Class II furcations became class 0 and 60% (3) became Class I, while in the connective tissue graft treated group, 20% (1) remained as Class II with a horizontal probing depth of 3 mm or above. The bioresorbable collagen membrane (GTR) treated group showed 60% (3) of initial Class II furcation converted to Class I, whereas 40% (2) of the defects remained unchanged. Bouchard et al. reported bone closure of Class II furcation defects for 2 of 11 of a connective tissue graft treated group and 4 of 11 for an acid plus ePTFE membrane-treated group. It has been reported that the complete resolution of the furcation involvement using GTR is not highly predictable. Moreover, in some studies, none of the membrane-treated mandibular Class II furcations were closed. Although the evaluation period and the number of patients for the present clinical trial do not allow for a conclusion that favours one procedure over the other, within the limits of this 6-month report, it may be assumed that a connective tissue graft has a better clinical effect on the soft tissue closure of furcation defects compared with the use of a GTR membrane. The present study showed an increase in the depth of gingival recession from baseline to 6 months post-operatively in both of the groups. An increase in the amount of gingival recession has previously been reported to occur following surgical periodontal procedures and this
was considered, as it was an expected side effect of the GTR procedure\textsuperscript{23,33}.

In the present study, higher gingival recession was observed in the GTR group compared with the CTG group, but the difference was not statistically significant. This confirms previous findings by Blumethal et al\textsuperscript{34}, who reported similar changes in gingival recession for collagen membrane in the treatment of mandibular Class II furcation defects. Belal et al\textsuperscript{16} also reported an insignificant change in gingival recession at both 6 and 12 months post-operatively for connective tissue graft-treated mandibular Class II furcation defects. Re-entry procedures involving a second surgical entry of the operated sites provide an accurate assessment of the hard tissue fill at the regenerated sites. However, these procedures do not provide sufficient information on the interface between the bone and the root surface, and do not allow assessment as to whether or not new cementum and a new periodontal ligament are also present\textsuperscript{25}. Histological evaluation remains the only reliable method for determining all the components of attachment apparatus. Thus, the absence of histological evidence in the present study precludes the presumption that true periodontal regeneration occurred. However, in a study by Parodi et al\textsuperscript{36}, in which a maxillary molar was treated with a similar membrane used in the present study, after 5 months of healing, regeneration of cementum and bone as well as a functionally oriented periodontal ligament at the site of the previously diseased root surface was demonstrated. Histological studies\textsuperscript{27,28} on connective tissue grafts have shown that the attachment of the graft to the root surface appears to be mediated by a combination of epithelial down growth, and connective tissue attachment with minimal sign of new cementum-like tissue formation in the apical portion of the recession areas coronal to the base of the instrumented root surface. Coronal growth of alveolar bone has been observed to a minimum extent and does not parallel the height of newly formed cementum-like tissue.

\textbf{Conclusions}

From the analysis of the results, and within the limitations of the present study, it can be concluded that, 6 months after surgery, both connective tissue graft as a barrier and resorbable collagen membrane resulted in improvement in terms of V-RAL gains and V-PD and HPD reductions.

The study also had a few limitations. The small sample size limited the statistical analysis of the results. In addition, the ethical considerations as well as associated patient non-acceptance have restricted the re-entry surgery to assess actual bone fill. Long-term analysis is also required to determine the stability of the results. Further, well-controlled studies are needed to confirm the findings of the present study.

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