



## Human Chorion Membrane Allograft Versus Subepithelial Connective Tissue Graft In The Treatment of Miller's Class I and Class II Gingival Recession

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### Abstract

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### Introduction

Gingival recession is one of the most common features of periodontitis and of particular importance as it compromises aesthetic appearance of the patient and may also predispose the patient to pathologies such as root caries and abrasion.<sup>1</sup>

Various surgical modalities have been developed for the manipulation of gingival tissue to cover recession defects. Modified coronally advanced flap is one such technique which involves coronal advancement of a partial thickness flap raised around the recession defect site. However autogenous grafts derived from the palate (Subepithelial connective tissue graft, Free gingival grafts) with modified coronally advanced flap technique have shown good results.<sup>2,3</sup>

Traditionally, connective tissue graft has been considered as gold standard for root coverage procedures but the major drawback is the creation of a second surgical site. Harvesting of graft is a technique sensitive procedure which causes patient discomfort and increases the chance of post-operative complications. To overcome such drawbacks, research has been directed to find biocompatible materials to achieve similar results without the inherent drawbacks of an autograft. Of late, placenta derived membranes have been used for root coverage procedures, based on the biologic properties in placenta which enhances wound healing and may even propagate regeneration.<sup>4,5</sup>

Since there are less studies in this aspect, a study was conducted to compare the efficacy of subepithelial connective tissue graft with human chorion membrane using modified coronally advanced flap technique for the treatment of miller's class I and class II gingival recession defects.

### Materials and Methods

This was a split mouth comparative study conducted on patients who reported to the Department of Periodontics, V.S. Dental College and Hospital, Bengaluru, Karnataka, India during the period from September 2016 to August 2017. The patients who fulfilled the inclusion criteria were selected for this study, after which ethical clearance was obtained from the institution. Participation of the subjects was voluntary and a written informed consent was obtained before treatment.

A total of 20 sites with gingival recession were selected and the sites were divided into 2 groups –. The test group consisted of 10 sites, in which the sites were treated with human chorion membrane and the Control group consisted of 10 sites, in which the sites were treated with subepithelial connective tissue graft. In both the groups, the flaps were closed by modified coronally advanced technique.

### Selection Of Cases

### Inclusion Criteria

1. Systemically healthy subjects with the age range of 25-60 years
2. Patients with Millers class I or class II recession defects
3. Subjects who are compliant
4. Radiographic evidence of sufficient interdental bone.

#### Exclusion Criteria

1. Subjects showing compromised oral hygiene during/after phase I therapy
2. Medically compromised subjects
3. Crowding/Mal-aligned teeth
4. Smokers

#### Method Of Evaluation

After case selection, all patients were subjected to Phase 1 therapy, which included full- mouth scaling and root planing. The patients were monitored, and after achieving a satisfactory level of plaque control, the baseline data were collected. The parameters evaluated were the recession height (RH), probing depth (PD), width of the attached gingiva (WAG) and the gingival biotype. The gingival biotype was evaluated based on the transparency of the probe. Clinical photographs and IOPA radiographs were taken at various surgical steps and during follow-up visit at 6 months. UNC-15 probe was used to take clinical measurements and a custom-made acrylic stent to guide the placement of probe at subsequent measurements.<sup>5</sup>

#### Source Of Allograft

The Human chorion membrane was obtained from the tissue bank, TATA memorial hospital, Mumbai.

#### Surgical Procedure

Preparation of the recipient site - After administration of local anaesthesia, a modified coronally advanced flap was raised by making submarginal incisions in the interdental areas, intra-sulcular incisions around the teeth, and two vertical incisions at the end of the two horizontal incisions extending into the alveolar mucosa. The resulting trapezoid-shaped flap was elevated with a split–full–split approach in the coronal–apical direction. The papillae were de-epithelialized to create a connective tissue (CT) bed.

The exposed root surfaces were thoroughly planed with currettes.<sup>2</sup>

In the test group, a chorion membrane of the desired defect dimension was placed over the defect. The membrane was found to adhere to the root surface, and no attempt was made to suture the membrane independently. After this the flap was repositioned 1 mm coronal to the cemento-enamel junction (CEJ) covering the tip of the deepithelialized papillae and sutured<sup>5</sup> (Fig1 – Fig3).

In the control group, after flap elevation, a template was fabricated using tin foil. An outline of the required dimension was marked on the palate. Subepithelial connective tissue graft was harvested according to langer and langer technique, positioned at the recipient site and sutured<sup>3</sup> (Fig5 – Fig7).

Suturing of the recipient site - Monofilament Polypropylene 4-0 suture was used for suturing the recipient sites with the coronal portion of the flap stabilized using a sling suture and multiple direct loop sutures were placed to close vertical incisions.

Suturing of the donor site - Multifilament 5-0 silk suture was used to suture the donor site. Donor site wound was stabilized using multiple direct loops and cross mattress sutures.

After the procedure, the patients were given postoperative instructions and medications (Augmentin 625mg, twice daily for 5 days, and Ibuprofen 400 mg, thrice daily for 3 days) and were instructed to rinse with chlorhexidine (0.2%) twice daily for 1 minute.

#### Follow Up

Subjects were recalled after 1 week for suture removal and then at 6 months post operatively. Oral prophylaxis of the subjects was performed at each visit. PD, RH, WAG and gingival biotype were assessed at each visit using an acrylic stent and UNC-15 probe along with clinical photographs.

#### Statistical Analysis

Statistical analysis was done using Statistical Package for Social Sciences [SPSS] for Windows Version 22.0 Released 2013. Armonk, NY: IBM Corp. Descriptive analysis was done using mean and standard deviation for quantitative variables,

frequency and proportions for categorical variables. Mann Whitney U test was used to compare the mean values of study variables (in mm) between two groups at different time intervals. Similarly, the percentage change between groups was compared using the same test. Gingival Biotypes between the two groups during time intervals were compared using Chi Square test. Mc Nemar's test was used to compare the Gingival Biotypes between Pre and post treatment periods in each study group. The mean values of different variables (in mm) between Pre & Post treatment period in each study group were compared using Wilcoxon Signed ranked test. The level of significance was set at  $P < 0.05$ .

## Results

All the patients tolerated the surgical procedure well and there was no post-operative complication. The mean age of the subjects in the test group was  $34.2 \pm 4.1$  years of which 33.3% were males and 66.7% were females. In the control group, the mean age was  $41.3 \pm 9.4$  years of which 66.7% were males and 33.3% were females.

At baseline, no statistically significant difference was found between the two groups when the PD, RH, WAG and gingival biotype were evaluated (Table 1).

**INTRA GROUP COMPARISON** - In the test group, there was a statistically significant decrease in the PD and RH, 6 months after intervention. The mean change in PD was from  $1.75 \pm 1.01$  mm to  $1.10 \pm 0.21$  mm ( $P$  value - 0.04) and the mean change in RH was from  $2.90 \pm 1.20$  mm to  $2.00 \pm 1.45$  mm ( $P$  value - 0.02) post operatively after 6 months. The mean difference between the pre- and post-operative PD and RH was 0.65 mm and 0.90 mm respectively. But the results also showed that there was no statistically significant change in the WAG, the mean change being  $2.35 \pm 0.91$  mm to  $2.50 \pm 0.71$  mm (Table 2, Figure 1 & 4).

Similarly, comparison of PD, RH and WAG (in mm) between pre and post treatment period was done in the control group also. The results showed that there was statistically significant reduction in RH, 6 months after intervention. The mean reduction in RH was from  $3.30 \pm 1.06$  mm to  $1.60 \pm 1.60$  mm ( $P$  value - 0.01) and the mean difference between the pre- and post-operative RH was 1.70 mm. When the pre and post treatment PD and WAG were compared, the

results showed no statistically significant difference. The change in the PD was from  $1.15 \pm 0.34$  mm to  $1.05 \pm 0.16$  mm & the change in WAG was from  $2.60 \pm 0.70$  mm to  $2.80 \pm 0.92$  mm (Table 3, Figure 5 & 8)

**INTER GROUP COMPARISON** - PD, RH, WAG and gingival biotype was compared 6 months after intervention between test and control group. The results showed that there was no statistically significant difference in PD, RH, and WAG between the two groups when the post treatment mean values were compared. The post-operative PD, RH and WAG in the test group were  $1.10 \pm 0.21$  mm,  $2.00 \pm 1.45$  mm and  $2.50 \pm 0.71$  mm respectively. In the control group, the PD, RH and WAG were  $1.05 \pm 0.16$  mm,  $1.60 \pm 1.60$  mm and  $2.80 \pm 0.92$  mm respectively (Table 4, Graph1)

The mean percentage reduction in PD in the test group and control group was  $24.17 \pm 27.90$  % and  $5.00 \pm 15.81$  % respectively which was not statistically significant. Similarly, the mean percentage decrease in RH in the test group and control group was  $37.33 \pm 31.58$  % and  $55.24 \pm 32.45$  % respectively which was not statistically significant. There was also no statistically significant mean percentage change in the WAG between the test and control groups ( $13.57 \pm 25.52$  % and  $6.67 \pm 14.05$  % respectively).

Chi Square test and McNemar's test was done to evaluate the change in the gingival biotype between the two groups, before and 6 months after the intervention. The results showed no statistically significant change in the gingival biotype before and at 6 months after the intervention. (Table 5, Table 6, Graph 2)

## Discussion

Several studies have shown promising results when placental membranes were used for root coverage procedures. The advantages of using this membrane are adequate amount of membrane available for single use, avoids second surgical site, limits post-operative morbidity, non-immunogenic, available in different dimensions and good shelf life of 2 years. Added advantage with chorion membrane allograft is to self-adhere at the site which aids in the stabilization of the membrane.<sup>5-12</sup>

The mean age of the subjects in the test group was  $34.2 \pm 4.1$  years and in the control group was  $41.3 \pm 9.4$

years. Patients reported minimal pain, discomfort postoperatively and uneventful soft tissue healing was observed in both the groups. These results suggest that chorion membrane is relatively a safe material for clinical use. This is in accordance with a study done in 2015 by *Chakraborty et. al.*<sup>7</sup>

On intra group comparison, it was found that both test and the control groups showed significant reduction in the RH. In the test group moreover along with the reduction in RH, there was also statistically significant reduction in the PD following therapy. The change in the RH before and after 6 months in the test group was in accordance with the finding of a study conducted by *Esteves J et.al* and *Pundir AJ et. al.* This reduction in the probing depth can be attributed to the fact that the chorion membrane contains cell adhesion bioactive factors such as fibronectin and laminin which promote cell adhesion, growth and differentiation and hence could have contributed for the reduction in PD.<sup>5,9</sup>

In the control group, no change in the PD was seen but there was statistically significant reduction in the RH 6 months after the intervention. The mean reduction in RH was from  $3.30 \pm 1.06$  mm to  $1.05 \pm 0.16$  mm post operatively. Our finding of SCTG showing significant reduction in RH in root coverage procedure is in accordance with the data evaluated in a study conducted by *Ramskrishnan T et al.* and *Nart J et. Al*, and *Randall J.*<sup>13-15</sup>

Like in the previously mentioned studies, there was an increase in the WAG in the test and control group after intervention. However, the change in our study was not statistically significant. This can be attributed to the relatively small sample size of the study. There was also no significant difference in the gingiva biotype at base line and at 6 months after intervention. In a study to evaluate if the gingival thickness played an important role in long term results following periodontal therapy the authors concluded that tooth brushing habits play an important part in maintaining gingival margin position post operatively than the thickness of the gingiva. In this study, patients were made aware about the most common cause of gingival recession and were educated in appropriate brushing technique to avoid chronic trauma to treated site. The patients maintained good oral hygiene that could have

contributed to the stability of the results despite not showing significant improvement in gingival biotype post operatively. However, long term follow-up of cases is required to arrive at definite conclusion.<sup>16</sup>

When inter group comparison was done, there was no statistically significant difference in the measured variable at the baseline and also, post-operatively, six months after the intervention. A split mouth study performed by *Lafzi et. al.* in 2016 has shown similar results.<sup>17</sup>

Although the result of both the test and control groups was comparable, the procedure like SCTG is very technique sensitive and difficult to perform in areas with thin biotype. The chorion membrane being an allograft carries a risk of graft rejection or transmission of disease from the source, however complications are rare. Though there was no significant difference in PD, RH, WAG and gingival biotype when the results of the test and control groups were compared, the test group showed significant reduction in RH and PD in the test group and hence can be successfully used as an alternative in case where harvesting a SCTG is not a feasible option due to factors such as anatomic limitations, patient co-operation, cost etc.

Limitations of this study: sample size is small and long-term follow-up are required.

### Conclusion

On intra group comparison, both test and control group showed significant reduction in RH, 6 months after intervention. Along with reduction in RH, the test group also showed reduction in PD. But when both the test and control groups were compared, there was no significant difference in the measured parameters such as RH, PD, WAG and gingival biotype. Hence it can be concluded that HCMA is equally as efficient as SCTG in the treatment of gingival recession.

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TABLE 1

Variables	Group	N	Mean	SD	Mean Diff	Z	P-Value
PD	Test	10	1.75	1.01	0.60	-1.801	0.07
	Control	10	1.15	0.34			
RH	Test	10	2.90	1.20	-0.40	-0.806	0.42
	Control	10	3.30	1.06			
WAG	Test	10	2.35	0.91	-0.25	-0.436	0.66
	Control	10	2.60	0.70			

\*Comparison of mean values of PD, RH and WAG (in mm) at Baseline between the Test and Control groups using Mann Whitney U test. The level of significance was set at P<0.05

TABLE 2

Variables	Time	N	Mean	SD	Mean Diff	Z	P-Value
PD	Pre Rx	10	1.75	1.01	0.65	-2.032	0.04*
	Post Rx	10	1.10	0.21			
RH	Pre Rx	10	2.90	1.20	0.90	-2.388	0.02*
	Post Rx	10	2.00	1.45			
WAG	Pre Rx	10	2.35	0.91	-0.15	-1.134	0.26
	Post Rx	10	2.50	0.71			

†Comparison of mean values of PD, RH and WAG (in mm) at baseline and 6 months postoperatively in Test Group using Wilcoxon Signed ranked test. The level of significance was set at P<0.05.

TABLE 3

Variables	Time	N	Mean	SD	Mean Diff	Z	P-Value
PD	Pre Rx	10	1.15	0.34	0.10	-1.000	0.32
	Post Rx	10	1.05	0.16			
RH	Pre Rx	10	3.30	1.06	1.70	-2.536	0.01*
	Post Rx	10	1.60	1.60			
WAG	Pre Rx	10	2.60	0.70	-0.20	-1.414	0.16
	Post Rx	10	2.80	0.92			

‡Comparison of mean values of PD, RH and WAG (in mm) at baseline and 6 months postoperatively in control Group using Wilcoxon Signed ranked test. The level of significance was set at P<0.05.

TABLE 4

Variables	Graft	N	Mean	SD	Mean Diff	Z	P-Value
PD	Test	10	1.10	0.21	0.05	-0.610	0.54
	Control	10	1.05	0.16			
RH	Test	10	2.00	1.45	0.40	-0.735	0.46
	SCTG	10	1.60	1.60			
WAG	Test	10	2.50	0.71	-0.30	-0.706	0.53
	Control	10	2.80	0.92			

§ Comparison of mean values of PD, RH and WAG (in mm) at 6 months Post treatment period between test and control group using Mann Whitney U test. The level of significance was set at P<0.05

TABLE 5

Time	Gingival Biotype	Test		Control		c <sup>2</sup> Value	P-Value
		n	%	n	%		
Pre Rx	Thin	4	40%	1	10%	2.400	0.12
	Thick	6	60%	9	90%		
Post Rx	Thin	1	10%	0	0%	1.053	0.31
	Thick	9	90%	10	100%		

k Comparison of Gingival Biotypes between Test and Control groups at baseline and at 6 months using Chi Square test. The level of significance was set at P<0.05.

TABLE 6

Group	Gingival Biotype	Pre Rx		Post Rx		P-Value
		n	%	n	%	
Test	Thin	4	40%	1	10%	0.25
	Thick	6	60%	9	90%	
Control	Thin	1	10%	0	0%	0.96
	Thick	9	90%	10	100%	

¶ Comparison of Gingival Biotypes between Test and Control groups at baseline and at 6 months using McNemar's test. The level of significance was set at P<0.05.

## LEGENDS

Figure 1 - Preoperative picture with baseline measurement (test site).

Figure 2 - Chorion membrane placed after incision and reflection of the flap.

Figure 3 - Flap being coronally advanced and sutured.

Figure 4- Postoperative picture after 6 months with measurement (test site).

Figure 5 - Preoperative picture with baseline measurement (control group).

Figure 6 - subepithelial connective tissue placed after incision and reflection of the flap.

Figure 7 - Flap being coronally advanced and sutured.

Figure 8- Postoperative picture after 6 months with measurement (control site).

Figure 9 - Comparison of mean values of PD, RH and WAG (in mm) after 6 months postoperatively between Test and Control groups.

Figure 10 - Comparison of gingival biotype at baseline and after 6 months postoperatively between Test and Control groups.

**Figure 1**





**Figure 2**



**Figure 3**

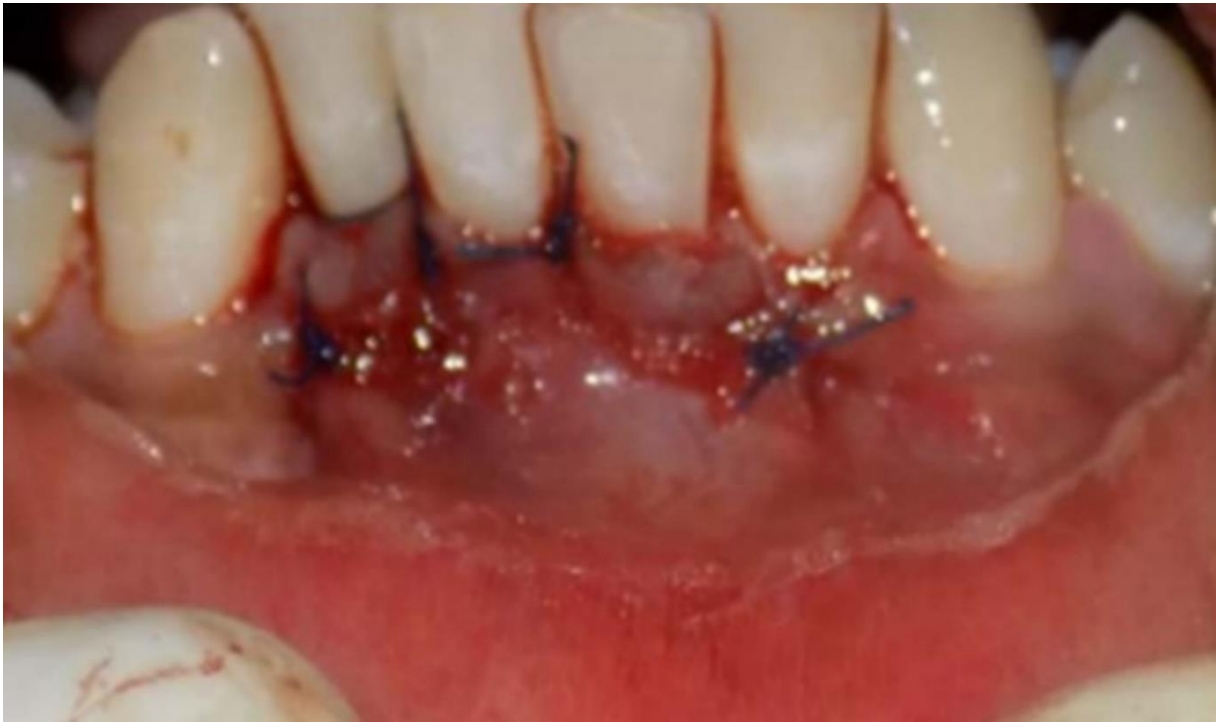


Figure 4



Figure 5



**Figure 6**



**Figure 7**



Figure 8



Figure 9

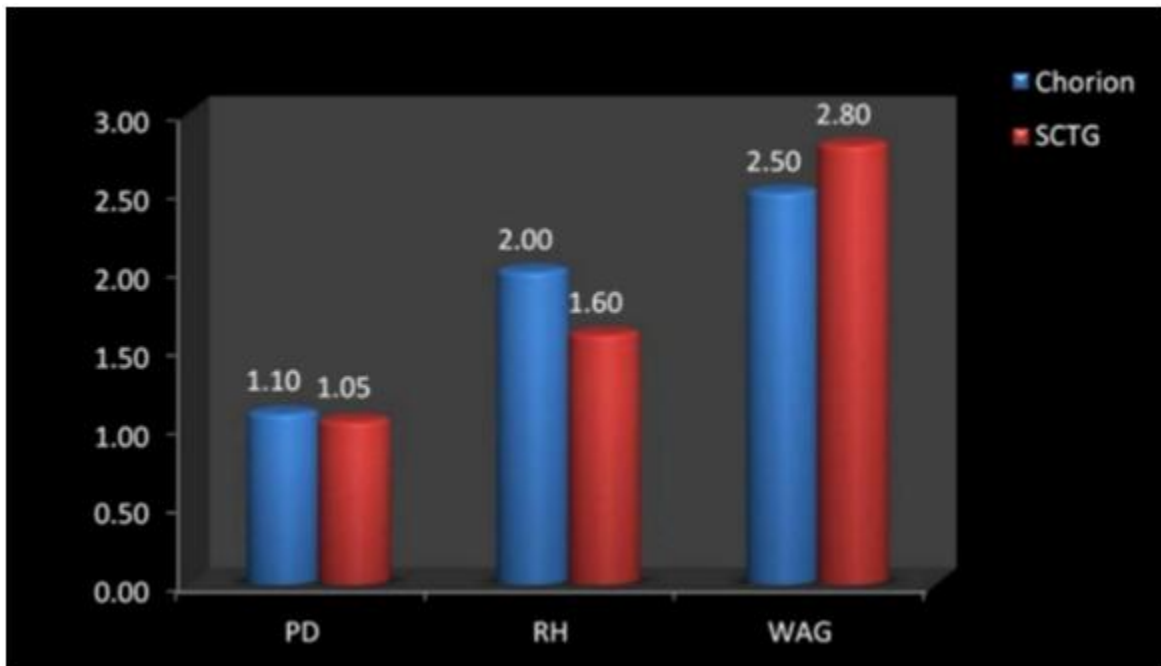


Figure 10

