



UNIVERSIDADE ESTADUAL DO OESTE DO PARANÁ
CENTRO DE CIÊNCIAS BIOLÓGICAS E DA SAÚDE
PROGRAMA DE PÓS-GRADUAÇÃO EM
ODONTOLOGIA (PPGO) - MESTRADO



EDSON OLIVEIRA SILVA

Fibrina Rica em Plaquetas *Advanced* (A-PRF) associada à técnica de tunelização
no tratamento de recessões gengivais tipo 1 de Cairo: Estudo clínico,
randomizado e cego.

Cascavel-PR
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Dissertação apresentada ao Programa de Pós-Graduação em Odontologia *Stricto Sensu* – Nível Mestrado, do Centro de Ciências Biológicas e da Saúde, da Universidade Estadual do Oeste do Paraná, como requisito parcial para obtenção do título de Mestre em Odontologia.

Área de concentração: Odontologia

Orientador: Profa. Dra. Patricia Oehlmeyer Nassar

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Oliveira Silva, Edson
Of Fibrina Rica em Plaquetas Advanced (A-PRF)
associada à técnica de tunelização no tratamento de
recessões gengivais tipo 1 de Cairo: Estudo clínico,
randomizado e cego / Edson Oliveira Silva;
orientadora Patrícia Oehlmeyer Nassar. --
Cascavel, 2021.
57 p.

Dissertação (Mestrado Acadêmico Campus de
Cascavel) -- Universidade Estadual do Oeste do
Paraná, Centro de Ciências Biológicas e da Saúde,
Programa de Pós-Graduação em Odontologia, 2021.

1. fibrina rica em plaquetas. 2. recessão
gengival. 3. hipersensibilidade dentinária. I.
Oehlmeyer Nassar, Patricia , orient. II. Título.

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Dissertação apresentada ao Programa de Pós-Graduação em Odontologia em cumprimento parcial aos requisitos para obtenção do título de Mestre em Odontologia, área de concentração Odontologia, linha de pesquisa Patologia Aplicada à Clínica Odontológica, APROVADO(A) pela seguinte banca examinadora:



Orientador(a) - Patrícia Oehlmeyer Nassar

Universidade Estadual do Oeste do Paraná - Campus de Cascavel (UNIOESTE)



Ricardo Augusto Conci

Universidade Estadual do Oeste do Paraná - Campus de Cascavel (UNIOESTE)



Greison Rabelo de Oliveira

Centro Universitário Ingá (UNINGA)

AGRADECIMENTOS

A Deus, pela dádiva da vida, e por me dar forças para não desistir nos momentos de angústia, por ser minha direção e me guiar na busca pelos meus sonhos. Gratidão a ti, Senhor!

Aos meus pais, Aureo e Belquis, e meus irmãos **Jhosiane e Rafael**, por serem meus exemplos, por ensinarem os valores que tenho comigo, por muitas vezes abdicarem de seus sonhos para que os meus se realizassem, por serem minha base. Sem vocês, eu não seria a pessoa que sou hoje. Gratidão por tudo, amo vocês!

A minha esposa, Bruna, por sempre acreditar em mim, mesmo quando eu mesmo já não acreditava, por me incentivar a ser melhor a cada dia, e não me deixar desistir, sendo meu refúgio e porto seguro frente aos momentos ruins, pelo apoio todos os dias ao longo destes anos, e por entender as vezes que tive de renunciar a momentos nossos, para poder me dedicar as atividades do mestrado. Obrigado por tudo! Eu amo você!

A minha orientadora, Profa. Patricia, por sempre estar presente, e por personalizar de maneira tão perfeita a palavra Professor. Desde os tempos da graduação fazendo aflorar em mim, o amor pela periodontia. Gratidão pela amizade, o carinho, por todo conhecimento compartilhado, me ajudando em todos os momentos, e por sempre acreditar em mim. Você é, e sempre será minha referência na docência. Minha eterna gratidão!

Ao Prof. Carlos, por todo conhecimento compartilhado, pela amizade, pela direção dada e dúvidas sanadas durante as cirurgias, sempre pronto para responder, de forma humilde, clara e simples. Você também demonstra o real significado da palavra professor. Gratidão por tudo.

Aos amigos por sempre me incentivarem a ser a minha melhor versão, sempre com uma palavra de apoio nos momentos em que mais precisei. Obrigado.

Ao Leonardo, Simone e Leticia, grandes profissionais e amigos que sempre estavam dispostos a ajudar na realização desta pesquisa, pela ajuda na realização das cirurgias, nas triagens de pacientes, por uma palavra amiga frente a uma dificuldade, ou simplesmente pelas risadas que compartilhamos neste período. Muito obrigado!

A Unioeste, minha segunda casa, pelo acolhimento incomparável, e por fornecer o que foi possível para a realização desta pesquisa.

Aos funcionários da Unioeste, em especial a **Maria Cristina**, que realizou as venopunções, sempre pronta para ajudar no que fosse possível.

Aos pacientes desta pesquisa que confiaram seus anseios sob minha responsabilidade profissional, e sempre estiverem dispostos a colaborar com este estudo. Foi através de vocês que esta conquista foi possível.

“Quanto mais aumenta nosso conhecimento, mais evidente fica nossa ignorância”.

(John F. Kennedy)

Fibrina Rica em Plaquetas *Advanced* (A-PRF) associada à técnica de tunelização no tratamento de recessões gengivais tipo 1 de Cairo: Estudo clínico, randomizado e cego.

RESUMO

O uso de Fibrina Rica em Plaquetas *advanced* (A- PRF) vem demonstrando bons resultados na cirurgia plástica periodontal para o tratamento de recessões gengivais. Este estudo tem por objetivo avaliar a efetividade do uso do A-PRF no recobrimento radicular e nos parâmetros clínicos periodontais de pacientes portadores de recessão gengival Tipo I de Cairo. Foram selecionadas 44 recessões gengivais Tipo 1 de Cairo bilaterais. As recessões foram divididas em dois grupos de acordo com o procedimento cirúrgico realizado (n=44): ETCS: Enxerto de Tecido Conjuntivo Subepitelial e A-PRF, utilizando a técnica cirúrgica de tunelização em ambos os lados. Os pacientes foram acompanhados aos 0, 90 e 180 dias para avaliação da altura e largura da recessão; Profundidade de sondagem e Sangramento à Sondagem; Nível de inserção clínica; Largura da faixa de tecido queratinizado; Espessura do tecido gengival queratinizado e Grau de sensibilidade. Os dados obtidos foram analisados e avaliados inicialmente por meio do teste de SHAPIRO-WILK para normalidade e posteriormente, utilizado o teste de ANOVA e TUKEY para dados normais ou Teste KRUSKALL-WALLIS e DUNN para dados não normais. Para análise do parâmetro de sensibilidade, o teste de escolha foi o de KRUSKALL-WALLIS e DUNN, com nível de significância de 5%. Na análise entre grupos foi realizado o teste T de STUDENT. Ambos os grupos apresentaram melhora nos parâmetros periodontais, porém sem diferença estatística significativa entre eles, o que sugere que o A-PRF pode ser uma alternativa ao ECTS nas cirurgias de recobrimento radicular de recessões tipo I de Cairo.

Palavras-chave: fibrina rica em plaquetas, recessão gengival, hipersensibilidade da dentina.

Advanced platelet-rich fibrin (A-PRF) associated to tunnel technique in the treatment of Cairo type 1 gingival recession: Clinical, randomized and blind study.

ABSTRACT

The use of advanced platelet-rich fibrin (A- PRF) has presented good results in periodontal plastic surgery for treatment of gingival recessions. The present study aims at assessing the effectiveness of the use of A-PRF in root coverage and periodontal clinical parameters of patients with Cairo Type 1 gingival recession. Forty-four (44) bilateral Cairo Type 1 gingival recessions were selected. The recessions were split in two groups according to the surgical procedure made (n=44): SCTG: Subepithelial Connective Tissue Graft and A-PRF, using tunnel surgical technique in both sides. The patients were monitored on 0, 90, and 180 days for assessment of the recession height and width; probing depth and bleeding on probing; clinical attachment level; keratinized tissue band width; thickness of keratinized gingival tissue and sensitivity level. The data obtained were initially analyzed and assessed with SHAPIRO-WILK test for normality and later ANOVA and TUKEY tests were used for normal data, or KRUSKALL-WALLIS and DUNN test for non-normal data. For analysis of sensitivity parameter, the test chosen was KRUSKALL-WALLIS and DUNN, with 5% significance level. In the analysis across groups the Student's *t*-test was conducted. Both groups presented improvement in periodontal parameters, however without significant statistical difference between them, which suggests that A-PRF can be an alternative to SCTG in root coverage surgeries of Cairo type 1 recession.

Keywords: platelet-rich fibrin; gingival recession; dentin hypersensitivity.

LISTA DE SIGLAS E ABREVIATURAS

JCE – Junção cimento esmalte

mm – Milímetros

NG – Nível Gengival

NI – Nível de inserção

PS – Profundidade de sondagem

Dissertação elaborada e formatada conforme as normas das publicações científicas: *Saudi Dental Journal*, Disponível em: https://www.elsevier.com/wps/find/journaldescription.cws_home/722876?generatepdf=true

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1- Introdução

As recessões gengivais podem causar danos estéticos ao sorriso, além de trazerem complicações como hipersensibilidade dentinária e cáries cervicais. A recessão gengival é definida como o deslocamento apical da margem gengival em relação à junção cimento-esmalte (JEC) e está associada à perda de inserção e à exposição da superfície da raiz ao meio bucal. A inflamação gengival em resposta ao acúmulo do biofilme dental ou ao trauma mecânico de escovação tem sido indicada como fator etiológico principal dessas lesões (Bedoya e Park, 2014; Öncü, 2017). O acúmulo de biofilme, trauma local e a ausência de gengiva inserida são os fatores etiológicos associados a essa patologia (Cortellini et al., 2018).

Os seus fatores etiológicos podem ser divididos em: fatores desencadeantes e predisponentes (Fu J et al., 2012). Dentro dos fatores desencadeantes estão o trauma de escovação, o trauma oclusal, a inflamação gengival, os tratamentos iatrogênicos e o tabagismo (Borghetti A., 2002). Já os fatores predisponentes são essencialmente as características anatômicas do paciente e que podem influenciar na ocorrência de recessões gengivais, bem como: o biotipo gengival, as variações da morfologia óssea subjacente, posição dentária incorreta, a presença de freios de inserção alta e de vestíbulo curto (Fu J et al., 2012).

Atualmente a classificação mais utilizada é a de Cairo que tem como parâmetro a perda de tecido de inserção interproximal: RT1) recessões de tecido mole que não apresentam perda de inserção interproximal; RT2) quando há perda de inserção interproximal, com distância da junção amelocementária - JCE ao fundo de sulco/bolsa menor ou igual à perda de inserção vestibular (medida da JCE ao fundo de sulco/bolsa vestibular); e RT3) Perda de inserção interproximal, com distância da JCE ao fundo de sulco/bolsa maior que a perda de inserção vestibular (Cairo et al., 2011; Steffens e Marcantonio, 2018).

Para o tratamento das recessões gengivais, diversos procedimentos cirúrgicos foram utilizados nos últimos anos, incluindo o reposicionamento lateral do retalho, enxerto gengival livre, cirurgia de tunelização com e sem enxerto de tecido conjuntivo subepitelial (ETCS), retalho posicionado coronalmente com ou sem ETCS, regeneração tecidual guiada com membranas, matriz dérmica acelular, plasma rico em plaquetas (PRP) e fibrina rica em plaquetas (PRF). (Aroca et al., 2009; Padma et al., 2013; Gupta et al., 2015; Tunali et al., 2015; Oncu et al., 2017).

A aplicação de ETCS tem sido amplamente utilizado no tratamento de recessões gengivais, mesmo possuindo como sua maior desvantagem, a necessidade de tecido a partir de uma área doadora, o que normalmente resulta em um maior desconforto pós-operatório, sendo que, em casos em que são necessárias várias áreas a serem intervencionadas, é preciso uma grande quantidade de tecido doador. É neste âmbito que o L-PRF surge como proposta alternativa, no sentido de substituir a utilização do ETCS, pois demonstra grandes quantidades de fatores de crescimento, uma facilidade na sua obtenção e com um pós-operatório muito favorável (Moraschini e Barboza, 2016).

A Fibrina Rica em Plaquetas e Leucócitos (L – PRF) é um agregador plaquetário obtido por meio da centrifugação sanguínea, que passou a ser utilizada em vários procedimentos de regeneração tecidual na medicina e na odontologia a partir dos estudos de Choukroun em 2001. Trata-se de um arcabouço de fibrina tridimensional com células (leucócitos), fatores de crescimento, citocinas e fatores angiogênicos no seu interior, potencializando o reparo e regeneração tecidual. Na odontologia, vem sendo empregado amplamente em implantodontia, nas reconstruções ósseas, e nas cirurgias periodontais, como no tratamento das recessões gengivais (Choukroun et al., 2016; Dohan et al., 2009; Aroca et al., 2009).

Os efeitos do A-PRF (*Advanced* PRF), vem sendo relatado por possuir uma maior quantidade de células (macrófagos e monócitos) e maior quantidade de fatores de crescimento, devido a sua centrifugação mais lenta, quando comparado ao L-PRF. (Kobayashi et al., 2016). O L-PRF padrão, foi melhorado em A-PRF (*Advanced* – PRF) por possuir uma maior quantidade de células (macrófagos e monócitos) e por suportar maior liberação de fator de crescimento em A-PRF (Ghanaati et al., 2014).

Em princípio, um menor tempo de centrifugação aumenta o número total de células contidas na camada superior do PRF, permitindo um número maior de leucócitos “aprisionados” dentro da matriz de fibrina. (Choukroun et al., 2006; Ghanaati et al., 2014; Kobayashi et al., 2016).

Sendo assim, o objetivo primário desta pesquisa foi avaliar a efetividade das membranas de Fibrina Rica em Plaquetas *Advanced* (A-PRF) associado a técnica de tunelização no recobrimento radicular em pacientes portadores de recessões gengivais tipo I de Cairo. objetivo secundário, comparar os resultados do recobrimento radicular na técnica de tunelização utilizando-se o enxerto de tecido conjuntivo sub epitelial versus a membrana A-PRF, avaliar o grau de hipersensibilidade dentinária após a realização do recobrimento radicular, bem como a estabilidade clínica dos resultados nas duas situações com preservação de 180 dias.

2- Metodologia

2.1- Tipo de Estudo

A pesquisa realizada é de natureza clínica aplicada, sendo um estudo prospectivo, com grupos em boca-dividida e paralelos e randomizado.

2-2 Aspectos Éticos

O estudo foi submetido e aprovado no Comitê de Ética e Pesquisa em Seres Humanos da Unioeste (número do parecer 3.359.508, CAAE 13806519.0.0000.0107, 31 de maio de 2019). O local de sua realização foi nas Clínicas do Curso de Odontologia do Campus de Cascavel da Unioeste. Após a aprovação pelo Comitê de Ética, foi explicado a todos os pacientes o objetivo deste estudo, sendo então incluídos como participantes do estudo após a assinatura do termo de consentimento livre e esclarecido (TCLE).

2.3- Coleta de Dados

2-3.1- Critérios de Inclusão

Pacientes que apresentaram recessões gengivais tipo 1 de Cairo (figura 01), bilaterais, com faixa etária de 18 a 50 anos, de ambos os gêneros, em pelo menos uma das arcadas, com recessões variando entre 2 e 5 milímetros em altura e entre 2 a 4 milímetros em largura. Os demais dentes da arcada dentária com saúde periodontal, com todos os sítios apresentando profundidade de sondagem menor ou igual a 3mm, com índice de sangramento gengival menor ou igual a 5%, sem inflamação gengival e livres de cáries.



Figura 01 – Recessões Gengivais Tipo I de Cairo bilaterais, selecionadas na pesquisa

2.3.2- Critérios de Exclusão

Os seguintes critérios de exclusão foram utilizados: história positiva de antibioticoterapia nos últimos seis meses e de antiinflamatórios esteróides ou não esteróides nos três meses antecessores ao estudo; história positiva de gestação; história positiva de tabagismo ou interrupção definitiva do hábito em até 5 (cinco) anos; história positiva de qualquer problema sistêmico que contra-indicasse o procedimento cirúrgico; tratamento endodôntico ou patologia pulpar no dente envolvido; interferências oclusais patogênicas e história de cirurgia prévia no local. O histórico médico de cada paciente foi obtido por meio de anamnese e todos os participantes foram submetidos a um exame clínico.

2.4- Cálculo Amostral

O cálculo do tamanho da amostra para o número de recessões gengivais foi realizado através do Teste T para amostras independentes com um poder de teste de 80% e nível de alfa de 0.05, sendo esses dados baseados em estudos prévios do grupo de pesquisadores. (Tonet et al., 2010; Nassar et al., 2014; Spada et al., 2017).

2.5- Desenho do Estudo

2.5.1- Avaliação Periodontal

O exame clínico/periodontal inicial foi realizado por um único operador previamente calibrado que, por meio de uma sonda periodontal do Tipo Willians nº 23, determinando:

1. Altura da recessão: medida como a distância da junção cimento-esmalte à margem gengival, mensurada em mm;
2. Largura da recessão: distância mésio-distal da margem gengival, mensurada em mm;
3. Profundidade de sondagem: distância da margem gengival (MG) até o fundo do sulco/bolsa com registro de presença ou ausência e mensurada em mm.
4. Sangramento à Sondagem: a cada três dentes com intervalo de 30 segundos aproximadamente para registro do parâmetro o que corresponde ao tempo de sondagem com registro de presença ou ausência.
5. Nível de inserção clínica: distância da junção cimento-esmalte (JCE) até o fundo do sulco/bolsa, com registro de presença ou ausência e mensurada em mm. Obtida a partir da soma entre a recessão gengival e profundidade clínica de sondagem.

6. Altura da faixa de tecido queratinizado: sendo a distância da junção mucogengival até a margem gengival;
7. Espessura do tecido gengival queratinizado em 3 pontos (Mesial, central e distal);
8. Grau de sensibilidade (Índice modificado da U.S. Public Health Service composto de três escores: 0 (Alpha) - sem sensibilidade; 2 (Bravo) - pequena sensibilidade; 3 (Charlie) - alta sensibilidade. (Camilotti et al., 2012).

Após o exame clínico periodontal inicial, as recessões foram divididas, aleatoriamente, em dois grupos (Gerador de Grupo Aleatório – Web Site - <https://pt.rakko.tools/tools/59/>), segundo os tratamentos propostos na figura 02.

Foram selecionados um total de 49 recessões na faixa etária de 26 a 46 anos de ambos os gêneros. Um total de quarenta e quatro recessões foram selecionadas para receberem os procedimentos cirúrgicos periodontais. Cada paciente recebeu as duas técnicas cirúrgicas, em uma hemi-arcada a tunelização associado ao enxerto de tecido conjuntivo subepitelial (Tunelização + ECTS) e na outra arcada a tunelização associado as membranas de A-PRF, sendo que a distribuição de qual hemi-arcada receberia determinada técnica foi feita de forma aleatória. As recessões foram alocadas em cada grupo de forma aleatória através de sorteio (Gerador de Grupo Aleatório – Web Site).

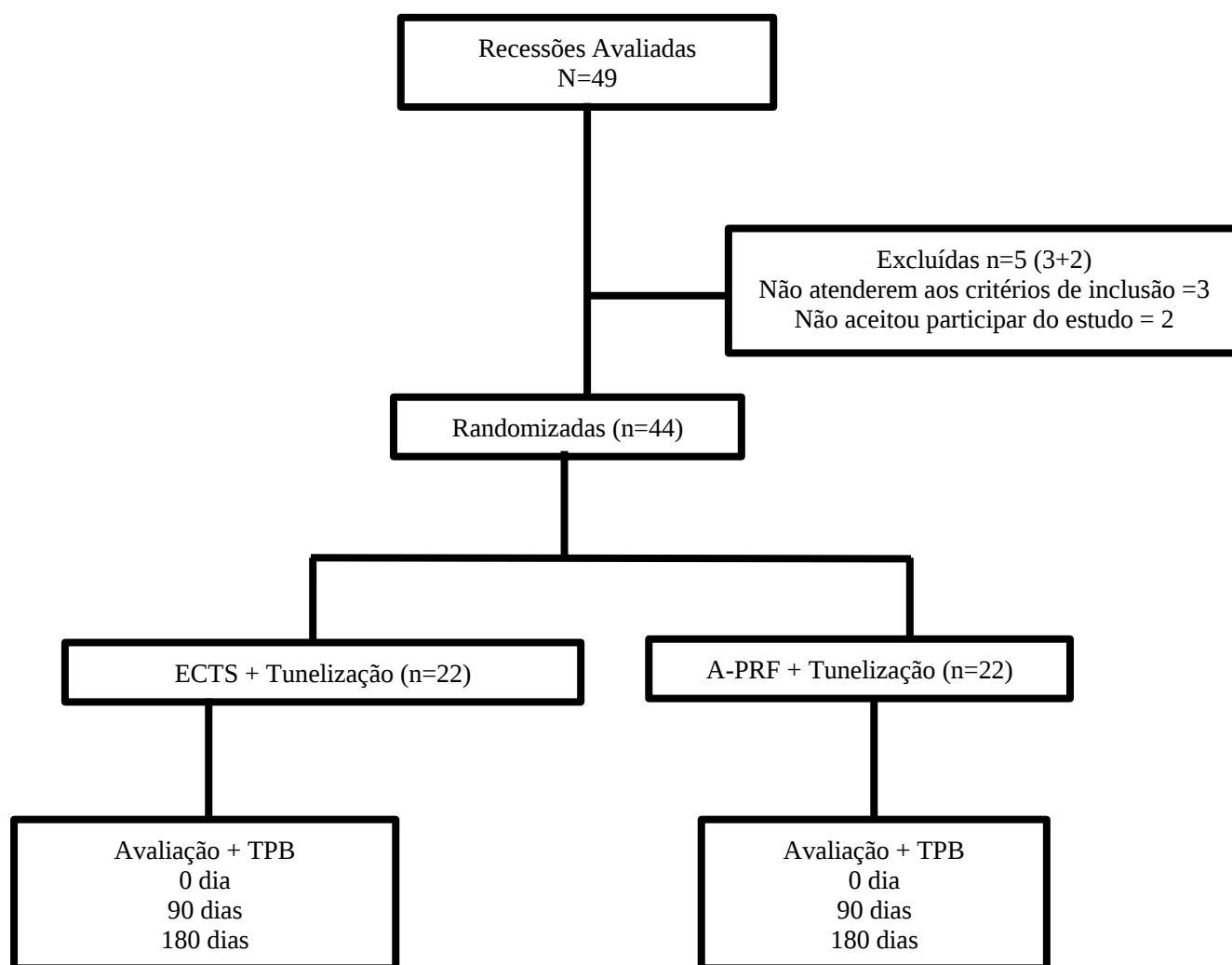


Figura 02. Distribuição das 44 recessões de acordo com os tratamentos propostos.

O tratamento periodontal foi realizado por um único operador, sendo realizado instrumentação manual e ultra-sônica. Para instrumentação manual, foram utilizadas curetas periodontais de Gracey 5/6, 7/8, 11/12 e 13/14 e para instrumentação ultra-sônica foi utilizado um aparelho ultra sônico. Após a realização do tratamento periodontal básico, instrução de higiene oral e o exame clínico inicial, cada recessão foi incluída em um dos dois grupos selecionados, de acordo com a técnica cirúrgica.

2.6- Técnicas cirúrgicas

2.6.1- Técnica 01: Enxerto de tecido conjuntivo subepitelial com tunelização:

A área receptora foi anestesiada com anestesia terminal infiltrativa, com o anestésico articaina 4% com Epinefrina 1:100.000 (DFL, Rio de Janeiro, RJ, Brasil). Foi realizada incisão intrasulcular com lâmina 15c (*SOLIDOR®*, Suzhou Kyuan Medical Apparatus Co. Ltd., Suzhou City, Beiqiao Town, China), incisões intrasulculares na região cervical, ao nível da JCE, para mesial e distal. Criação do túnel com a utilização de tunelizadores (*Supremo Instrumentais Cirúrgicos*, São Paulo, Brasil), por meio de um retalho de espessura total próximo a margem gengival, até ultrapassar a junção mucogengival, de modo a se obter um tecido sem tensões. A raiz do dente foi raspada e alisada com cureta periodontal do tipo Gracey específica para a região, seguida por irrigação abundante com soro fisiológico 0,9% (Figura 03).

O ETCS foi obtido do palato, através da técnica de incisão linear. A região doadora foi suturada com suturas simples com fio de sutura Nylon 5.0 (*SHALON®* São Luis M. Belos, Goiás, Brasil), em seguida posicionado sobre a recessão gengival e estabilizado nas proximais com fio de polipropileno 5.0 (*TECHSUTURE®*, Bauru, São Paulo, Brasil) ao nível da JCE. Posteriormente o tecido foi tracionado coronalmente por meio de suturas verticais fixadas na região incisal/vestibular dos elementos dentários envolvidos, utilizando um fio de polipropileno 5.0 (*TECHSUTURE®*, Bauru, São Paulo, Brasil).



Figura 03 – Leito receptor preparado através da técnica de tunelização.

2.6.2- Técnica 02: Tunelização e *Advanced* - Fibrina Rica em Plaquetas (A-PRF)

Protocolo para obtenção e utilização do A-PRF (Choukroun e Ghanaati et al., 2018):

A sequência do processo de obtenção do A-PRF é basicamente dividida em três etapas:

1ª Etapa: Punção venosa e coleta do sangue.

2ª Etapa: Separação celular (centrifugação).

3ª Etapa: Preparo das membranas de A-PRF.

1ª Etapa: Punção venosa e coleta do sangue: Antes do início do procedimento cirúrgico para recobrimento radicular, foi realizada a venopunção de cerca de 60 ml de sangue (de acordo com a extensão do leito receptor). O protocolo de venopunção seguiu todas as recomendações do Ministério da Saúde, com dispositivo de Coleta à Vácuo, seguindo os passos: Rosquear a agulha no adaptador (canhão). Ajuste do garrote e escolha a veia; anti-sepsia do local da coleta com algodão umedecido em álcool a 70% ou álcool. Realização da punção, introduzir o tubo no suporte, pressionando-o até o limite; soltar o garrote assim que o sangue começar a fluir no tubo até o enchimento; orientar o paciente a pressionar com algodão a parte puncionada, mantendo o braço estendido, sem dobrá-lo.

2ª Etapa: Separação celular (centrifugação): O sangue coletado, foi imediatamente à centrífuga (*Centrifuga Spinlab, São Paulo, Brasil*), em volumes por tubos de 10ml. As membranas de fibrina foram obtidas por centrifugação de 1500 rpm por 14 minutos. Ao final da centrifugação, os tubos foram recolhidos e a fibrina não foi removida de imediato. Foi aguardado pelo menos 30 minutos, até o momento da sua utilização no leito cirúrgico.

3ª Etapa: Preparo das membranas de A-PRF: Com o Kit próprio para confecção de membranas de PRF em inox (*Supremo Instrumentais Cirúrgicos, São Paulo, Brasil*) a porção intermediária centrifugada, o coágulo de fibrina, foi separada da porção de células vermelhas e do plasma pobre em plaquetas, e depositado na caixa de inox, sendo aplicada a tampa compressiva, sem apertá-la, por um período de 15 minutos. O próprio peso (130g) é suficiente para comprimir o coágulo para obter as membranas, sem oferecer danos às estruturas celulares presentes na malha de fibrina. (Figura 04).

Preparo do leito receptor e inserção das membranas de A-PRF: (Aroca et al., 2009): A área receptora foi anestesiada com anestesia terminal infiltrativa, com o anestésico articaína 4% com Epinefrina 1:100.000 (DFL, Rio de Janeiro, RJ, Brasil). Foi realizada incisão intrasulcular com lâmina 15c (*SOLIDOR®*, Suzhou Kyuan Medical Apparatus Co. Ltd., Suzhou City, Beiqiao Town, China), na região cervical, ao nível da JCE, para mesial e distal. Criação do túnel com a utilização de tunelizadores (*Supremo Instrumentais Cirúrgicos, São Paulo, Brasil*), por meio de um retalho de espessura total próxima a margem gengival, até ultrapassar a junção mucogengival. de modo a se obter um tecido sem tensões. Foram utilizadas 4 membranas de A-PRF que foram posicionadas juntas e suturadas no interior do túnel mucoperiosteal criado. Posteriormente o tecido foi tracionado coronalmente por meio de suturas verticais fixadas com resina composta na região incisal/vestibular dos elementos dentários envolvidos, utilizando um fio de polipropileno 5.0 (*TECHSUTURE, Bauru, São Paulo, Brasil*). (Figura 05)



Figura 04 – Aspecto da membrana de fibrana após a centrifugação.



Figura 05 – Membranas de A-PRF e ECTS suturadas no leito receptor.

2.7- Controle e avaliações pós-operatórias

Os pacientes receberam medicações para controle da dor, antibioticoterapia, anti-inflamatório não esteroide, analgésico e colutório bucal (Amoxicilina 500mg de 8/8 horas por 7 dias, nimesulida 100mg de 12/12 horas por 3 dias, dipirona 500mg/5ml por 3 dias e digluconato de clorexidina 0,12% de 12/12 horas por 15 dias). As suturas da região doadora foram removidas com 7 dias e as suturas do leito receptor removidas com 15 dias.

Os pacientes foram avaliados por um período total de 180 dias, sendo que os exames clínicos foram realizados nos períodos de 0, 90 e 180 dias e em todos os períodos, os pacientes receberam a terapia de manutenção, sendo reavaliados todos os critérios do exame periodontal inicial. (Figuras 06 e 07). Após os 180 dias de período experimental, todos os pacientes foram incluídos em um programa de manutenção periodontal.



Figura 06 – Aspecto pós-operatório de 180 dias do Grupo A-PRF.



Figura 07 – Aspecto pós-operatório de 180 dias do Grupo ECTS.

2.8- Análise Estatística

Quanto à análise estatística, o programa utilizado foi o GraphPad Prism 8.0®. Os dados clínicos foram analisados e avaliados inicialmente por meio do teste de SHAPIRO-WILK para verificação de normalidade, o teste de ANOVA e TUKEY foi utilizado para os dados normais e o Teste KRUSKALL-WALLIS e DUNN para dados não normais. Para análise do parâmetro de sensibilidade e sangramento foram utilizados os testes de KRUSKALL-WALLIS e DUNN. Na análise entre grupos foi utilizado o teste T de STUDENT, com nível de significância de 5%.

3- Resultados

Foram analisadas 44 recessões gengivais, sendo 22 do grupo tunelização + ECTS e 22 do grupo tunelização + A-PRF.

A tabela 1 demonstra os valores dos parâmetros clínicos periodontais do grupo tunelização + A-PRF analisados ao final de 180 dias, com todos os parâmetros apresentando melhoras estatisticamente significantes após o acompanhamento de 180 dias ($p < 0,05$).

Tabela 1 - Análise comparativa dos parâmetros periodontais do grupo Tunelização + A-PRF no período inicial e final. Os valores representam média $\pm$ desvio padrão

Parâmetros (mm)	Tunelização + A-PRF (inicial)	Tunelização + A-PRF (90 dias)	Tunelização + A-PRF (180 dias)
Largura da Recessão	2,68 $\pm$ 0,64A	1,63 $\pm$ 0,95B	1,53 $\pm$ 0,63B
Altura da Recessão	2,13 $\pm$ 0,83A	1,05 $\pm$ 0,62B	1,13 $\pm$ 0,51B
Profundidade de Sondagem	1,71 $\pm$ 0,38A	1,35 $\pm$ 0,37B	1,08 $\pm$ 0,26B
Nível de Inserção Clínica	3,18 $\pm$ 0,85A	2,21 $\pm$ 0,71B	2,13 $\pm$ 0,51B
Altura da Gengiva Inserida	1,37 $\pm$ 0,47A	2,21 $\pm$ 0,53B	2,33 $\pm$ 0,81B
Grau de Sensibilidade	1,68 $\pm$ 0,99A	0,68 $\pm$ 0,94B	0,4 $\pm$ 0,63B
Índice se Sangramento	0,31 $\pm$ 0,64A	0 $\pm$ 0A	0,6 $\pm$ 0,50B
Espessura do Tecido	1,40 $\pm$ 0,50A	2,42 $\pm$ 0,76B	2,26 $\pm$ 0,70B

NOTA: Letras diferentes significam que são diferentes estatisticamente dentro de cada grupo de tratamento ($p < 0,05$).

A Tabela 2, demonstra os valores dos parâmetros clínicos periodontais do grupo tunelização + ECTS analisados ao final de 180 dias, onde todos os parâmetros periodontais apresentaram uma melhora estatisticamente significativa, exceto o índice de sangramento.

Tabela 2 - Análise comparativa dos parâmetros periodontais do grupo Tunelização + ECTS, no período inicial e final. Os valores representam média $\pm$ desvio padrão.

Parâmetros (mm)	Tunelização + ECTS (inicial)	Tunelização + ECTS (90 dias)	Tunelização + ECTS (180 dias)
Largura da Recessão	2,5 $\pm$ 0,51A	1,73 $\pm$ 0,73B	1,26 $\pm$ 0,59B
Altura da Recessão	2,04 $\pm$ 0,57A	0,89 $\pm$ 0,56B	0,93 $\pm$ 0,45B
Profundidade de Sondagem	0,71 $\pm$ 0,52A	1,24 $\pm$ 0,39B	1,06 $\pm$ 0,13B
Nível de Inserção Clínica	3,18 $\pm$ 0,73A	2 $\pm$ 1B	2,13 $\pm$ 0,74B
Altura da Gengiva Inserida	1,36 $\pm$ 0,49A	2,15 $\pm$ 0,50B	2,2 $\pm$ 0,67B
Grau de Sensibilidade	1,68 $\pm$ 1,08A	0,42 $\pm$ 0,50B	0,13 $\pm$ 0,35B
Índice se Sangramento	0,27 $\pm$ 0,45A	0,05 $\pm$ 0,22A	0,4 $\pm$ 0,63A
Espessura do Tecido	1,27 $\pm$ 0,55A	2,36 $\pm$ 0,49B	2,53 $\pm$ 0,74B

NOTA: Letras diferentes significam que são diferentes estatisticamente dentro de cada grupo de tratamento ($p < 0,05$).

A tabela 3, demonstra a comparação entre as variações das médias (inicial e 180 dias) de cada um dos parâmetros periodontais entre os grupos ECTS e A-PRF. Observou-se que não houve diferenças estatisticamente significantes entre os dois grupos, sendo que ocorreu um ganho de altura gengival de 1,40 $\pm$ 0,66mm no grupo tunelização + ECTS e 1,36 $\pm$ 0,72mm no grupo tunelização + A-PRF. Além disso, foi obtido um ganho na espessura do tecido nos dois grupos, sendo no grupo tunelização + ECTS -0,45 $\pm$ 1,40mm e 0,13 $\pm$ 1,45mm no grupo tunelização + A-PRF. A sensibilidade dentinária também obteve uma melhora significativa ($p < 0,05$) de 1,68 $\pm$ 1,08 para 0,13 $\pm$ 0,35 no grupo ECTS, e de 1,68 $\pm$ 0,99 para 0,4 $\pm$ 0,63.

Tabela 3 - Análise comparativa da variação (0-180dias) dos valores dos parâmetros periodontais entre os grupos ECTS e A-PRF. Os valores representam média $\pm$ desvio padrão.

Parâmetros (mm)	Tunelização + ECTS	Tunelização + A-PRF
Largura da Recessão	1,73 $\pm$ 0,78	1,63 $\pm$ 1,04
Altura da Recessão	1,40 $\pm$ 0,66	1,36 $\pm$ 0,72
Profundidade de Sondagem	0,98 $\pm$ 0,96	0,96 $\pm$ 0,80
Nível de Inserção Clínica	1,72 $\pm$ 1,12	1,72 $\pm$ 0,93
Altura da Gengiva Inserida	-0,13 $\pm$ 1,20	-0,27 $\pm$ 1,27
Espessura do Tecido	-0,45 $\pm$ 1,40	-0,13 $\pm$ 1,45

NOTA: Dados não significantes estatisticamente ($p > 0.05$).

Alguns pacientes relataram dor de maior intensidade na região doadora. Inchaço, edema e desconforto foram as complicações relatadas pelos pacientes deste estudo nos primeiros dias de pós-operatório, sendo que após a primeira semana, não foi relatado algum tipo de desconforto.

Todos os pacientes relataram que realizariam o procedimento novamente, caso fosse necessário, devido principalmente a melhora estética e melhora da sensibilidade dentinária apresentada inicialmente.

4- Discussão

Para o recobrimento das recessões gengivais, a cirurgia plástica periodontal é um desafio diário na prática clínica, pois muitas variações anatômicas devem ser levadas em consideração no seu tratamento, o que pode influenciar no seu prognóstico. A presença de osso interproximal, a espessura do tecido queratinizado, o tamanho das papilas adjacentes, o posicionamento dentário, bem como a destreza, habilidade e experiência do profissional, foram relatadas por Corteleni et al., 2018; Cortellini et al., 2009; e Tonetti e Jepsen, 2014.

A técnica de tunelização associada com o posicionamento coronal fornece extrema segurança com manipulação e tensão mínima do enxerto, proporcionando um excelente suprimento sanguíneo para os materiais de enxerto, garantindo assim, um menor desconforto pós-operatório, sendo esta técnica indicada na literatura para recessões gengivais múltiplas. (Zabalegui et al., 1999).

Os pacientes que apresentam recessões gengivais, com queixas relacionadas a hipersensibilidade dentinária e a estética, são candidatos a receberem os tratamentos de cirurgia plástica periodontal para recobrimento radicular. O recobrimento radicular em locais com perda de tecido gengival, seja ele localizado ou generalizado, é um dos objetivos das cirurgias plásticas periodontais. (Nassar et al., 2014)

O presente estudo mostrou uma melhora significativa nos parâmetros periodontais em ambos os grupos, após 6 meses de pós-operatório, estando estes resultados, de acordo com estudos feitos por Sato et al., 1993 e Cugini et al., 2000.

Apesar de o ECTS apresentar resultados melhores na maioria dos parâmetros periodontais, estes não foram significantes estatisticamente, demonstrando que as membranas de A-PRF podem ser indicadas para pacientes que não queiram se submeter ao procedimento de remoção do enxerto, e em casos em que o paciente não possua tecido gengival suficiente no local doador. Aliado a isso, a ausência de risco de complicações pós-operatórias como dor, necrose do palato, e acidente vascular da artéria palatina, são vantagens atribuídas para a utilização do A-PRF.

Várias modalidades de tratamento vêm sendo adicionada, sendo que diversos desenhos de retalhos, enxertos autógenos associados ou não com outros materiais, materiais sintéticos e até mesmo tratamento ortodôntico (Kassab et al.; 2010). A matriz de fibrina rica em plaquetas advanced (A-PRF), possui a capacidade de proteção de lesões abertas (Martins et al., 2017), e principalmente, de aumentar a cicatrização dos tecidos (Lebarbenchon et al., 2019), pelo poder de controlar o processo inflamatório, por meio da liberação gradativa de citocinas e de fatores de crescimento, estimulando a síntese de fibroblastos e de colágeno, fazendo também a

estimulação da angiogênese e da migração epitelial. (Martins, 2017; Macedo, 2019; Culhaoglu, 2018).

Em estudos anteriores com recessões bilaterais classe I e II de Miller, boca dividida, que avaliaram o uso do PRF para recobrimento radicular em comparação ao enxerto de tecido conjuntivo, relataram resultados sem diferença estatística no grau de recobrimento radicular entre os grupos. (Tunali 2015; Oncu 2017). Entretanto de acordo com revisão sistemática de Miron et al., 2017 os resultados do recobrimento radicular realizados com a técnica de Reposicionamento Coronário + enxerto de tecido conjuntivo, são maiores aos encontrados com a utilização de PRF. A quantidade de gengiva inserida foi estatisticamente maior nos dois grupos do presente estudo, quando comparado ao valor inicial, havendo diferença estatística entre os dois grupos com valores favoráveis ao grupo que utilizou enxerto conjuntivo. Esse resultado está de acordo com revisões sistemáticas anteriores (Moraschini 2016; Miron et al., 2017; Castro 2017).

Tunali 2015, em seu estudo, não apresentou diferença estatisticamente significativa na quantidade de gengiva inserida aos 12 meses. O aumento de gengiva inserida no grupo do enxerto conjuntivo se explica pelo fato do enxerto de tecido conjuntivo transferir sua expressão genética do local doador, aumentando a faixa de tecido queratinizado e a espessura de gengiva inserida na área receptora.

A faixa menor de tecido queratinizado no grupo do PRF pode ser explicada pelo fato de que a fibrina autóloga funciona pelo aumento da angiogênese, promovendo e induzindo a formação da neovascularização e novos tecidos no leito receptor. Para o procedimento de recobrimento radicular obter êxito a qualidade do leito receptor é fundamental, portanto, quando uma faixa de tecido queratinizado estiver disponível, ela promoverá a formação de mais tecido queratinizado. Entretanto, se houver apenas um tecido mucoso frouxo e não aderido, uma formação semelhante deste tecido com pouca qualidade será encontrada. (Dohan et al., 2006).

Os resultados positivos do PRF são concedidos a seus diversos fatores de crescimento (PDGF, VEGF E TGF), citocinas, glicoproteínas e principalmente a rede densa de matriz de fibrina (100x maior que a normal) que aumenta e promove a angiogênese e a síntese de matriz (Keceli et al., 2008, 2015). Neste estudo houve uma eficácia no recobrimento das recessões em ambos os grupos (tabelas 1 e 2), estando de acordo com outros estudos que encontraram resultados semelhantes como os de Oncu 2017 e Keceli et al., 2008.

Estudos recentes têm dado ênfase quanto a importância do biotipo tecidual para a estabilidade da margem gengival ao longo do tempo (Pini Prato et al., 2017). As técnicas que utilizam o enxerto de tecido conjuntivo são relatadas com maior ganho em espessura gengival (Baldi et al., 1999). No presente estudo, observou-se que nas situações em que se empregou o A-PRF não houve diferença estatisticamente significativa na espessura gengival.

Neste estudo observou-se uma melhora na altura de gengiva inserida nos dois grupos, porém com o A-PRF obteve um ganho maior, porém sem diferença estatística entre eles (aumento de $0,13 \pm 1,20$ contra $0,27 \pm 1,27$), resultado este, semelhante ao encontrado no estudo de Oncu et al., 2017.

A qualidade e quantidade de tecido mole alcançado após o recobrimento radicular com enxerto de membranas de A-PRF está diretamente relacionado com a quantidade de matriz de fibrina que é enxertada. Ghanaati et al 2014, descobriram que ao se usar 3 a 4 membranas de PRF, a fibropromoção poderia ser clinicamente previsível.

As vantagens do uso do L-PRF ou A-PRF são baseadas nas citocinas plaquetárias que desempenham um papel fundamental nos mecanismos iniciais do processo de cicatrização devido a sua capacidade de estimular a migração e a proliferação celular, induzir remodelação da matriz de fibrina e a secreção de uma matriz de colágeno. Essas citocinas ficam aprisionadas na malha de fibrina. O aumento do número de leucócitos desempenha um papel central na fagocitose de microorganismos e tecidos necróticos, bem como direcionando a futura regeneração desses tecidos através da liberação de citocinas e fatores de crescimento, além de estimular a angiogênese e formação dos tecidos (Ghanaati et al., 2014; Isobe et al., 2017; Kobayashi et al., 2016).

5- Conclusão

Com base na significância clínica dos resultados, foi possível concluir que tanto o ECTS quanto A-PRF, associado a técnica de tunelização, apresentaram melhora significativa nos parâmetros periodontais avaliados, não apresentando diferença estatística significativa entre eles, sugerindo que o A-PRF, associado a técnica de tunelização, pode ser utilizado como alternativa ao ECTS no tratamento de recessões gengivais tipo I de Cairo.

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ANEXOS

ANEXO A

ARTIGO CIENTÍFICO

Advanced Platelet-Rich Fibrin (A-PRF) Associated with the Tunneling Technique in the Treatment of Cairo Type 1 Gingival Recessions: Randomized and Blinded Clinical Study.

Edson Oliveira Silva¹, Leticia Nadal², Simone Karine Rothen³, Carlos Augusto Nassar⁴,
Patricia Oehlmeyer Nassar^{5*}

¹ Dentistry, Post-Graduate Student of the Master's Degree in Dentistry - Western Paraná State University - UNIOESTE - Cascavel, PR – Brazil – dredson.odontologia@hotmail.com

² Dentistry, Post-Graduate Student of the Master's Degree in Dentistry - Western Paraná State University - UNIOESTE - Cascavel, PR – Brazil – leticia_nadal@hotmail.com

³ Dentistry, Post-Graduate Student of the Master's Degree in Dentistry - Western Paraná State University - UNIOESTE - Cascavel, PR – Brazil – pro-simonekarine@hotmail.com

⁵ Periodontics, Western Paraná State University - UNIOESTE - Cascavel, PR – Brazil – canassar@yahoo.com

⁶ Periodontics, Western Paraná State University - UNIOESTE - Cascavel, PR – Brazil – ponassar@yahoo.com

* Mail Address: Avenida das Torres, 200 – casa 249 – FAG – CEP: 85806-095 – Cascavel – PR – A/C Patricia Oehlmeyer Nassar

Phone: (45) 99117 0903 - Email: ponassar@yahoo.com

Advanced platelet-rich fibrin (A-PRF) associated to tunnel technique in the treatment of Cairo type 1 gingival recession: Clinical, randomized and blind study.

ABSTRACT

The use of advanced platelet-rich fibrin (A- PRF) has presented good results in periodontal plastic surgery for treatment of gingival recessions. The present study aims at assessing the effectiveness of the use of A-PRF in root coverage and periodontal clinical parameters of patients with Cairo Type 1 gingival recession. Forty four (44) bilateral Cairo Type 1 gingival recessions were selected. The recessions were split in two groups according to the surgical procedure made (n=44): SCTG: Subepithelial Connective Tissue Graft and A-PRF, using tunnel surgical technique in both sides. The patients were monitored on 0, 90, and 180 days for assessment of the recession height and width; probing depth and bleeding on probing; clinical attachment level; keratinized tissue band width; thickness of keratinized gingival tissue and sensitivity level. The data obtained were initially analyzed and assessed with SHAPIRO-WILK test for normality and later ANOVA and TUKEY tests were used for normal data, or KRUSKALL-WALLIS and DUNN test for non-normal data. For analysis of sensitivity parameter, the test chosen was KRUSKALL-WALLIS and DUNN, with 5% significance level. In the analysis across groups the Student's *t*-test was conducted. Both groups presented improvement in periodontal parameters, however without significant statistical difference between them, which suggests that A-PRF can be an alternative to SCTG in root coverage surgeries of Cairo type 1 recession.

Keywords: platelet-rich fibrin; gingival recession; dentin hypersensitivity.

1. Introduction

Gingival recessions can cause esthetic damages to smile, in addition to bringing complications, like dentin hypersensitivity and cervical caries. Gingival recession is defined as the apical displacement of the gingival margin in relation to the cemento-enamel junction (CEJ) and is associated to the loss of attachment and exposure of the root surface to the oral environment. The gingival inflammation in response to the accumulation of dental biofilm, or mechanical trauma caused by tooth brushing, has been indicated as main etiological factor of these injuries (Bedoya and Park, 2014; Öncü, 2017). Biofilm accumulation, local trauma and absence of gingival attachment are etiological factors associated to this pathology (Cortellini et al., 2018).

Its etiological factors can be divided into: triggering factors and predisposing factors (Fu J et al., 2012). The triggering factors include tooth-brushing trauma, occlusal trauma, gingival inflammation, iatrogenic treatments and tabagism (Borghetti A., 2002). The predisposing factors, on the other hand, are essentially the anatomic characteristics of the patient that can influence the occurrence of gingival recessions, such as: gingiva biotype, variations in the underlying bone morphology, incorrect dental position and the presence of high attachment and short vestibule frena (Fu J et al., 2012).

Currently, the most used classification is that of Cairo, whose parameter is the loss of interproximal attachment tissue: RT1) soft tissue recessions that do not present loss of interproximal attachment; RT2) when there is loss of interproximal attachment, with distance from the cemento-enamel junction – CEJ in the bottom of sulcus shorter or equal to the vestibular attachment loss (measured from the CEJ to the bottom of vestibular sulcus); and RT3) Loss of interproximal attachment, with distance from CEJ to the sulcus bottom longer than the loss of vestibular attachment (Cairo et al., 2011; Steffens e Marcantonio, 2018).

Several surgical procedures were used for the treatment of gingival recessions in the last years, including the lateral repositioning of the flap, free gingival graft, tunneling surgery with and without subepithelial connective tissue (SCTG) graft, coronally positioned flap with or without SCTG, residual regeneration with membranes, acellular dermal matrix, platelet-rich plasma (PRP) and platelet-rich fibrin (PRF) (Aroca et al., 2009; Padma et al., 2013; Gupta et al., 2015; Tunali et al., 2015 Oncu et al., 2017).

The application of SCTG has been widely used in the treatment of gingival recessions, even presenting as its worse disadvantage the need of tissue from a donor area, which normally results in greater post-operative discomfort, and, in cases where several areas must be treated, a large amount of donor tissue is necessary. It is in this ambit that the L-PRF arises as

alternative, in terms of replacing the use of SCTG, because it demonstrates large amounts of growth factors, easiness to obtain, in addition to presenting a very favorable post-operative (Moraschini e Barboza, 2016).

The leukocyte- and platelet-rich fibrin (L – PRF) is a platelet aggregator obtained by blood centrifugation that started to be used in several tissue regeneration procedures in medicine and odontology after Choukroun studies in 2001. It is a framework of tridimensional fibrin with cells (leukocytes), growth factors, cytokines, and angiogenesis factors inside it, potentiating tissue repair and regeneration. In dentistry, it has been widely used in implants, bone reconstructions, and periodontal surgeries, as in the treatment of gingival recessions (Choukroun et al., 2016; Dohan et al., 2009; Aroca et al., 2009).

The A-PRF (*Advanced* PRF) effects have been reported for presenting a larger amount of cells (macrophages and monocytes) and larger amount of growth factors, due to its slower centrifugation, when compared to L-PRF (Kobayashi et al., 2016). The standard L-PRF was improved to A-PRF (*Advanced* – PRF) for having larger amounts of cells (macrophages and monocytes) and for supporting more release of growth factor in A-PRF (Ghanaati et al., 2014).

Normally, a shorter time of centrifugation increases the total number of cells contained in the PRF upper layer, producing a higher number of leukocytes “trapped” inside the fibrin matrix (Choukroun et al., 2006; Ghanaati et al., 2014; Kobayashi et al., 2016).

Thus, the objective of the present study was to assess the effectiveness of Advance platelet-rich fibrin (A-PRF) membranes associated to the tunnel technique for root coverage in patients with Cairo type 1 gingival recession for a period of 180 days.

2. Materials and Methods

The study conducted is of applied clinic nature, randomized, prospective with split-mouth and, parallel groups. The study was submitted and approved by Unioeste Committee of Ethics and Research on Human Beings (opinion number 3.359.508, CAAE 13806519.0.0000.0107, May 31, 2019). It was conducted at the Clinics of Unioeste Cascavel Campus Odontology Course.

Forty four recessions were selected, of patients from 23 to 47 years old, of both sexes, that presented bilateral Cairo type 1 gingival recessions, with probing depth inferior to 3mm in all teeth, without gingiva inflammation and free of caries (Figure 01). Patients with history of systemic disease, smokers, pregnant women or breastfeeding women and those with previous

surgeries in the recession location were excluded. The calculation of the sample size to the number of gingival recessions was made with t-test for independent samples with test power of 80% and alpha level of 0.05. These data were based on previous studies of the researchers group (Tonet et al., 2010; Nassar et al., 2014; Spada et al., 2017).

The initial clinical/periodontal examination was conducted by a single operator, previously calibrated that, with Willians n° 23 type periodontal probe, determined: 1. Recession height: measured as the distance from the cementoenamel junction to the gingival margin, in mm; 2. Recession width: mesio-distal distance of the gingival margin, measured in mm; 3. Probing depth: distance from the gingival margin (GM) to the bottom of the sulcus with presence or absence and measured in mm; 4. Bleeding on probing: at each three teeth with interval of approximately 30 seconds for record of the parameter, which corresponds to the probing time with record of presence or absence. 5. Clinical attachment level: distance from the cementoenamel junction (CEJ) to the bottom of the sulcus, with record of presence or absence, and measured in mm. Obtained from the sum of the gingival recession and the probing clinical depth; 6. Height of the keratinized tissue strip: the distance from the mucogingival junction to the gingival margin; 7. Thickness of the keratinized gingival tissue at three points (Mesial, central and distal); 8. Degree of sensitivity (U.S. Public Health Service modified index comprising three scores: 0 (Alpha) – no sensitivity; 2 (Bravo) – slight sensitivity; 3 (Charlie) – high sensitivity (Camilotti et al., 2012).

After the initial periodontal clinical examination, the recessions were randomly divided into two groups (Random Group Generator – Web Site - <https://pt.rakko.tools/tools/59/>), according to the treatments proposed in Figure 1. Each patient received both surgical techniques, in one hemiarch, tunneling associated to subepithelial connective tissue (Tunneling + SCTG) and in the other hemiarch, tunneling associated to A-PRF membranes (Tunneling + A-PRF), and the distribution to hemiarchs was randomly made.

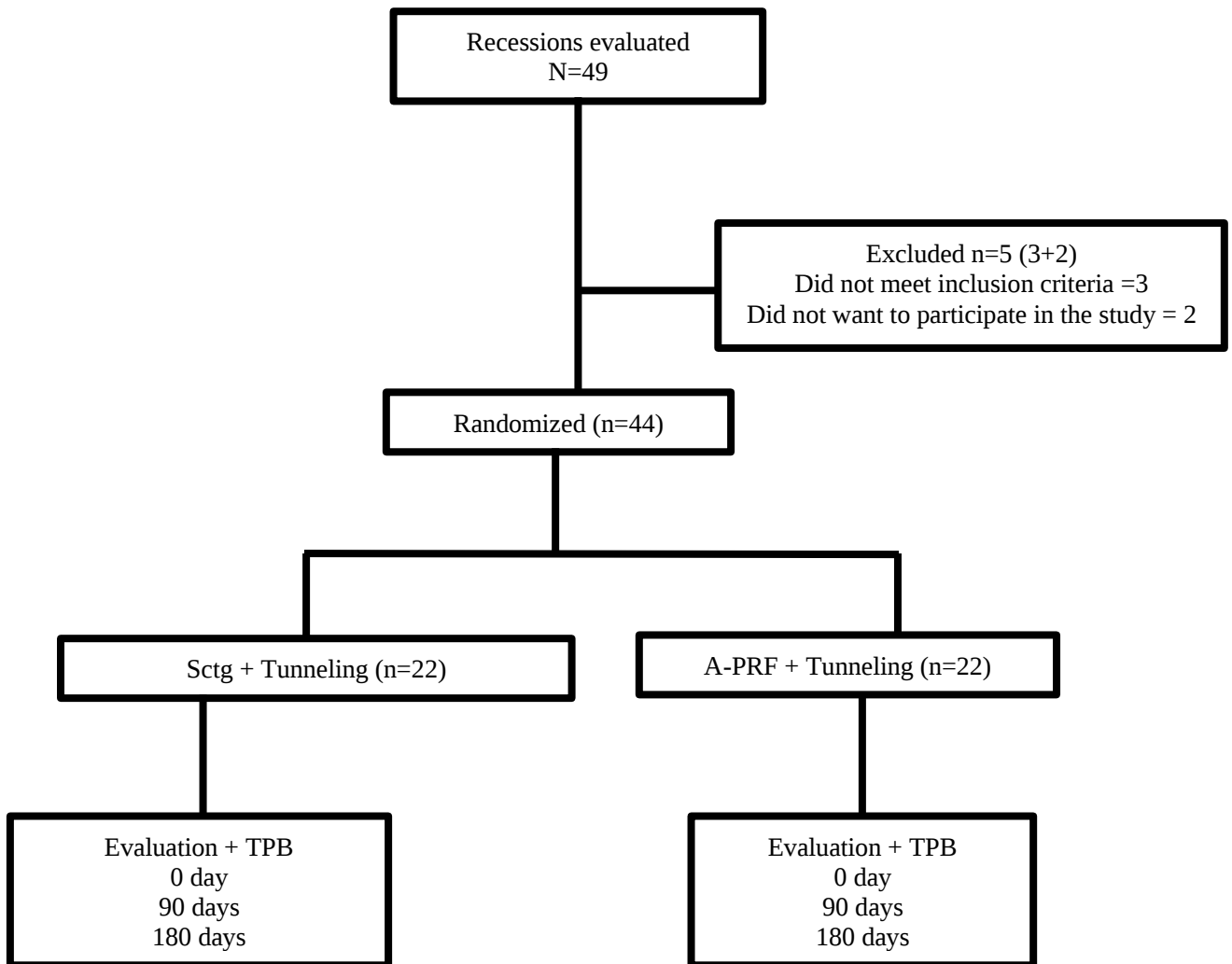


Figure 1. Distribution of the 44 recessions according to the treatments proposed.

2.1 Surgical techniques

2.1.1 Recipient bed preparation:

The two surgical techniques were performed in one single surgical act, and so, the bilateral receptor areas were simultaneously prepared. The receptor area was anesthetized with infiltrative terminal anesthesia, with anesthetic articaine 4% and epinephrine 1:100.000 (*DFL, Rio de Janeiro, RJ, Brazil*). Intra-sulcus incision was performed in the cervical region, at CEJ level, for mesial and distal with 15c blade (*Swann Morton, England*), creation of tunnel with tunnelers (*Supremo Instrumentais Cirúrgicos, São Paulo, Brazil*), by means of a flap with total thickness close to the gingival margin, until going beyond the mucogingival junction, so as to obtain a tissue without tensions. The tooth root was scraped and smoothed with periodontal curette, Gracey type, specific to the region, followed by abundant irrigation with saline solution 0.9%.

2.1.2 Obtention of the connective tissue graft and adaptation to the recipient bed (Tunneling + SCTG Group)

The subepithelial connective tissue graft was obtained from the palate, through single incision technique (linear incision) (Xavier e Alves, 2015). The donor region was sutured with simple sutures using Nylon 5.0 prolene (*SHALON® São Luis M. Belos, Goiás, Brazil*), then it was positioned on the gingival recession and stabilized in proximals with propylene thread 5.0 (*TECHSUTURE® , Bauru, São Paulo, Brazil*) at CEJ level. It was later coronally tractioned with vertical sutures fixed in the incision/vestibular region of the dental elements involved, using polypropylene thread 5.0 (*TECHSUTURE® , Bauru, São Paulo, Brazil*).

2.1.3 Preparation of Advanced Platelet-rich Fibrin and adaptation in the recipient bed (Tunneling + A-PRF Group)

Protocol for obtention and use of A-PRF (Choukroun and Ghanaati et al., 2018): 1st Stage: Venipuncture and blood collection; 2nd Stage: Cell separation (centrifugation); 3rd Stage: Preparation of A-PRF membranes. 1st Stage: Venipuncture and blood collection: Before starting the surgical procedure for root coverage, venipuncture of around 60 ml of blood was performed (according to the extension of the recipient bed). 2nd Stage: Cell separation (centrifugation): The blood collected was immediately placed in the centrifuge (*Centrifuga Spinlab, São Paulo, Brazil*), not exceeding 3 minutes, in 10 ml portions. The fibrin membranes were obtained by 1,500 rpm centrifugation for 14 minutes. When the centrifugation was finished, the tubes were collected, and the fibrin was not immediately removed. At least 30 minutes passed before its use in the surgical bed. 3rd Stage: Preparation of A-PRF membranes: With the appropriate stainless-steel kit for making PRF membranes (*Supremo Instrumentais Cirúrgicos, São Paulo, Brazil*) the intermediary portion centrifuged, the fibrin clot, was separated from the portion of red cells and platelet-poor plasma and deposited in the stainless steel box. The compressive lid was applied, without tightening it, for 15 minutes. The weight itself (130g) is enough to compress the clot to obtain the membranes, without posing damages to the cell structures present in the fibrin mesh. Four A-PRF membranes were used, and they were positioned and sutured inside the mucoperiosteal tunnel created. The tissue was later coronally tractioned with vertical sutures fixed in the incision/vestibular region of the dental elements involved, using polypropylene thread 5.0 (*TECHSUTURE, Bauru, São Paulo, Brazil*). (Figure 4)

2.2 Post-operative control and evaluations

The patients received pain relief medications, antibiotic therapy, non steroidal anti-inflammatory, analgesic and mouthwash (Amoxicillin 500mg at each 8 hours, for seven days, nimesulide 100mg at each 12 hours for 3 days, dipyron 500mg/5ml for 3 days and chlorhexidine digluconate 0.12% at each 12 hours for 15 days). The donor region sutures were removed after 7 days, and the recipient bed sutures were removed after 15 days.

The patients were assessed for a period of 180 days, and the clinical examinations were made in periods of 0, 90 and 180 days, and in all periods the patients received maintenance therapy, with all criteria of the initial periodontal examination reassessed. After the 180 days of experimental period, all patients were included in a periodontal maintenance program.

2.3 Statistical analysis

The program used for statistical analysis was GraphPad Prism 8.0®. The clinical data were initially analyzed and assessed with SHAPIRO-WILK test for normality verification, ANOVA and TUKEY tests for normal data and KRUSKALL-WALLIS and DUNN tests for non-normal data. For analysis of sensitivity parameter and bleeding KRUSKALL-WALLIS and DUNN tests were used. In the analysis between groups the STUDENT t-test was used, with significance level of 5%.

3. Results

Forty four gingival recessions were analyzed, 22 from the tunneling + SCTG group and 22 from the tunneling +A-PRF group.

Table 01 displays the values of periodontal clinical parameters for the tunneling + A-PRF group analyzed after 180 days, with all parameters presenting statistically significant improvements after the 180 days of follow-up ($p<0.05$).

Table 1 – Comparative analysis of periodontal parameters of the Tunneling + A-PRF group in the initial and final periods. The values represent mean $\pm$ standard deviation.

Parameters (mm)	Tunneling + A-PRF (initial)	Tunneling + A-PRF (90 days)	Tunneling + A-PRF (180 days)
Recession width	2.68 $\pm$ 0.64A	1.63 $\pm$ 0.95B	1.53 $\pm$ 0.63B
Recession height	2.13 $\pm$ 0.83A	1.05 $\pm$ 0.62B	1.13 $\pm$ 0.51B
Probing depth	1.71 $\pm$ 0.38A	1.35 $\pm$ 0.37B	1.08 $\pm$ 0.26B
Clinical attachment level	3.18 $\pm$ 0.85A	2.21 $\pm$ 0.71B	2.13 $\pm$ 0.51B
Height of attached gingiva	1.37 $\pm$ 0.47A	2.21 $\pm$ 0.53B	2.33 $\pm$ 0.81B
Degree of sensitivity	1.68 $\pm$ 0.99A	0.68 $\pm$ 0.94B	0.4 $\pm$ 0.63B
Bleeding rate	0.31 $\pm$ 0.64A	0 $\pm$ 0A	0.6 $\pm$ 0.50B
Tissue thickness	1.40 $\pm$ 0.50A	2.42 $\pm$ 0.76B	2.26 $\pm$ 0.70B

NOTE: Different letters mean that they are statistically different in each treatment group ($p<0.05$).

Table 02 presents the values of periodontal clinical parameters for the tunneling + SCTG group analyzed after 180 days, where all periodontal parameters presented statistically significant improvement, except for bleeding rate.

Table 2 - Comparative analysis of periodontal parameters of the Tunneling + SCTG group in the initial and final periods. The values represent mean $\pm$ standard deviation.

Parameters (mm)	Tunneling + SCTG (initial)	Tunneling + SCTG (90 days)	Tunneling + SCTG (180 days)
Recession width	2.5 $\pm$ 0.51A	1.73 $\pm$ 0.73B	1.26 $\pm$ 0.59B
Recession height	2.04 $\pm$ 0.57A	0.89 $\pm$ 0.56B	0.93 $\pm$ 0.45B
Probing depth	0.71 $\pm$ 0.52A	1.24 $\pm$ 0.39B	1.06 $\pm$ 0.13B
Clinical attachment level	3.18 $\pm$ 0.73A	2 $\pm$ 1B	2.13 $\pm$ 0.74B
Height of attached gingiva	1.36 $\pm$ 0.49A	2.15 $\pm$ 0.50B	2.2 $\pm$ 0.67B
Degree of sensitivity	1.68 $\pm$ 1.08A	0.42 $\pm$ 0.50B	0.13 $\pm$ 0.35B
Clinical attachment level	0.27 $\pm$ 0.45A	0.05 $\pm$ 0.22A	0.4 $\pm$ 0.63A
Tissue thickness	1.27 $\pm$ 0.55A	2.36 $\pm$ 0.49B	2.53 $\pm$ 0.74B

NOTE: Different letters mean that they are statistically different in each treatment group ($p < 0.05$).

Table 03 presents the comparison of the means variation (initial and after 180 days) of each periodontal parameters among SCTG and A-PRF groups. No statistically significant differences were observed, and there was gain of 1.40 ± 0.66 mm in gingival height in tunneling + SCTG group and 1.36 ± 0.72 mm in tunneling + A-PRF group. Besides, gain in the tissue thickness was obtained for both groups: 0.45 ± 1.40 mm for the tunneling + SCTG group, and 0.13 ± 1.45 mm for the tunneling + A-PRF group. Dentin sensitivity also presented significant improvement ($p < 0.05$) from 1.68 ± 1.08 to 0.13 ± 0.35 in the SCTG group, and from 1.68 ± 0.99 to 0.4 ± 0.63 for the A-PRF group.

Table 3 - Comparative analysis of the variation (0 – 180 days) of periodontal parameters in SCTG and A-PRF groups. The values represent mean $\pm$ standard deviation.

Parameters (mm)	Tunneling + SCTG	Tunneling + A-PRF
Recession width	1.73 ± 0.78	1.63 ± 1.04
Recession height	1.40 ± 0.66	1.36 ± 0.72
Probing depth	0.98 ± 0.96	0.96 ± 0.80
Clinical Attachment Level	1.72 ± 1.12	1.72 ± 0.93
Height of attached gingiva	-0.13 ± 1.20	-0.27 ± 1.27
Tissue thickness	-0.45 ± 1.40	-0.13 ± 1.45

NOTE: Data not statistically significant ($p > 0.05$).

Some patients reported more intense pain in the donor region. Swelling and discomfort were the complications reported by the patients in this study in the first post-operative days, while after the first week no type of discomfort was reported.

All patients stated that they would undergo the procedure again, if necessary, chiefly due to the improvement in esthetic and in the initially presented dentin sensitivity.

4. Discussion

For gingival recession coverage, the gingival plastic surgery is a daily challenge in the clinical practice, because many anatomic variations must be considered in their treatment, which can influence their prognosis. The presence of interproximal bone, the thickness of the keratinized tissue, the size of adjacent papillae, the teeth positioning, as well as the dexterity, skills and experience of the professional were reported by Corteleni et al., 2018; Cortellini et al., 2009; and Tonetti e Jepsen, 2014.

The tunneling technique associated to the coronal positioning provides high safety with minimum handling and tension in the graft, thus ensuring less post-operative discomfort. This technique is indicated for multiple gingival recessions in the literature (Zabalegui et al., 1999).

Patients that present gingival recessions with complaints associated to dentin hypersensitivity and esthetics are eligible to receive periodontal plastic surgery for root coverage. Root coverage in locations with loss of gingival tissue, whether localized or generalized, is one of the objectives of periodontal plastic surgeries (Nassar et al., 2014).

The present study showed significant improvement in periodontal parameters in both groups after 6 months of post-operative, and such results are aligned with those of studies conducted by Sato et al., 1993 and Cugini et al., 2000.

Though SCTG presents better results in most periodontal parameters, they were not statistically significant, which demonstrates that A-PRF membranes can be indicated for patients that don't want to undergo the procedure for removal of the graft; and in cases where the patient does not have enough gingival tissue at the donor location. Moreover, the absence of risk of post-operative complications like pain, palate necrosis, and vascular accident of the palatine artery, is an advantage assigned to the use of A-PRF.

Several treatment modalities are being added, like different flap designs, autogenous grafts associated or not to other materials, synthetic materials, and even orthodontic treatment (Kassab et al.; 2010). The advanced platelet-rich fibrin (A-PRF) matrix can protect open injuries (Martins et al., 2017), and, mainly, it can increase tissues cicatrization (Lebarbenchon et al., 2019), because it can control the inflammatory process by gradual release of cytokines and growth factors, thus stimulating the synthesis of fibroblasts and collagen, also promoting angiogenesis and epithelial migration (Martins, 2017; Macedo, 2019; Culhaoglu, 2018).

Previous studies on Miller class I and II bilateral recessions, split mouth, that assessed the use of PRF for root coverage compared to connective tissue graft reported results without statistical differences in the level of root coverage across groups (Jandovic 2012; Tunali 2015; Oncu 2017). However, according to Miron et al. systematic review, 2017, the results of root coverage performed

with Coronary Repositioning + connective tissue graft are better than those found with the use of PRF. The amount of gingiva attached was statistically higher in both groups of the present study when compared to the initial value, with statistical differences between the two groups, and with values favorable to the group that used connective graft. Such result corroborates previous systematic reviews (Moraschini 2016; Miron et al., 2017; Castro 2017).

Tunali 2015, in his study, did not observe statistically significant differences in the amount of gingiva attached after 12 months. The increase of gingiva attached in the connective graft group is explained by the fact that the connective tissue graft transfers its genetic expression from the donor location, increasing the strip of keratinized tissue and the thickness of the gingiva attached to the recipient area.

The shorter strip of keratinized tissue in the PRF group can be explained by the fact that the autologous fibrin works with angiogenesis increase, promoting and inducing the formation of neovascularization and new tissues in the recipient bed. For the root coverage procedure to be successful, the quality of the recipient bed is fundamental, therefore, when a strip of keratinized tissue is available, it will promote the formation of more keratinized tissues. However, in case there is only a loose mucous tissue, not adhered, a similar formation of this tissue, with poor quality, will be found (Dohan et al., 2006).

The positive PRF results are granted to the different growth factors (PDGF, VEGF E TFG), cytokines, glycoproteins, and chiefly the dense network of fibrin matrix (100x greater than normal) that increases and promotes angiogenesis and matrix synthesis (Keceli et al., 2008, 2015). In this study there was efficacy in recession coverage for both groups (tables 1 and 2), which is aligned to other studies that found similar results, like those by Oncu 2017 and Keceli et al., 2008.

Recent studies have emphasized the tissue biotype importance for the gingival margin stability over time (Pini Prato et al., 2017). Techniques that utilize connective tissue graft are reported with more gain in gingival thickness (Baldi et al., 1999). In the present study, it was observed that in situations where A-PRF was used there was no statistically significant difference in gingival thickness.

Improvement in the height of the attached gingiva was observed in both groups, however, the A-PRF group obtained greater gain, though without statistical differences between them (increase of 0.13 ± 1.20 against 0.27 ± 1.27), result that is similar to that found in the study by Oncu et al., 2017.

The quality and amount of soft tissue reached after the root coverage with PRF membrane graft is directly associated to the amount of fibrin matrix grafted. Ghanaati et al 2014 found out that while using 3 to 4 PRF membranes, fibropromotion could be clinically predictable.

The advantages of using L-PRF or A-PRF are based on the platelet cytokines that play a fundamental role in the initial mechanisms of the cicatrization process, due to their capacity to stimulate cell migration and proliferation, induce remodeling of the fibrin matrix and the secretion of a collagen matrix. These cytokines stay trapped in the fibrin mesh. The increase in the number of leukocytes plays a central role in the phagocytosis of microorganisms and necrotic tissues, and also directs the future regeneration of these tissues through the release of cytokines and growth factors, in addition to stimulating angiogenesis and the formation of tissues (Ghanaati et al., 2014; Isobe et al., 2017; Kobayashi et al., 2016).

5. Conclusion

Based on the clinical significance of results, it was possible to conclude that both SCTG and A-PRF, associated to the tunneling technique, presented significant improvement in the periodontal parameters assessed, without presenting significant statistical difference between them, suggesting that A-PRF associated to the tunneling technique, can be used as alternative to SCTG in the treatment of Cairo type I gingival recessions.

Declaration of Conflict of Interests

The authors declare that there is no conflict of interests.

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 2. Two authors: both authors' names and the year of publication;
 3. Three or more authors: first author's name followed by "et al." and the year of publication.
- Citations may be made directly (or parenthetically). Groups of references should be listed first alphabetically, then chronologically. Examples: "as demonstrated (Allan, 1996a, 1996b, 1999;

Allan and Jones, 1995). Kramer et al. (2000) have recently shown" List: References should be arranged first alphabetically and then further sorted chronologically if necessary. More than one reference from the same author(s) in the same year must be identified by the letters "a", "b", "c", etc., placed after the year of publication. Examples: Reference to a journal publication: Van der Geer, J., Hanraads, J.A.J., Lupton, R.A., 2000. The art of writing a scientific article. *J. Sci. Commun.* 163, 51-59. Reference to a book: Strunk Jr., W., White, E.B., 1979. *The Elements of Style*, third ed. Macmillan, New York. Reference to a chapter in an edited book: Mettam, G.R., Adams, L.B., 1999. How to prepare an electronic version of your article, in: Jones, B.S., Smith, R.Z. (Eds.), *Introduction to the Electronic Age*. E-Publishing Inc., New York, pp. 281-304. Web references: As a minimum, the full URL should be given. Any further information, if known (DOI, author names, dates, reference to a source publication, etc.), should also be given. Web references can be listed separately (e.g., after the reference list) under a different heading if desired, or can be included in the reference list.

Journal abbreviations source

Journal names should be abbreviated according to the List of Title Word Abbreviations.

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AFTER ACCEPTANCE

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ANEXO C**PARECER CONSUBSTANCIADO DO CEP**

**UNIOESTE - CENTRO DE
CIÊNCIAS BIOLÓGICAS E DA
SAÚDE DA UNIVERSIDADE**



PARECER CONSUBSTANCIADO DO CEP

DADOS DO PROJETO DE PESQUISA

Título da Pesquisa: FIBRINA RICA EM PLAQUETAS (PRF) NO RECOBRIMENTO DE RAÍZES COM RECESSÃO TIPO 1 DE CAIRO E RESTAURADAS COM CIMENTO DE IONÔMERO DE VIDRO MODIFICADO POR RESINA.

Pesquisador: Carlos Augusto Nassar

Área Temática:

Versão: 1

CAAE: 13806519.0.0000.0107

Instituição Proponente: Centro de Ciências Biológicas e da Saúde CCBIS - UNIOESTE

Patrocinador Principal: Financiamento Próprio

DADOS DO PARECER

Número do Parecer: 3.359.508

Apresentação do Projeto:

Estudo clínico randomizado do tipo boca dividida em que deverão ser incluídos 40 pacientes com recessão gengival Tipo 1 de Cairo bilaterais que buscam tratamento da Clínica de Odontologia da Unioeste, com idade entre 20 e 60 anos, sem comorbidades sistêmicas que impossibilitem a realização de procedimentos cirúrgicos. Os pacientes serão submetidos a cirurgia periodontal de recobrimentos radicular associada a enxerto de tecido conjuntivo subepitelial - ETCS de um lado da arcada dentária e do outro membranas de fibrina rica em plaquetas – PRF, sendo realizadas as técnicas de Túnel e Retalho Reposicionado Coronariamente. Os pacientes serão acompanhados por 90 e 180 dias, para verificação da altura e largura da recessão; Profundidade de sondagem e Sangramento à Sondagem; Nível de inserção clínica; Nível Gengival (Recessão); Largura da faixa de tecido queratinizado; Espessura do tecido gengival queratinizado em 3 pontos (Mesial, central e distal); Grau de sensibilidade (Índice modificado da U.S. Public Health Service composto de três escores: 0 (Alpha) - sem sensibilidade; 2 (Bravo) - pequena sensibilidade; 3 (Charlie) - alta sensibilidade. Para análise será utilizado o teste de SHAPIRO-WILK para verificação da distribuição da normalidade; depois, serão utilizados o teste de ANOVA e TUKEY para verificar diferença entre os grupos. Para análise do parâmetro de sensibilidade, serão utilizados os testes de KRUSKAL-WALLIS e DUNN, com nível de significância de 5%. O programa Bioestat 5.4 (Instituto Mamirauá, Amazonas, Brasil) será utilizado para a realização de toda a análise estatística do estudo.

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Bairro: UNIVERSITÁRIO

CEP: 85.815-110

UF: PR

Município: CASCAVEL

Telefone: (45)3220-3092

E-mail: cep-propg@unioeste.br

**UNIOESTE - CENTRO DE
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Continuação do Parecer: 3.358.500

Objetivo da Pesquisa:

Comparar os resultados do recobrimento radicular utilizando-se o ETO3 ou PRF, associados com Retalho Posicionado Coronalmente (RPC) ou a Técnica do Tunelamento (Tunel) em raízes restauradas com CIV-R.

Objetivo Secundário: * Conhecer os efeitos do PRF em recobrimento radicular utilizando duas técnicas cirúrgicas distintas, RPC e Tunel; * Analisar a morbidade pós-operatória em ambas as técnicas cirúrgicas; * Verificar se há melhora da sensibilidade após a realização do recobrimento radicular; * Averiguar clinicamente sobre a estabilidade dos resultados em ambas as técnicas e ambos os materiais de enxerto, em 0, 90 dias e 180 dias.

Avaliação dos Riscos e Benefícios:

Riscos: Estão relacionados à utilização de anestésicos locais, de rotina no atendimento odontológico, dor, desconforto, necroses gengivais (tanto do enxerto como do leito receptor) e sangramento pós-operatório. Em relação aos riscos dos anestésicos, estes podem ser diminuídos pela avaliação de episódios anteriores de reações alérgicas ou alterações na pressão arterial e, caso sejam relatadas alterações de qualquer natureza, o paciente será encaminhado para avaliação médica e somente poderá participar do estudo quando houver autorização do profissional. Com relação aos demais riscos, para que os mesmos sejam minimizados, os pacientes deverão seguir as recomendações pós-operatórias, como por exemplo a realização de compressas de gelo no local, repouso e alimentação leve.

Benefícios: Os benefícios esperados são de que se estabeleça uma melhora dos parâmetros clínicos periodontais das áreas com recessões gengivais de todos os pacientes, bem como, o recobrimento radicular, formação de um bom volume de tecido queratinizado, melhora da sensibilidade e do contorno estético da margem gengival, além da não necessidade de remoção de enxertos autógenos no grupo teste.

Comentários e Considerações sobre a Pesquisa:

Pesquisa apresenta relevância científica e uma revisão breve do tema.

Considerações sobre os Termos de apresentação obrigatória:

Apresentou todos os termos obrigatórios.

Endereço: RUA UNIVERSITÁRIA 2089	CEP: 85.819-110
Bairro: UNIVERSITÁRIO	
UF: PR	Município: CASCAVEL
Telefone: (45)3220-3032	E-mail: cep.prpq@unioeste.br

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Continuação do Parecer: 3.359-536

Conclusões ou Pendências e Lista de Inadequações:

A pesquisa atende os preceitos éticos da pesquisa que envolve seres humanos.

Este parecer foi elaborado baseado nos documentos abaixo relacionados:

Tipo Documento	Arquivo	Postagem	Autor	Situação
Informações Básicas do Projeto	PE_INFORMAÇÕES_BÁSICAS_DO_PROJETO_1333453.pdf	24/04/2019 10:59:43		Aceito
TCLE / Termos de Assentimento / Justificativa de Ausência	termodeconsentimentolivreesclarecido.pdf	20/04/2019 10:54:37	Carlos Augusto Nassar	Aceito
Outros	termodecompromissousodedados.pdf	11/04/2019 20:23:23	Carlos Augusto Nassar	Aceito
Declaração de Pesquisadores	declaracao.pdf	11/04/2019 20:22:57	Carlos Augusto Nassar	Aceito
Projeto Detalhado / Brochura Investigador	Projetocomite.pdf	11/04/2019 16:28:35	Carlos Augusto Nassar	Aceito
Outros	termocienciadoresponsavel.pdf	11/04/2019 16:28:25	Carlos Augusto Nassar	Aceito
Outros	instrumentocoletadedados.pdf	11/04/2019 16:27:51	Carlos Augusto Nassar	Aceito
Parecer Anterior	parecerambulatorioodontio.pdf	11/04/2019 16:27:34	Carlos Augusto Nassar	Aceito
Folha de Rosto	folhaderosto.pdf	11/04/2019 16:27:22	Carlos Augusto Nassar	Aceito

Situação do Parecer:

Aprovado

Necessita Apreciação da CONEP:

Não

CASCAVEL, 31 de Maio de 2019

Assinado por:
Dartel Ferrari de Lima
(Coordenador(a))

Endereço: RUA UNIVERSITÁRIA 2009
Bairro: UNIVERSITÁRIO
UF: PR Município: CASCAVEL
Telefone: (45)3220-3002 CEP: 85.819-110
E-mail: cep.prppq@unioeste.br