

Original Research Paper

# Comparison Between the Use of Advanced Platelet-Rich Fibrin (A-PRF) and Connective Tissue Graft in the Treatment of Cairo Type 1 Gingival Recession Associated with the Coronally Advanced Flap

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**Abstract:** The effects of Advanced Platelet Rich Fibrin (A-PRF) are promising in periodontal surgeries because of the high number of cells and growth factors due to slower centrifugation. Then, the aim of the present study was to evaluate the efficacy of A-PRF in combination with Coronally Advanced (CAF) and compare it with the use of Connective Tissue Graft (CTG) also associated with CAF in Cairo type 1 gingival recession. The study was approved by the Human Research Ethics Committee of the University and patients signed the informed consent. Forty gingival recessions were selected, with 20 belonging to the CAF + A-PRF group (test group) and 20 to the CAF + CTG group. The height of the gingival recession, height, and thickness of the attached gingiva, probing depth, level of clinical insertion, and tooth sensitivity were evaluated at the beginning and 6 months after surgery. GraphPad Prism 8.0 program® was used for statistical analysis. The normality was assessed through Shapiro-Wilk. T-student was performed for the parametric data and MANN-WHITNEY test for the non-parametric. The significance level was set at 5%. There was a gain in gingival height of  $1.45 \pm 0.96$  mm in the CAF + CTG group and  $1.04 \pm 0.95$  mm in the CAF + A-PRF group, resulting in a coverage of 54.3% in the test group and 73% in the control group. Complete coverage (100%) of the recessions was found at 7 sites (35%) in the group with A-PRF and 10 sites (50%) with the CTG. In both groups, there was a significant improvement in tooth sensitivity ( $p < 0.05$ ). Regarding the other clinical parameters evaluated, there was no statistically significant difference; however, slightly higher rates of gum gain were observed in both height and thickness in the control group. After 180 days, both techniques showed significant improvement in the evaluation + ed periodontal parameters, concluding that both can be used for treating Cairo type I gingival recessions.

**Keywords:** Gingival Recession, Surgical Flap, Platelet-Rich Fibrin, Connective Tissue

## Introduction

Affecting approximately 84.6% of the population (Sarfati *et al.*, 2010), gingival recessions are a common concern related to smile disharmony, often accompanied by complications such as dentin hypersensitivity and cervical caries. Gingival Recession (GR) is defined as the exposure of the root surface due to the migration of the gingival margin, with the gingiva falling short of the cement junction

(Fernandes *et al.*, 2021; Mancini *et al.*, 2021). It can manifest as localized or generalized and may be associated with one or more surfaces (Kassab *et al.*, 2010; Öncü, 2017). Etiological factors most commonly linked to this condition include biofilm accumulation, local trauma, and local anatomy, such as alveolar bone dehiscence and a thin gingival biotype (Öncü, 2017).

Initially, the Miller classification (class I, II, III, and IV) was employed to categorize gingival recessions. However, in 2018, a consensus was reached between the American

Academy of Periodontology and the European Federation of Periodontology during a world workshop for the classification of periodontal and peri-implant diseases and conditions. The Cairo classification, based on the loss of interproximal insertion tissue, was introduced, comprising RT1 soft tissue recessions without interproximal insertion loss; RT2 cases with the loss of interproximal attachment, with the distance from the Cemento-Enamel Junction (CEJ) to the sulcus/pouch being less than or equal to the vestibular attachment loss (measured from the CEJ to the sulcus/pouch in the buccal socket); and RT3 interproximal attachment loss, where the distance from the CEJ to the sulcus/pouch base exceeds the vestibular attachment loss (Bin *et al.*, 2023; Cairo *et al.*, 2014; Nassar *et al.*, 2022).

Indications for the treatment of gingival recessions include reduced sensitivity, a decreased risk of cervical and root caries, increased gingival banding, and improved aesthetics (Imber and Kasaj, 2021; Kassab *et al.*, 2010; Mancini *et al.*, 2021). Surgical therapeutic options are well-documented and have high success rates, achieving up to 100% coverage in cases of RT1 (Bin *et al.*, 2023; Chan *et al.*, 2015; Nassar *et al.*, 2022).

In recent years, various surgical procedures have been employed to address gingival recession defects, such as lateral flap repositioning, free gingival grafting, Coronally Advanced Flap (CAF), subepithelial Connective Tissue Grafting (CTG) and membrane-guided tissue regeneration, acellular dermal matrix, Platelet-Rich Plasma (PRP) and Platelet-Rich Fibrin (PRF) (Aroca *et al.*, 2009; Eren and Atilla, 2014; Fernandes *et al.*, 2021; Padma *et al.*, 2013; Uzun *et al.*, 2018).

CAF associated with CTG is considered the gold standard in the treatment of gingival recessions (Kassab *et al.*, 2010; Mancini *et al.*, 2021; Moraschini and Barboza, 2016; Öncü, 2017; Pini Prato *et al.*, 2018; Rodas *et al.*, 2020). However, it has some disadvantages, including greater morbidity, the need for another surgical site, the risk of hemorrhage due to the palatine artery, and limited donor tissue supply. Thus, materials with fiber conduction potential, such as platelet aggregates, are being studied as potential substitutes for these grafts (Dohan *et al.*, 2006; Fernandes *et al.*, 2021).

Platelet and Leukocyte Rich Fibrin-L-PRF is a platelet aggregate obtained through blood centrifugation, first utilized in various tissue regeneration procedures in medicine and dentistry following Choukroun's studies in 2001. It serves as a three-dimensional fibrin scaffold containing cells (leukocytes), growth factors, cytokines, and angiogenic factors, enhancing tissue repair and regeneration. In dentistry, it has found applications in implantology and periodontal reconstructive surgeries, particularly in the treatment of gingival recessions (Choukroun *et al.*, 2006;

Dohan *et al.*, 2006; El Bagdadi *et al.*, 2019). Currently, the effects of Advanced PRF (A-PRF) are under speculation, reported to have a higher number of cells and growth factors than L-PRF due to slower centrifugation (El Bagdadi *et al.*, 2019; Ghanaati *et al.*, 2014; Kobayashi *et al.*, 2016). It has been promising as an alternative to the CTG.

The objective of the present study was to evaluate and compare the results of the level of insertion, height, and thickness of the attached gingiva and the degree of sensitivity after root covering of Cairo type 1 gingival recessions using the coronally advanced flap technique associated with subepithelial connective tissue grafting versus the advanced platelet-rich fibrin membrane.

## Materials and Methods

This is an applied clinical study, designed as a prospective, split-mouth, parallel-group, and randomized trial. The project underwent submission and approval by the human research ethics committee of Unioeste, with the identification number 3.359.508, CAAE 13806519.0.0000.0107.

The study involved the selection of 12 patients, ranging in age from 23-47 years, comprising both genders. These individuals presented bilateral Cairo type 1 gingival recession, with probing depth of less than 3 mm in all teeth, no signs of gingival inflammation, and were free of caries (Fig. 1). Exclusion criteria included individuals with a history of systemic disease, smokers, pregnant or lactating women and those who had undergone previous surgeries at the recession site. The initial screening involved 48 recessions in 12 patients and the sample size was determined based on previous analyses, considering the number of GR with at least two teeth presenting GR in each side. Test power was set at 80%, alpha level at 0.05 and the data were derived from prior studies by the group of researchers (Bin *et al.*, 2023; Nassar *et al.*, 2014; Bombardelli *et al.*, 2010).



**Fig. 1:** The gingival recessions selected were bilateral Cairo 1 recessions

The initial clinical and periodontal examination was conducted by a previously calibrated researcher who utilized a Williams Type 23 periodontal probe. The following parameters were determined:

- Height of the recession (measured at the center of the recession from the amelocemental junction to the gingival margin)
- Clinical attachment level (the result of probing depth added to the height of the gingival recession)
- Height of the attached gingiva (measurement from the gingival margin to the mucogingival junction)
- Thickness of keratinized gingival tissue at 3 points (mesial, central, and distal, performed with tissue perforation and periodontal probe)
- Recession width
- Height of clinical crown
- Probing depth
- Degree of sensitivity (pain score on a scale of 0-10 after an air jet on recession, using the visual pain scale)

Following the initial periodontal clinical examination, the recessions were assigned to specific root coverage techniques and randomly (Random Group Generator-web site-<https://pt.rakko.tools/tools/59/>) divided into two groups based on the treatments. Each patient received both surgical, with one hemiarch undergoing a coronally Advanced Flap associated with A-PRF membranes (CAF + A-PRF) and the other hemiarch undergoing a coronally advanced flap associated with subepithelial Connective Tissue Graft (CAF + CTG). The allocation of which hemiarch received which technique was randomized.

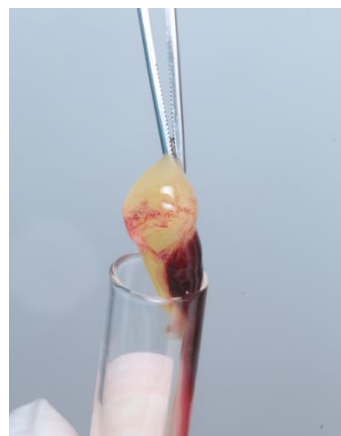
### *Surgical Technique*

The surgical procedure involved the simultaneous application of two surgical techniques for preparing the bilateral recipient areas. Regional blockade anesthesia was administered. An intrasulcular incision was made using a 15c blade (Swann Morton, England), followed by horizontal incisions at the base of the papillae, at the level of the CEJ, for mesial and distal aspects and relaxing vertical incisions. The epithelium of the papillae was then removed with scissors. The total mucoperiosteal detachment was performed up to the mucogingival line and from this point, a partial-thickness flap was created to obtain a flap without tension. The tooth root was scraped with a Gracey curette (Hu-Friedy, Rio de Janeiro, Brazil), followed by copious irrigation with saline solution. The recipient bed was protected with gauze soaked in saline solution while the grafts were prepared (Fig. 2) (Spada *et al.*, 2016).

The subepithelial connective tissue graft was harvested from the palate using the single incision technique (linear incision) (Xavier and Alves, 2015). The graft was then positioned over the recession and stabilized with suspensory sutures (Fig. 4). Subsequently, the flap was sutured over the graft using nylon 5.0 thread (Fig. 5) (Technofio, Goiânia, Brazil) (Spada *et al.*, 2016).



**Fig. 2:** Clinical aspect in the receptor region



**Fig. 3:** Clinical aspect of the fibrin clot formed after centrifugation



**Fig. 4:** A-PRF membranes were sutured in the right upper canine, lateral incisor, and central incisor, while CTG was placed in the left upper canine, lateral incisor, and central incisor



**Fig. 5:** Immediate postoperative appearance, after sutures were performed in a vertical mattress with 5-0 nylon

The protocol for obtaining and using Advanced platelet-rich fibrin (A-PRF) was based on the process outlined by Ghanaati *et al.* (2014) and involved three main stages.

- 1<sup>st</sup> stage: Venipuncture and blood collection:

Before initiating the root covering surgical procedure, venipuncture was performed by a nurse, collecting approximately 60 mL (6 glass-based vacuum tubes with no additive (Montserrat, China)) blood, depending on the length of the recipient bed. The venopuncture protocol followed the recommendations of the Ministry of health (Bin *et al.*, 2023).

- 2<sup>nd</sup> stage: Cell separation (centrifugation):

The collected blood was promptly taken to the centrifuge (centrifuge Montserrat FibrinFUGE25, Montserrat, China) in 10 mL portions, with each collection not exceeding 3 min. Fibrin membranes were obtained through centrifugation at approximately 1500 rpm for 14 min, resulting in a G-force of 210. Then, they were left at least 30 min in repose.

- 3<sup>rd</sup> stage: Preparation of A-PRF membranes:

Using the kit designed for making PRF membranes in stainless steel (Intra-Lock®), the intermediate portion of the centrifuged blood, containing the fibrin clot, was separated from the portion with red cells and platelet-poor plasma (Fig. 3). The fibrin clot was then deposited in the stainless-steel box and the compressive cap was placed over it without excessive tightening. The weight of the cap (130 g) was sufficient to compress the clot and produce the membranes without causing damage to the cellular structures present in the fibrin mesh.

The obtained A-PRF membranes, typically around 4 membranes on average, were positioned and sutured below the flap in the recipient bed using mattress sutures (Fig. 4). Subsequently, the flap was sutured over the graft with nylon 5.0 thread, following the same procedure as in the CAF + CTG group (Öncü, 2017) (Fig. 5).

### Follow-Up

Following the root-covering surgical procedure, a postoperative care plan was implemented, including medication prescriptions and follow-up appointments.

Medication prescriptions: Amoxicillin 500 mg every 8 h for 7 days (antibiotic therapy), Dipyron 500 mg every 6 h for 3 days (pain control), and Chlorhexidine 0.12% mouthwash twice daily for 15 days.

The sutures from the palate were removed after 7 days and the sutures from the recipient bed were removed at a later stage, 15 days post-surgery.

Patients were followed up for a total period of 180 days (Fig. 6), with clinical examinations conducted at 0 and 180 days and an intermediate follow-up at 90 days.

After the 180-day trial period, all patients were enrolled in a periodontal maintenance program to ensure ongoing oral health.



**Fig. 6:** Clinical aspect in 6 months follow-up

This comprehensive postoperative plan aimed to manage pain, prevent infection, and support the proper healing of the surgical sites, contributing to the overall success of the root covering procedures.

### Statistical Analysis

The statistical analysis of the clinical data obtained from the study involved the use of GraphPad Prism 8.0®. The following steps were performed:

1. Normality assessment: The Shapiro-wilk test was employed to evaluate the distribution of normality for the obtained data
2. Parametric analysis: For normally distributed data, the student's t-test was utilized for the analysis
3. Non-parametric analysis: The MANN-Whitney test was employed for the sensitivity parameter, which is a non-parametric test
4. Significance level: The significance level for the statistical tests was set at 5%

These statistical analyses aimed to assess the significance of the results obtained from the clinical data, providing valuable insights into the effectiveness of the different techniques used for root covering in Cairo type 1 gingival recession.

### Results

The study initially included a certain number of participants, but due to the exclusion of 2 patients (representing 8 gingival recessions) who chose to discontinue the postoperative follow-up visits, the final analysis was conducted on 40 RT1 gingival recessions (in 10 patients), evenly distributed between the CAF + A-PRF and CAF + CTG groups. Here are some key details about the participants:

- Total participants: 10 patients
- Total gingival recessions analyzed: 40 RT1 gingival recessions

Additional participant demographics:

- Gender distribution: 8 females and 2 males

- Tooth location: 16 gingival recessions in anterior teeth (40%) and 24 in posterior teeth (60%)
- Arch distribution: 34 recessions in the upper arch (85%) and 8 in the lower arch (15%)

The preoperative and postoperative results after 180 days within each group are summarized in Table 1. It indicates that both techniques demonstrated significant improvements in the evaluated periodontal parameters after 180 days. Notably, the CAF + CTG group exhibited a slightly superior outcome ( $p < 0.05$ ) in the gingival thickness parameter compared to the CAF + A-PRF group. However, for other parameters, both techniques showed similar and significant improvements ( $p > 0.05$ ).

Table 2 presents the values of clinical parameters for the CAF + CTG group after 180 days. The table indicates that all parameters demonstrated significant improvements ( $p < 0.05$ ) except for probing depth, which did not show a statistically significant variation at the end of the observation period. The specific values and changes for each parameter are outlined in the table.

**Table 1:** Postoperative comparisons (180 days) between the groups of mean values

| Measures (mm) | Groups       |              |
|---------------|--------------|--------------|
|               | CAF + CTG    | CAF + A-PRF  |
| HR            | 1.450±0.960  | 1.04±0.95    |
| CAL           | 1.450±0.940  | 1.05±1.19    |
| HG            | -0.860±0.940 | -0.31±1.28   |
| GT            | -0.055±0.080 | -0.015±0.83* |
| RW            | 1.180±1.360  | 1.04±1.49    |
| HC            | 1.860±1.850  | 1.27±1.07    |
| PD            | 0.000±0.610  | -0.16±0.65   |

HR = Height of Recession; CAL = Clinical Attachment Level; HG = Height of the attached Gingiva; GT = Gingival Thickness; RW = Recession Width; HC = Height of the Clinical crown. PD = Probing Depth. (\*) The statistically significant difference within the same evaluation parameter ( $p < 0.05$ )

**Table 2:** Comparison of preoperative and postoperative measurements in the CAF + CTG group

| Measures (mm) | Groups       |               |
|---------------|--------------|---------------|
|               | Preoperative | Postoperative |
| HR            | 2.30±0.86    | 0.70±0.80*    |
| CAL           | 3.45±0.94    | 2.00±1.07*    |
| HG            | 3.55±1.63    | 4.50±1.31*    |
| GT            | 2.06±0.80    | 2.64±0.71*    |
| RW            | 3.00±1.45    | 1.70±1.94 *   |
| HC            | 11.20±1.32   | 9.63±0.83 *   |
| PD            | 1.40±0.58    | 1.40±0.64     |

HR = Height of Recession; CAL = Clinical Attachment Level; HG = Height of the attached Gingiva; GT = Gingival Thickness; RW = Recession Width; HC = Height of the Clinical crown. PD = Probing Depth. (\*) Statistically significant differences within the same

**Table 3:** Comparison of preoperative and postoperative measurements in the CAF + A-PRF group

| Measures (mm) | Groups       |               |
|---------------|--------------|---------------|
|               | Preoperative | Postoperative |
| HR            | 2.15±1.03    | 1.00±0.91*    |
| CAL           | 3.20±1.28    | 2.15±1.03*    |
| HG            | 3.30±1.41    | 3.65±1.59     |
| GT            | 2.11±0.76    | 2.17±0.79     |
| RW            | 3.15±1.42    | 2.00±1.74     |
| HC            | 11.30±2.00   | 9.90±1.33*    |
| PD            | 1.35±0.53    | 1.36±0.60     |

HR = Height of Recession; CAL = Clinical Attachment Level; HG = Height of the attached Gingiva; GT = Gingival Thickness; RW = Recession Width; HC = Height of the Clinical crown. PD = Probing Depth. (\*) Statistically significant differences within the same

Table 3 presents the values of clinical parameters for the CAF + A-PRF group after 180 days. The table indicates that only the parameters of recession height, clinical attachment level, and clinical crown height demonstrated significant improvements after 180 days ( $p < 0.05$ ). Other clinical parameters did not exhibit significant differences at the end of the observation period ( $p > 0.05$ ). The specific values and changes for each parameter are outlined in the table.

The study observed a gain in gingival height of 1.45±0.96 mm in the CAF + CTG group and 1.04±0.95 mm in the CAF + A-PRF group, resulting in coverage of 54.3% in the test group and 73% in the control group. Full coverage (100%) of recessions was achieved at 7 sites (35%) in the A-PRF group and 10 sites (50%) with CTG. However, one patient in the A-PRF group experienced palate necrosis and more intense pain. Pain and swelling were common complications reported by all patients in the first postoperative week. Despite this, 100% of the patients expressed willingness to undergo the procedure again, primarily due to the improvement in sensitivity reported at all sites, a common complaint among patients. In the CAF + CTG group, there was a significant improvement ( $p < 0.05$ ) in dentin sensitivity from 2.75±3.29-0.30 ± 0.92, and in the CAF + A-PRF group, it improved from 3.60±3.31 to 0.80±1.32.

## Discussion

The study by Aroca *et al.* (2009) was among the pioneers in investigating the treatment of gingival recessions with L-PRF. This study, conducted through a word-of-mouth referral system, compared the use of CAF associated with L-PRF (test group) versus CAF alone (control), resulting in a mean coverage of 80.7% in the test group and 91.5% in the control group. In terms of total coverage of recessions, the current study achieved successful outcomes in sites treated with CTG and A-PRF (Tables 2-3). These results can be compared with findings

in the literature, where the percentage of coverage in the PRF group ranged from 50% (Öncü, 2017) to 52.2% (Aroca *et al.*, 2009; Kuka *et al.*, 2018), 55% (Keceli *et al.*, 2008) and 92.2% (Eren and Atilla, 2014). Comparative studies between CAF alone and CAF associated with L-PRF have consistently demonstrated full coverage and superior mean coverage in the groups where the L-PRF membrane was incorporated (Aroca *et al.*, 2009; Jankovic *et al.*, 2010; Kuka *et al.*, 2018). Moreover, when comparing the association of CTG with the PRF membrane at the same surgical site, 89% coverage was achieved, compared to 79.9% at the site with CTG alone (Keceli *et al.*, 2015).

The CAF technique, which has been employed for several decades, does not, in isolation, exhibit improvements in the width and thickness of keratinized tissue, crucial factors for the long-term maintenance of root coverage (Pini Prato *et al.*, 2018). In our study, we observed an enhancement in the gingival band in both groups, with a greater gain in the CTG group, although without statistical significance between them (an increase of  $0.86 \pm 0.94$  mm versus  $0.31 \pm 1.28$  mm). This finding contrasts with the results reported by Öncü (2017), which demonstrated the superiority of the L-PRF test group. In terms of the thickness of the attached gum, the control group also exhibited a more significant improvement in the parameters compared to the test group, showing a statistically significant difference. This aligns with similar findings in other studies (Eren and Atilla, 2014; Öncü, 2017) (Table 1). In a systematic review, (Rodas *et al.*, 2020) demonstrated the superiority of connective tissue regarding the increase in both height and thickness of the attached gingiva. Similar results were found in the systematic review developed by Mancini *et al.* (2021).

While our study demonstrated superior results with CTG, A-PRF membranes might be recommended for patients lacking adequate gingival thickness at the donor site or those unwilling to undergo graft harvesting. Moreover, the advantages of A-PRF include less postoperative pain and the absence of risks associated with palate artery injury (Bin *et al.*, 2023). The choice between these techniques should be based on individual patient characteristics, preferences, and specific clinical indications.

The positive effects of PRF have been attributed to its various growth factors, including PDGF, VEGF, and TGF, as well as cytokines, glycoprotein structures, and the dense fibrin matrix network (100 times larger than normal). This network enhances and promotes angiogenesis and matrix synthesis (Keceli *et al.*, 2008; 2015). Different centrifugation protocols have been reported in studies using various centrifuge models, influencing the quality of the membranes formed. In summary, the protocols adopted are often based on the studies of Choukroun (Dohan *et al.*,

2006), which form L-PRF membranes using 2700 rpm for 12 min. This study opted for a slower centrifugation protocol of 1500 rpm for 14 min, resulting in A-PRF membranes, following the approach of (Ghanaatif *et al.*, 2014). In immunohistochemical analysis, A-PRF demonstrated a lower density fibrin network, with cells spread more evenly in the clot, a higher number of platelets and progenitor cells, and a slower and more prolonged release of growth factors compared to L-PRF. These characteristics may contribute to an improvement in tissue regeneration (Ghanaati *et al.*, 2014; Isobe *et al.*, 2017; Kobayashi *et al.*, 2016).

The advantages of using L-PRF or A-PRF are rooted in platelet cytokines that play a crucial role in the initial mechanisms of the healing process. They stimulate cell migration and proliferation, induce fibrin matrix remodeling, and promote the secretion of a collagen matrix. These cytokines become trapped in the fibrin mesh. The increased number of leukocytes is pivotal in the phagocytosis of microorganisms and necrotic tissues. Moreover, they guide the future regeneration of these tissues by releasing cytokines and growth factors. Leukocytes also play a role in stimulating angiogenesis and tissue formation (Fernandes *et al.*, 2021; Ghanaati *et al.*, 2014; Isobe *et al.*, 2017; Kobayashi *et al.*, 2016).

## Conclusion

After 180 days, both the Coronally Advanced Flap associated with Advanced Platelet Rich Fibrin (CAF + A-PRF) and the Coronally Advanced Flap associated with Subepithelial Connective Tissue Grafting (CAF + CTG) techniques exhibited significant improvement in the evaluated periodontal parameters. Thus, both techniques can be effectively used for the treatment of Cairo Class I gingival recessions. However, the connective tissue grafting technique demonstrated a slight superiority, showing statistically significant improvements in clinical parameters over the 180-day period. Additionally, there was statistical superiority in gingival tissue thickness compared to the PRF technique.

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## Author's Contributions

**Letícia Nadal, Patricia Oehlmeyer Nassar, Simone Karine Rothen, Edson Oliveira Silva and Carlos Augusto Nassar:** The work was prepared and developed.

**Luiza Roberta Bin:** The work was published by all authors, and submitted.

## Ethics

The authors have declared that no competing interests exist. The study was submitted and approved by Unioeste Human Research Ethics Committee 3.359.508, CAAE 13806519.0.0000.0107. All the patients received and signed the informed consent form.

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