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Cast versus 3D-printed cobalt-chromium framework for removable partial denture: a case report

Estrutura em cobalto-cromo fundida versus impressa para prótese parcial removível: relato de caso

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ABSTRACT

Background: The diverse steps in the Cobalt-Chromium (Co-Cr) casting process can induce misfit, highlighting the need for new methods to improve the fitting of metallic framework for Removable Partial Dentures (RPD). **Objective:** This case report describes and compares the clinical performance of two fabrication techniques mandibular metallic framework for RPDs. **Case report:** After preparing the abutment teeth and the impression to obtain the working model, two fabrication techniques were applied the metallic framework: lost wax technique and 3D printing. For the 3D printing technique, the working model was digitized, and the framework was planned using Exocad (GmbH) software and fabricated with a 3D printer (EOS M280). With the denture base conventionally processed for both metallic frameworks, they were digitized, and the distance between the metallic framework and the mandibular arch was measured using reverse engineering software (GOM Inspect). The RPDs were installed in an interleaved manner to collect data on OHRQoL using the OHIP-14 questionnaire and patient satisfaction (installation and after 3 months). **Conclusions:** The 3D printing process produced a metallic framework comparable to the cast Co-Cr alloy framework in terms of fit, quality of life related to oral health, patient satisfaction, and the absence of surface porosity.

KEYWORDS

3D-printed; Direct metal laser sintering; Framework; Patient satisfaction; Removable partial denture.

RESUMO

Contexto: As diversas etapas envolvidas no processo de fundição da liga de Cobalto-Cromo (Co-Cr) podem gerar desajustes, evidenciando a necessidade de métodos alternativos que promovam um melhor assentamento da estrutura metálica em Próteses Parciais Removíveis (PPR´s). **Objetivo:** Este relato de caso descreve e compara o desempenho clínico de duas técnicas de fabricação de estruturas metálicas mandibulares para PPR´s. **Relato de caso:** Após o preparo dos dentes pilares e a moldagem para obtenção do modelo de trabalho, duas técnicas de fabricação foram utilizadas para confecção da estrutura metálica: técnica da cera perdida e impressão 3D (tridimensional). Na técnica de impressão 3D, o modelo de trabalho foi escaneado, a estrutura foi planejada no *software* Exocad (GmbH) e impressa em uma impressora 3D (EOS M280). Com a base da prótese processada de forma convencional em ambas as estruturas metálicas, estas foram escaneadas e a distância entre a estrutura metálica e o arco mandibular foi mensurada por meio de um *software* de engenharia reversa (GOM Inspect). As PPR´s foram instaladas de forma alternada para coleta de dados sobre a QVRSB, utilizando o questionário OHIP-14, além da avaliação da satisfação do paciente (no momento da instalação e após 3 meses). **Conclusões:** O processo de impressão 3D resultou em uma estrutura metálica comparável àquela obtida por fundição da liga Co-Cr, em termos de ajuste, qualidade de vida relacionada à saúde bucal, satisfação do paciente e ausência de porosidades superficiais.

PALAVRAS-CHAVE

Impressão em 3D; Sinterização direta de metal a laser; Estrutura; Satisfação do paciente; Prótese parcial removível.

INTRODUCTION

Traditionally, metallic frameworks for removable partial dentures (RPDs) are fabricated using cobalt-chromium (Co-Cr) alloys, which are valued for their mechanical strength [1], resistance to corrosion [2,3], and durability against wear [4,5]. Despite these advantages, the conventional casting process for Co-Cr alloys may result in non-uniform microstructures and solidification flaws, and it relies heavily on the skill of the dental technician. These factors can lead to inconsistencies in framework fit, reduced mechanical performance, and overall diminished quality and clinical outcomes for RPDs [6].

The introduction of computer-aided design and computer-aided manufacturing (CAD-CAM) has enabled the production of RPD metallic frameworks through both subtractive and additive methods [6]. Milling Co-Cr alloy blocks or discs, an example of subtractive manufacturing, raises the overall expense of RPD fabrication and has limitations in precisely reproducing intricate features, such as retention and opposition clasps, while also producing material waste. Conversely, additive manufacturing permits the recovery and reuse of some unused powder [7,8]. Additionally, by avoiding the casting process and the application of force during fabrication, additive manufacturing reduces the risk of structural deformation and eliminates casting shrinkage, thereby minimizing associated defects [9,10].

Laser-sintering techniques are commonly referred to by different terms, such as selective laser melting (SLM), selective laser sintering (SLS), and direct metal laser sintering (DMLS) [11]. While SLM entails the full fusion of metal particles, SLS and DMLS generally involve only partial melting of the powder [12-14]. A key distinction between SLS and DMLS lies in the composition of the materials and the extent of melting achieved. SLS can process a variety of powders, including polymers, ceramics, and metals, with partial sintering [12-16]. In contrast, DMLS utilizes a blend of metal powders with varying melting temperatures, where the lower melting point component fully melts, allowing partial bonding with the higher melting point particles [12,13,16-18].

In vitro studies have evaluated Co-Cr alloy frameworks for RPD fabricated through additive manufacturing [19-25]. Some authors have reported homogeneity, smaller grain size in the microstructure, clinically acceptable load-to-fracture values [26], and low porosity related to the manufacturing parameters in Co-Cr alloy frameworks manufactured by SLM compared to those fabricated using cast Co-Cr alloys [27]. Two case reports [28,29] showed the absence of complications or clinical failures within a 4-year follow-up period [29] and acceptable fit of 3D-printed Co-Cr metallic frameworks compared to cast Co-Cr metallic frameworks [28]. Additionally, a clinical trial concluded that SLM technology can result in better patient satisfaction in the short-term follow-up [30]. However, there is little evidence of data in the literature with clinical evidence, proven by the existence of only one case report that evaluated patient satisfaction with an RPD featuring a metallic framework manufactured using the DMLS method, which was also used by the aforementioned case report [31].

Given the repetitive cycles of insertion and removal of the removable partial denture metallic framework, it is essential that the fundamental requirements retention, support and stability function correctly to avoid compromising oral health and reducing the clinical performance of RPDs [6]. Hence, evaluating these requirements from the patient's perspective is equally important. Therefore, given the limited evidence from case reports featuring three-dimensional and patient-centered analyses with follow-up data, the objective this case report describes and compares the clinical performance of two fabrication techniques mandibular metallic framework for RPDs.

CASE REPORT

A 56-year-old female patient visited the Department of Dental Prosthetics of the Blinded/Redacted without using any type of denture and in need of prosthetic rehabilitation. The clinical examination showed the absence of teeth 35, 36, 46 e 47 configuring a Kennedy Class III with modification 1. The radiographic examination revealed restorations without marginal infiltration or fracture and prosthetic abutments without loss of bone insertion or mobility. The prosthetic rehabilitation options were discussed with the patient, who agreed to a mandibular Removable Partial Denture (RPD). An Informed Consent Form was provided and signed by the patient and approved by the Ethics Committee of Blinded/Redacted (Blinded/Redacted 5.191.014).

The patient's mouth preparation began with the clinical procedure of scaling and root planing of the crown and root in the fourth, fifth and sixth sextants. Guide planing was performed on the distal face of prosthetic abutment teeth 37 and 48, and retentive areas were created by adding composite resin, exactly in the area of the flexible portion of the retention arms, on prosthetic abutment teeth 37 and 45.

The mandibular arch was molded with alginate (Avagel; Dentsply Sirona). The alginate impression was disinfected by storing it in a closed container and spraying it with 1% sodium hypochlorite for 10 minutes before being poured with Type III dental stone (Plaster stone type III; Asfer) to obtain a preliminary model. Using this model, the path of insertion and removal of the removable partial denture framework was planned by analyzing the guide planes areas, retentive areas in the prosthetic abutments, and the location of the rests. After transferring the planning to the mouth (Figure 1), a final impression of the mandibular arch was taken using alginate (Avagel; Dentsply Sirona). The impression was disinfected by storing it in a closed container and spraying it with 1% sodium hypochlorite for 10 minutes, then poured to obtain the working model (Plaster Dent-Mix type IV; Asfer). Based on the determined path of insertion and removal of the removable partial denture framework, two workflows were followed for processing the Cobalt-Chromium (Co-Cr) alloy: the cast method (lost wax technique) and 3D printing (*Direct Metal Laser Sintering*) (Figure 2).

Figure 1 - Partially edentulous mandibular arch.

To obtain the cast Co-Cr alloy metallic framework, the physical working model was sent to the dental prosthesis laboratory, where it was duplicated by using a reversible hydrocolloid (Duplicator; Kota Knebel) for phosphate coating. After 40 minutes, the model was removed and dehydrated for 30 minutes in an oven at 250°C and then immersed in liquid wax at 130°C for 10 seconds.

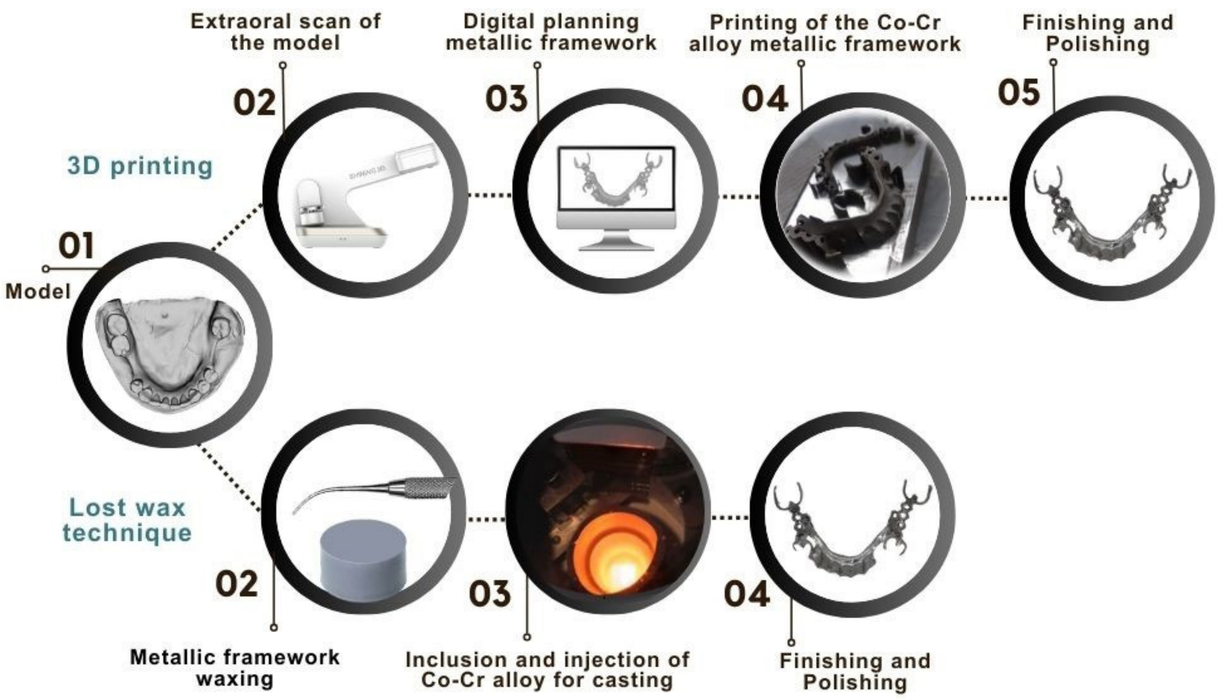


Figure 2 - Flowchart of the work sequence for the manufacture of metallic framework in cast and 3D-printed Co-Cr alloy.

After the wax was absorbed, the model was returned to the oven for 20 minutes at 180°C. Once the refractory model was done, wax plasty was performed and included for coating (VarseoVest P; Bego) of the Co-Cr alloy (Remanium GM 280) using the lost-wax technique. Then, the coated resin pattern was placed in a gas oven (Midither 200 MP; Bego) and initially heated at 650°C for 30 min; the temperature was then increased to 1010°C. The investment was placed in an induction casting machine (Fornax T; Bego) and the alloy was cast at 1440°C [32].

Regarding the fabrication of the 3D-printed Co-Cr alloy metallic framework (EOS CobaltChrome MP1), the working model was scanned with a laboratory scanner (3D shining DS-EX; Talmax) and the STL (Standard Tessellation Language) file was then uploaded to the software (PartialCAD Exocad; GmbH) for CAD (computer-aided design) planning of the metallic framework (Figure 3A). The framework was printed (EOSINT M280; EOS GmbH) with the specifications for 3D-printed framework were: 30 μm powder layer thickness, 170 W power, 200 W ytterbium fiber laser, particle size around 15–45 μm, and a working atmosphere of argon. After the printing process, the frameworks were stress-relieved at 650ºC for 6 h in the furnace (Jung J300, model TB09613, Blumenau, Brazil) and removed from the build platform (Figure 3B) [6].

Once both metallic frameworks were completed, the finishing and polishing process (Figure 3C), by a single operator, was carried out for correct fitting in the physical working model. For finishing process, the following protocol was followed: spruce cutting disc (Schelble), mounted tip for gray Co-Cr #4 (Schelble), mounted tip for gray Co-Cr #3 (Schelble), mounted tip for gray Co-Cr #2 (Schelble) and mounted tip for gray Co-Cr #1 (Schelble). For polishing process, the following sequence was followed: brown rubber tip (Cromox) and felt disc with brown polishing compound (Abraspol; Kota Knebel).

Clinically, the metallic frameworks were tested in the patient's mouth (Figure 4A, B), with adjustment required for adequate seating of the 3D-printed Co-Cr alloy framework. Later the color and shape of the artificial teeth (Biolux OMC; VIPI) as well as the gingiva color (STG; VIPI) were selected. To assemble the artificial teeth, the maxillary arch was molded (Avagel; Dentsply Sirona) to obtain a new impression that was then poured (type III stone plaster; Asfer) and mounted on a semi-adjustable articulator (4000-S; BioArt) with a Camper table (BioArt), followed by manual intercuspation of the mandibular model, once the patient presented occlusal stability.

Without clinical proof of mounting the teeth in wax, the acrylic base of the RPDs for both

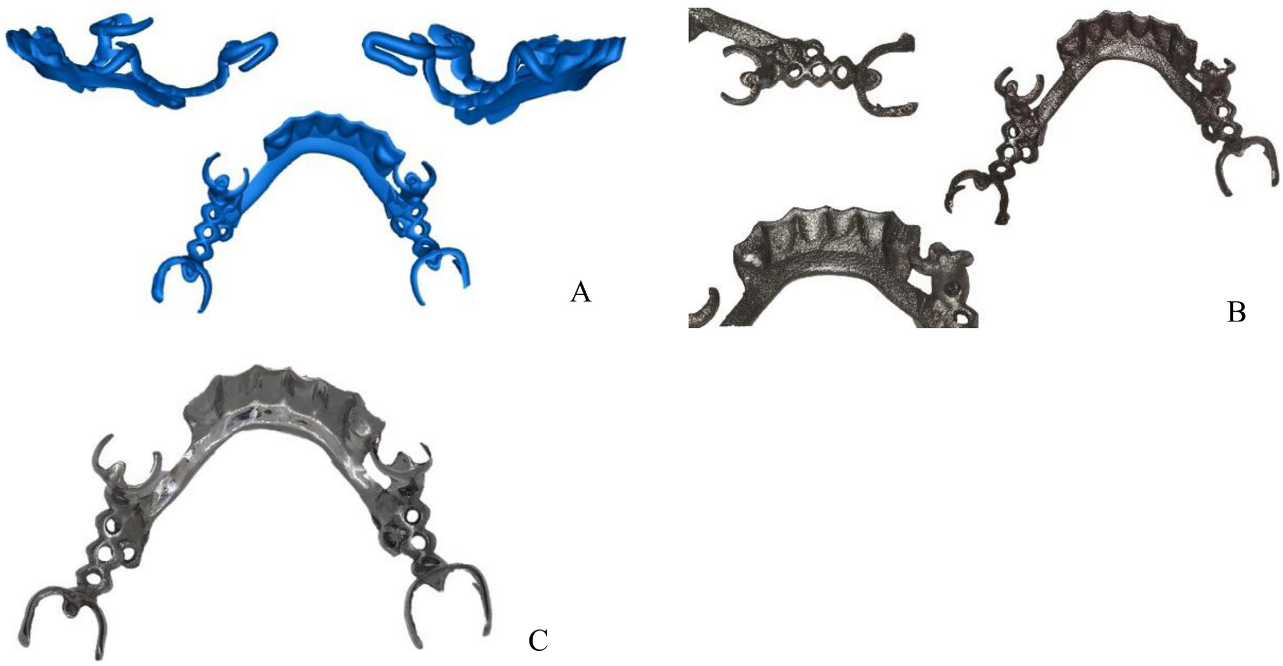
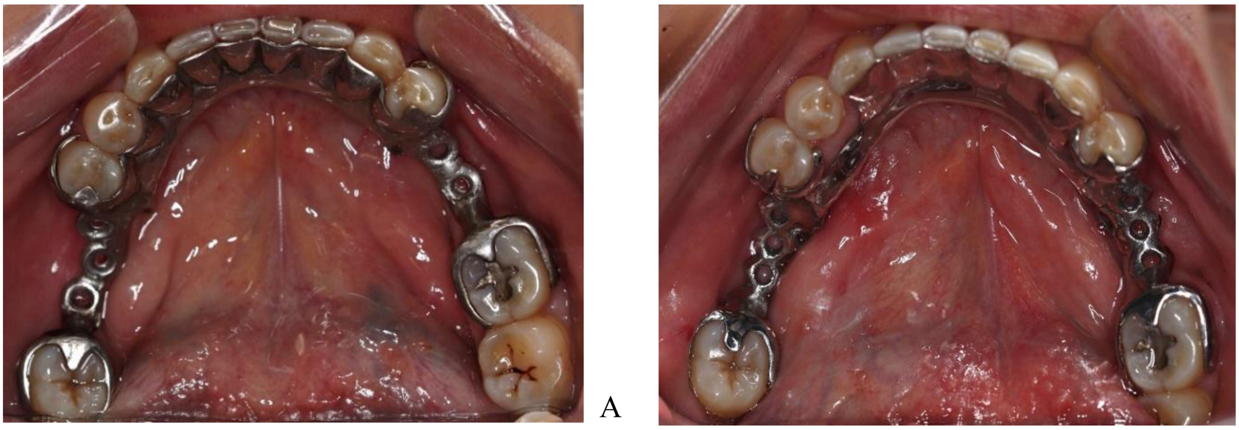
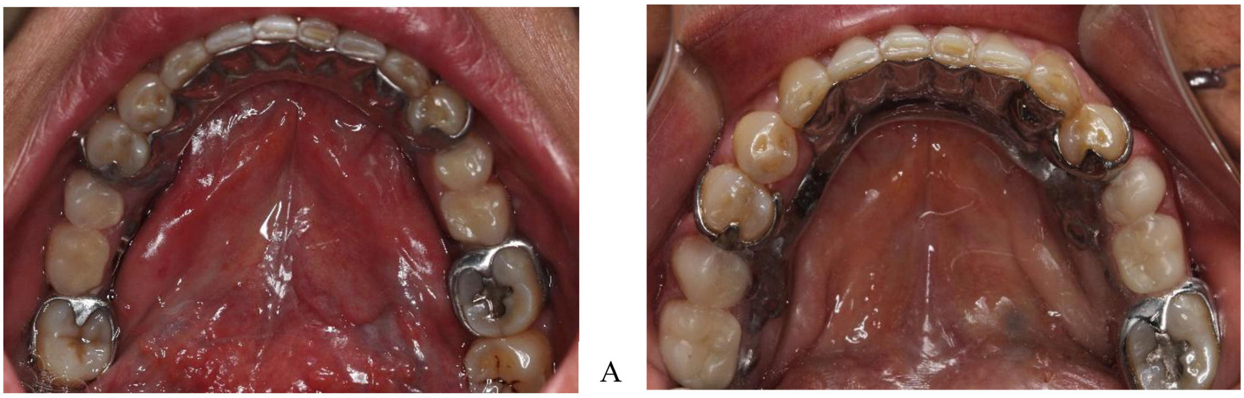


Figure 3 - Manufacturing of the metallic framework in the 3D-printed Co-Cr alloy. A, CAD planning of the metallic framework. B, Printing of the metallic framework. C, Finishing and polishing of the metallic framework by the conventional method.



A B

Figure 4 - Clinical test of the metallic framework. A, cast Co-Cr alloy, B, 3D-printed Co-Cr alloy.



A B

Figure 5 - Production of the acrylic base of the RPDs. A, RPD with cast Co-Cr alloy metallic framework. B, RPD with 3D-printed Co-Cr alloy metallic framework.

metallic frameworks was made using the same methods (Figure 5A, B). The metallic framework with the artificial teeth set was included (Muffle; OGP) and subjected to an initial hydraulic press for 12 hours. Afterwards, the mold obtained was filled with heat-cured denture base resin (Cril Plus; VIPI) in the proportion of 14 g of powder to 6.5 mL of liquid. A polyethylene sheet was positioned over the resin and a pressure of 1,250 kgf was applied for 12 hours. The classic procedure for polymerizing the acrylic resin was performed in a heated water cycle at 74°C for 12 hours [33]. The PPRs were installed without the need for any clinical adjustment.

The RPDs were scanned using an intraoral scanner (3Shape TRIOS), and the resulting STL files were imported into reverse engineering software (GOM Inspect; GmbH, Germany). Each RPD was superimposed onto the corresponding intraoral scan of the mandibular arch using a two-step alignment process (Figure 6A). Initially, a point-based pre-alignment was performed by selecting anatomical reference regions, including the middle third of the lingual surface of the mandibular central incisor (corresponding to the seating area of the lingual plate) and the center of the edentulous alveolar ridge crest on both sides (corresponding to the denture base). Subsequently, a global best-fit alignment was applied to optimize the superimposition between the RPD and the mandibular arch. Framework fit was quantitatively assessed by measuring the linear distance between the metallic framework and the abutment teeth at predefined regions. Measurements were obtained at multiple points for each component: rests (18 points) (Figure 6B), retention arms (52 points) (Figure 6C), opposition arms (52 points) (Figure 6D), and major connector (149 points) (Figure 6E). These distances represented local deviations between the framework and the supporting structures. Mean values were calculated for each component to represent overall fit [34].

To assess the differences between the cast and 3D-printed Co-Cr frameworks, a Shapiro–Wilk test was performed to verify the normality of the paired data for each component.

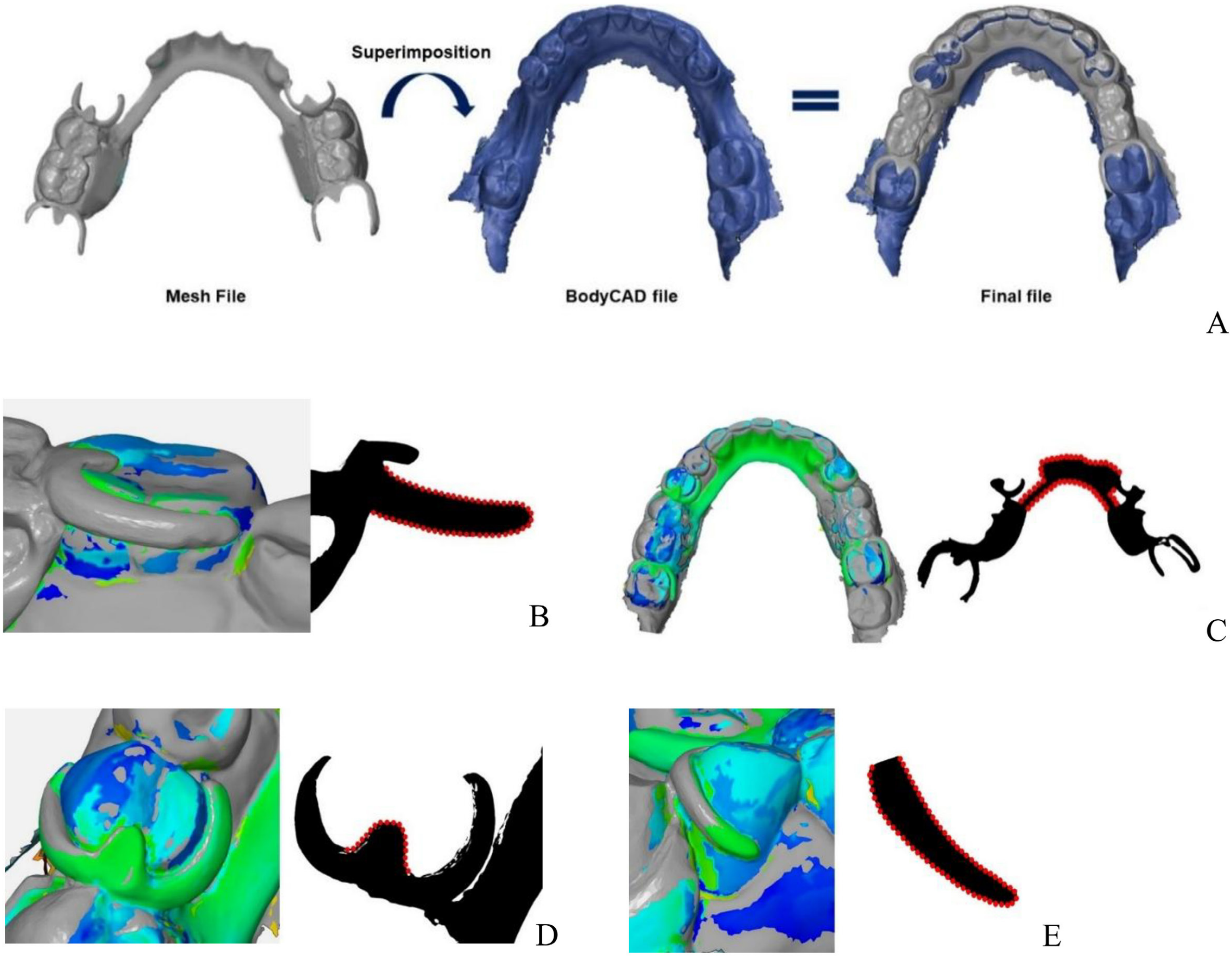


Figure 6 - Quantitative analysis of the RPDs fitting on the abutment teeth. A, Overlapping of the RPD on the mandibular arch. Measurement of specific areas of the metallic framework: rest (B), retention clasp (C), opposition clasp (D) and major connector (E). Red dots indicate measurement areas of the fitting between the metallic framework and abutment teeth.

Since the data were normally distributed (p>0.05), paired t-tests were applied to compare each component between the two fabrication methods. A significance level of α=0.05 was adopted for all tests.

Subsequently, the RPD fabricated using cast Co-Cr alloy metallic framework was installed and after three months, it was removed for the installation of the RPD with the 3D-printed Co-Cr framework, which also remained in the mouth for three months. At the installation and after three months wearing each RPD, the patient satisfaction and oral health-related quality of life (OHRQoL) variables were collected. Only at installation, occlusal radiographs (Dabi Atlante, Spectro 70X Seletronic) were taken to evaluate the presence of porosity in both Co-Cr alloy metallic frameworks.

Patient satisfaction with RPDs was assessed using an analog scale numbered from 1 to 5, with 1 being “unsatisfactory” and 5 being “excellent” to assess the following aspects: general satisfaction, aesthetics, retention, speech, and chewing [35]. To assess the influence of rehabilitation on oral health-related quality of life, a questionnaire based on the Oral Health Impact Profile (OHIP-14) was used, consisting of 14 questions to be answered on a simplified Likert scale, ranging from 0 (never), 1 (rarely), 2 (sometimes), 3 (repeatedly), and 4 (always). A lower score indicated better oral health-related quality of life [36].

RESULTS

The cast Co-Cr alloy framework showed better fit in the remaining regions, opposing arms, and major connector, while the 3D-printed Co-Cr alloy exhibited better fit in the retaining arms area. The statistical analysis revealed significant differences between the cast and 3D-printed frameworks for the opposition arms (p=0.02) and the major connector (p=0.01).

For the rest (p=0.15), retention arms (p=0.10), and overall mean deviation (p=0.08), no statistically significant differences were observed (Figure 7).

Both RPDs were free from areas of porosity (Figure 8A, B) and provided the patient with improved OHRQoL and satisfaction regarding aesthetics, retention, speech and chewing (Table I and II). Clinically, calculus was observed in the lingual plate region of cast Co-Cr alloy metallic framework, while the RPD with the 3D-printed Co-Cr alloy framework remained free of calculus (Figure 9).

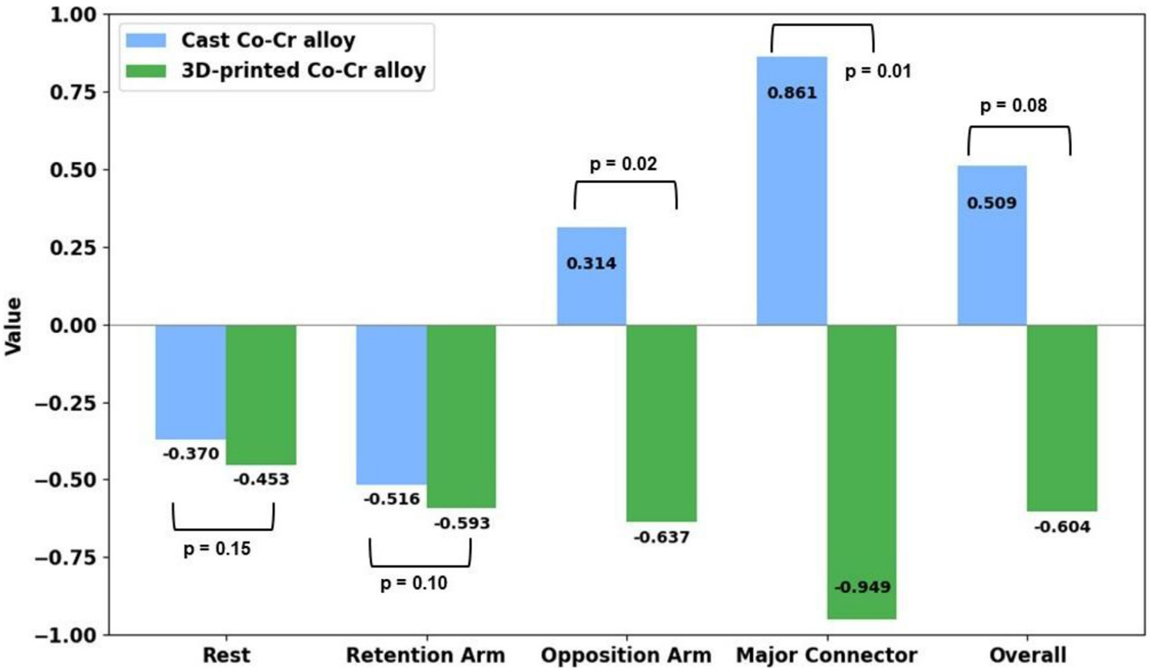


Figure 7 - Quantitative analysis of metal framework displacement in relation to the abutment teeth.

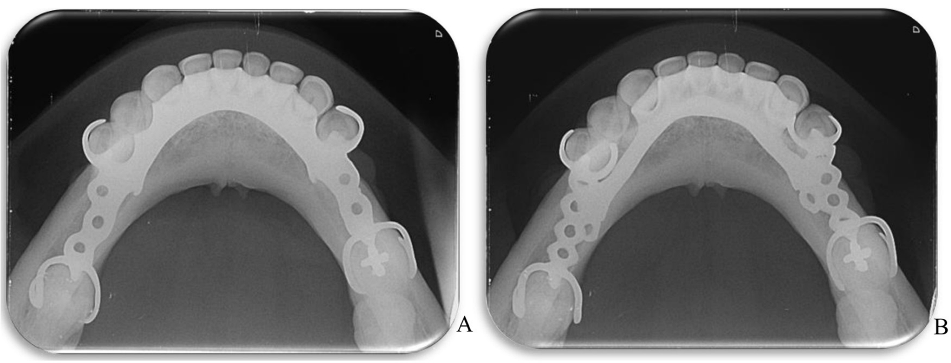


Figure 8 - Occlusal radiographs. A, RPD with cast Co-Cr alloy metallic framework. B, RPD with 3D-printed Co-Cr alloy metallic framework.

Figure 9 - Clinical condition of RPDs. (A) RPD with cast Co-Cr alloy metallic framework. (B) RPD with 3D-printed Co-Cr alloy metallic framework.

Table I - Oral health-related quality of life (OHRQoL) with RPDs obtained with the cast and 3D-printed Co-Cr alloy metallic framework

OHIP-14 item	Metallic framework			
	Cast Co-Cr alloy		3D-printed Co-Cr alloy	
Functional limitation	T0	T1	T0	T1
Trouble pronouncing words	0	0	0	0
Taste worse	0	0	0	0
Painful aching	0	0	0	0
Uncomfortable to eat	1	0	0	0
Psychological discomfort				
Self-conscious	0	0	0	0
Tense	0	0	0	0
Physical disability				
Diet unsatisfactory	0	0	0	0
Interrupt meals	0	0	0	0
Psychological disability				
Difficult to relax	0	0	0	0
Been embarrassed	0	0	0	0
Social disability				
Irritable with others	0	0	0	0
Difficulty doing jobs	0	0	0	0
Handicap				
Life unsatisfying	0	0	0	0
Unable to function	0	0	0	0

T0: installation, T1: after 3 months, 0: never, 1: rarely, 2: sometimes, 3: repeatedly, 4: always.

Table II - Patient satisfaction with the RPDs obtained with the cast and 3D-printed Co-Cr alloy metallic framework

Questions	Cast Co-Cr alloy		3D-printed Co-Cr alloy	
Aesthetics	T0	T1	T0	T1
Retention	5	5	5	5
Speaks	5	5	5	5
Mastication	5	5	5	5
Comfort	5	5	5	5
Overall satisfaction	5	5	5	5

T0: installation, T1: after 3 months.

DISCUSSION

The 3D printing Direct Metal Laser Sintering (DMLS) process produced a metallic framework made of Cobalt-Chromium (Co-Cr) alloy that is comparable to the cast Co-Cr alloy framework in terms of fit, quality of life related to oral health and patient satisfaction after 3-month follow-up, and the absence of surface porosity.

The residual thermal stress generated from rapid heating and cooling during the fabrication of cast Co-Cr alloy metallic frameworks can impair its physical and mechanical properties. This phenomenon may lead to retentive displacement, affecting the accommodation of the framework. However, when performed properly, this process may not have a clinical impact [37-39]. In this context, although there was no difference between the two frameworks for the rest, retention arm and overall, the cast Co-Cr alloy framework had a better overall fit, as confirmed in the study by Heiba et al. [39] when it was observed that the cast framework adapted better than the 3D-printed one [39].

Furthermore, the better fit of the cast Co-Cr alloy framework can be attributed to the challenges associated with the 3D printing metallic frameworks with larger dimensions and components using the DMLS method.

These challenges may arise from difficulties in construction orientation of objects and the placement of sprues during the 3D printing process, potentially impacting the fit of the metallic framework to the dental surfaces [40-43].

Finishing and polishing can influence the precision of metallic framework components, especially in the clamp region [44]. This is evident in the current case report, as clinical adjustments were necessary during the proofing of the 3D-printed Co-Cr alloy metallic framework for adequate fit. Brudvik & Reimers reported an average metal loss of 127 μm from the surface of Co-Cr alloy frameworks after finishing and polishing [45]. Therefore, the average misfit values in the 3D-printed Co-Cr alloy metallic framework in this case report can also be attributed to potential human-induced errors, as reported in other studies evaluating finished and polished metallic frameworks [38,43-49].

The superposition methodology allowed the collection of multiple measurement points and the calculation of the best possible fit between the mandibular arch and the metallic framework. Additionally, the use of color mapping helped identify overloaded or misaligned areas of the metallic framework. However, previous reports have shown that metallic frameworks exhibit clinically acceptable precision values (<311 μm) [50] to ensure their functionality and biomechanical performance [48]. Therefore, we emphasize that the fit values of both metallic frameworks (cast and 3D-printed) fall within the acceptable ranges. Consequently, the numerical differences reported between metallic frameworks may not negatively impact clinical performance or be perceptible to patients and clinicians [41,43,47].

Cast metal alloys may exhibit porosity or internal defects due to impurities and previously cast sprues, which can lead to changes in the properties of the reused alloy. Therefore, no new metal should be added in successive reformulations of predominantly base alloys, or only 50% of the new metal may be added to previously cast alloys. The foundation of these guidelines is that some essential secondary elements present in small quantities in the original alloy may be lost through evaporation or oxidation [51-58].

In this specific case report, the absence of porosity in the radiograph of the cast Co-Cr alloy metallic framework can be attributed to the technical quality of the casting process. Thus, the DMLS 3D-printed Co-Cr alloy metallic framework also showed no porosities due to the post-heat treatment applied to the alloy after processing [6]. Additionally, the porosity of laser-sintered alloys can be influenced and controlled by laser sintering technology operating parameters, such as layer thickness, laser power, laser wavelength, and scanning speed [6]. Based on these facts, printer parameters were crucial in ensuring the absence of porosity in the 3D-printed Co-Cr alloy metallic framework.

In most cases, patient satisfaction is subjective and can vary significantly between individuals. Furthermore, it has been shown that the evaluation of RPDs differs from dentists to patients. Variables such as survival rate, prosthesis longevity, and frequency of complications are often prioritized by dentists; whereas social and psychological impacts of treatment and cost-effectiveness are more relevant from the patient's perspective. The Oral Health Impact Profile (OHIP) effectively assesses patients' perceptions of oral health and has been used to measure oral health-related quality of life (OHRQoL) of removable denture users, proving to be a reliable measurement method [57-65].

In this context, this case report demonstrated favorable OHRQoL and satisfaction for the patient rehabilitated with a RPD, regardless of the processing method of the Co-Cr alloy. This was also observed by Muehlemann et al. [31] when comparing workflows for fabricating the metallic framework using the lost wax casting technique and DMLS, over periods of 3 months and 2 years of removable partial denture wear. Alongside this previous finding, it can be suggested that the overall satisfaction is attributed to the favorable biomechanical design of the RPD, which contributes to the adequate fitting of metallic frameworks. In parallel, the absence of compression areas and adherence to the functional limits of the RPD ensured comfort and safety during speech and chewing. Given that the case involves posterior edentulous areas with limited spaces, aesthetics was not the primary issue for the patient, but rather functionality.

After 3 months, the patient chose to keep the RPD obtained with the 3D-printed Co-Cr alloy metallic framework without knowing it was the 3D-printed. Clinically, calculus was observed in the lingual plate region of cast Co-Cr alloy metallic framework, while the RPD with the 3D-printed Co-Cr alloy framework remained free of calculus.

This occurred likely because the patient did not store it in an appropriate environment. However, the framework was cleaned and subsequently polished.

The Co-Cr alloy processed by DMLS for the fabrication of a metallic framework for a removable partial denture met the patient's functional needs. The findings of this case report reinforce that additive manufacturing can now be seen as a reality for rapid and cost-effective fabrication. In addition, it serves as a solution to the casting issues that frequently impact the quality of metallic frameworks for PPDs, along with reduced laboratory work time, improved dentist-CAD communication, and the ability to replicate metallic frameworks through archiving the design, often within just a few hours. However, this clinical case is limited in that we did not create a fully digital RPD and observed the interaction between Co-Cr, denture base resin and 3D-printed teeth. Furthermore, there was no randomization of the order of installation of the frameworks or a washout period between the installation of the two frameworks. Conducting randomized clinical trials with longitudinal follow-up to collect long-term data will provide a more reliable indication of the material's performance.

CONCLUSION

In this single clinical case, the 3D printing process produced a metallic framework comparable to the cast Co-Cr alloy in terms of fit, oral health-related quality of life, patient satisfaction after three months of prosthesis use, and absence of surface porosity. These findings should be interpreted with caution and cannot be generalized.

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Data availability

Data may be made available upon request.

Author's Contributions

ALCP: Conceptualization, Methodology, Investigation, Writing – Original Draft Preparation, Writing – Review & Editing. MRSC: Investigation, Writing – Original Draft Preparation, Writing – Review & Editing. PTMB: Investigation, Software, Writing – Original Draft Preparation, Writing – Review & Editing. EDFF: Investigation, Writing – Original Draft Preparation, Writing – Review & Editing. MHGV: Writing – Original Draft Preparation, Writing – Review & Editing. ALJ: Funding Acquisition, Writing – Original Draft Preparation, Writing – Review & Editing. AFPC: Project Administration, Supervision, Writing – Original Draft Preparation, Writing – Review & Editing.

Conflict of Interest

The authors have no conflicts of interest to declare.

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Regulatory Statement

This study was conducted in accordance with all the provisions of the local human subjects oversight committee guidelines and policies of: Universidade Federal do Rio Grande do Norte (UFRN). This study protocol was reviewed and approved by Research Ethics Committee of the Universidade Federal do Rio Grande do Norte, approval number 5.191.014.

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