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VARIÁVEIS RELACIONADAS AO COMPORTAMENTO EM FADIGA E MODO DE FALHA DE RESTAURAÇÕES CAD/CAM

Santa Maria, RS
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Tese apresentada ao Curso de Doutorado do Programa de Pós-Graduação em Ciências Odontológicas da Universidade Federal de Santa Maria (UFSM, RS), como requisito parcial para a obtenção do título de **Doutor em Ciências Odontológicas com Ênfase em Prótese Dentária**.

Orientador: Prof. Dr. Luiz Felipe Valandro
Coorientadora: Prof. Dra. Andressa Borin Venturini

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RESUMO

VARIÁVEIS RELACIONADAS AO COMPORTAMENTO EM FADIGA E MODO DE FALHA DE RESTAURAÇÕES CAD/CAM

AUTOR: Helder Callegaro Velho
ORIENTADOR: Luiz Felipe Valandro
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Esta tese é composta por 3 estudos laboratoriais. O estudo 1 avaliou se o material do pistão aplicador de carga (resina epóxi reforçada com fibra de vidro e aço inoxidável) e o diâmetro da ponta deste (6 ou 40 mm) influenciam o comportamento mecânico à fadiga, modo de falha e distribuição de tensões em restaurações simplificadas de cerâmica feldspática. O diâmetro do pistão demonstrou influência no comportamento à fadiga (quanto menor o diâmetro, pior comportamento em fadiga), modo de falha e distribuição de tensões de restaurações simplificadas de cerâmica feldspática. No entanto, a influência do material do pistão só foi observada quando usados pistões de 6 mm de diâmetro (resina epóxi reforçada com fibra de vidro resultam em um melhor comportamento em fadiga do que o aço inoxidável). Considerando que os pistões de 6 mm de diâmetro tendem a subestimar o desempenho à fadiga, eles devem ser evitados em testes de fadiga de restaurações simplificadas de cerâmica feldspática com espessura ≤ 1 mm. O estudo 2 se propôs a caracterizar o efeito da região de contato oclusal no desempenho à fadiga e na região de fratura de coroas monolíticas de dissilicato de lítio considerando 3 regiões de aplicação de carga (restrito às pontas das cúspides; restrito ao plano inclinado das cúspides; associando ponta da cúspide e plano inclinado das cúspides). A aplicação de carga em distintas regiões de contato oclusal afeta o padrão de distribuição de tensão e, consequentemente, o desempenho de fadiga mecânica e a região de fratura das coroas monolíticas de dissilicato de lítio. Uma combinação de carregamento em regiões distintas é recomendada para promover uma melhor avaliação do comportamento à fadiga de um conjunto restaurado. O estudo 3 avaliou o efeito da inclinação das cúspides na superfície oclusal do preparo protético no comportamento à fadiga, modo de falha e distribuição de tensões em laminados oclusais de dissilicato de lítio e de resina composta. Foram considerados três diferentes graus de inclinação das cúspides do substrato (0°, 15° e 30°) e dois tipos de material restaurador (cerâmica de dissilicato de lítio e resina composta). Apesar dos diferentes graus de inclinação dos preparos, as restaurações foram projetadas mantendo 30° de inclinação entre as cúspides na superfície oclusal e 0,7mm de espessura na região de fossa central. Considerando apenas uma perspectiva mecânica, os laminados oclusais em resina composta comportam-se melhor que os de dissilicato de lítio quando as inclinações das cúspides do preparo protético são de 30°, enquanto os laminados oclusais de dissilicato de lítio apresentam melhor performance do que os laminados de resina composta para inclinações de cúspides do preparo protético de 0°. Para uma inclinação de cúspides do preparo protético de 15°, ambos os materiais se comportam de forma semelhante.

Palavras-chave: Análise de falhas. Cerâmicas odontológicas. Estudos laboratoriais. Resina composta.

ABSTRACT

VARIABLES RELATED TO FATIGUE BEHAVIOR AND FAILURE MODE OF CAD/CAM RESTORATIONS

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CO-PROMOTER: Andressa Borin Venturini

This thesis is composed of 3 studies. Study 1 evaluated the influence of the piston material (glass fiber-reinforced epoxy resin or stainless steel) and the piston tip diameter (6 or 40 mm) on the fatigue mechanical behavior, failure mode, and stress distribution of feldspathic ceramic simplified restorations. The piston diameter showed an influence on the fatigue behavior (the smaller the diameter, the worse fatigue behavior), failure mode, and stress distribution of feldspathic ceramic simplified restorations. However, the influence of piston material is only observed when 6 mm diameter pistons are used (glass fiber-reinforced epoxy resin results in better fatigue behavior than stainless steel). Considering that the 6 mm diameter pistons tend to underestimate fatigue performance, they should be avoided in fatigue testing of simplified feldspathic ceramic restorations with a thickness ≤ 1mm. Study 2 set out to characterize the effect of the occlusal contact region on the mechanical fatigue performance and on the fracture region of monolithic lithium disilicate ceramic crowns. Load application on distinct occlusal contact regions affects the stress distribution pattern and consequently the mechanical fatigue performance and fracture region of the monolithic lithium disilicate ceramic crowns. A combination of loading at distinct regions is recommended to promote better evaluation of the fatigue behavior of a restored set. Study 3 evaluated the effects of cusp inclination of the prosthetic preparation’s occlusal surface on the fatigue behavior, failure mode and stress distribution of glass-ceramic and resin composite occlusal veneers. Three different occlusal surface cusp inclination degrees (0°, 15 and 30°) and two type of restorative material (lithium disilicate or resin composite) were considered. Despite different substrate preparation cusp inclination degrees, the restorations were designed maintaining 30° inclination between the cusps at the occlusal surface and a thickness of 0.7mm at the central groove region of the restorations. From a mechanical standpoint, RC occlusal veneers behave better than LD occlusal veneers when the inclinations of the cusps in the prosthetic preparation are 30°. LD occlusal veneers exhibit better mechanical behavior then RC occlusal veneers when the prosthetic preparation cusps have 0° inclinations. When a cusp inclination of 15° is preserved, both restorative materials behave similarly.

Keywords: Failure analysis. Dental ceramics. Laboratory studies. Resin composite.

SUMÁRIO

1 INTRODUÇÃO GERAL

Nas últimas décadas foi possível observar uma grande expansão do uso da tecnologia CAD/CAM (*Computer-Aided Design/Computer-Aided Manufacturing*) na odontologia. Expansão esta impulsionada pelo desenvolvimento e aprimoramento tanto dos sistemas CAD/CAM quanto dos materiais restauradores para CAD/CAM (SUGANNA et al., 2022). Os blocos de materiais restauradores para CAD/CAM são manufaturados industrialmente o que permite a confecção de restaurações dentárias com maior confiabilidade mecânica em relação as restaurações convencionais (restaurações em resina composta direta, restaurações cerâmicas estratificadas ou injetadas), uma vez que o processo industrial de produção dos blocos para CAD/CAM reduz a presença de defeitos e poros no interior do material restaurador (ZHANG; KELLY, 2017). Os diferentes tipos de restaurações dentárias como *inlays*, *onlays*, laminados, coroas totais, próteses parciais fixas, *abutments* de implantes e até reconstruções de arcadas completas podem ser executadas a partir dos materiais e sistemas para CAD/CAM. Além disso, a confecção das restaurações pode ser realizada em laboratórios de prótese dentária ou até mesmo diretamente nos consultórios odontológicos que possuem sistema CAD/CAM (MCLAREN, 2011).

No meio bucal, as restaurações estarão expostas à degradação de suas propriedades mecânicas associadas à fadiga, uma vez que, estão sujeitos à presença de umidade, cargas mastigatórias cíclicas, alterações de temperatura e de pH (DENRY; HOLLOWAY, 2010). Assim, ensaios de fadiga configuram-se importantes ferramentas para avaliar o comportamento a longo prazo dos materiais restauradores (KELLY et al., 2017; ZHANG; LAWN, 2004). Para isso, é importante que os diferentes aspectos a que as restaurações são submetidas na cavidade oral sejam bem reproduzidos pelos ensaios laboratoriais de fadiga na perspectiva de que os resultados possam reproduzir situações e desfechos clínicos (KELLY et al., 2010).

É importante ressaltar que não há padronização na escolha dos parâmetros de ensaio nos experimentos de fadiga, o pistão utilizado para aplicar a carga (material, formato e diâmetro da ponta) é um exemplo de parâmetro que apresenta grande variabilidade entre os estudos (VELHO et al., 2022). Sabe-se que o material, dimensões e formato da ponta do pistão são fatores que podem influenciar no modo de falha e distribuições de tenções durante o ensaio (KELLY, 1999). Embora o aço inoxidável seja o material de pistão mais reportado na literatura (VELHO et al., 2022), ainda não está claro qual material seria o mais adequado. Pistões de resina epóxi, por exemplo, geram uma distribuição de tensões mais uniforme na cerâmica feldspática, atingindo a superfície de tração de maneira mais homogênea e resultando na falha da cerâmica em um período mais curto em comparação aos pistões de aço inoxidável (MIRANDA et al., 2019). Em relação ao formato da ponta do pistão, sabe-se que os pistões de ponta esférica são os mais adequados, uma vez que a distribuição de tensões durante o carregamento se dá de forma mais homogênea em relação aos pistões de ponta plana (KELLY et al., 2010). Para isso, conforme Kelly e colaboradores (1999) o uso de esferas com pelo menos 40 mm de diâmetro seriam ideais para que se consiga reproduzir uma área de contato oclusal clínica. No entanto, apesar destas recomendações, permanece a lacuna na literatura sobre qual o material, formato e dimensões do pistão seriam os mais adequados para aplicação de cargas em fadiga (VELHO et al., 2022).

Ainda na perspectiva de aproximar os ensaios laboratoriais aos cenários clínicos, um outro ponto a ser reproduzido, é a região e o padrão de aplicação de carga. É importante considerar que o padrão de carregamento (distribuição dos contatos oclusais) e que a direção das forças aplicadas irão influenciar o desempenho das restaurações testadas (CORAZZA et al., 2015; ZHANG; SAILER; LAWN, 2013). A forma como as restaurações são carregadas dependerá particularmente do desenho da superfície oclusal e, consequentemente, da quantidade e localização dos contatos oclusais (DITTMER et al., 2011). Embora, a maioria dos estudos laboratoriais utilizem uma única carga no centro da superfície oclusal (VELHO et al., 2022), sabe-se que clinicamente, durante o ciclo mastigatório, os contados oclusais não acontecem de uma forma isolada e em uma única região (CORAZZA et al., 2015). O primeiro contato após as fases preparatória e de esmagamento ocorre em posição excêntrica (ponta da cúspide), seguido de deslizamento até a oclusão cêntrica (fossa central) (DELONG; DOUGLAS, 1983). Assim, a carga axial nas pontas das cúspides e na fossa central também precisa ser considerada quando se pretende aproximar do cenário clínico.

Além das variáveis mencionadas acima (tipo de pistão e região de aplicação de carga), clinicamente, diferentes tipos de restaurações podem ser utilizadas dependendo da necessidade restauradora do elemento dentário. Atualmente, os laminados oclusais têm sido indicados como técnicas restauradoras menos invasivas para dentes que apresentam desgaste oclusal e perda de dimensão vertical em comparação aos tratamentos convencionais com coroas metalocerâmicas ou coroas totalmente cerâmicas (MAGNE et al., 2010; SCHLICHTING et al., 2011). Nesta perspectiva, os laminados oclusais exigem um preparo dentário simples guiado pelo espaço interoclusal gerado pelo desgaste dentário e considerações anatômicas (MAGNE et al., 2010). Materiais como as resinas compostas e as cerâmicas são alternativas para restaurações do tipo laminados oclusais (ALBELASY et al., 2020). No entanto, estes materiais têm propriedades mecânicas diferentes (BELLI et al., 2014), que tendem a influenciar a capacidade de suportar cargas na região posterior (MORIMOTO et al., 2016). Assim, dado que o desenho do preparo dentário (SIROUS et al., 2022) e o tipo de material restaurador (ALBELASY et al., 2020) influenciarão o desempenho mecânico dos laminados oclusais e que diferentes graus de gravidade do desgaste dentário podem ser encontrados clinicamente, o que, consequentemente, determinará o desenho final da restauração necessária para restabelecer a estrutura dentária perdida, esta configura-se como mais uma temática relevante a ser explorada por estudos laboratoriais.

Frente ao exposto anteriormente, explorar variáveis relacionadas ao comportamento em fadiga e modo de falha de restaurações CAD/CAM configura-se como uma temática atual e importante que requer investigação. Neste sentido, a presente tese apresenta-se fragmentada em 3 artigos científicos.

ARTIGO 1: *Effects of material and piston diameter on the fatigue behavior, failure mode, and stress distribution of feldspathic ceramic simplified restorations.* Com objetivo de avaliar se o material do pistão (resina epóxi reforçada com fibra de vidro e aço inoxidável) e o diâmetro da ponta do pistão (6 ou 40 mm) influenciam o comportamento mecânico à fadiga, modo de falha e distribuição de tensões de restaurações simplificadas de cerâmica feldspática.

ARTIGO 2: *How does the occlusal contact region influence the mechanical fatigue performance and fracture region of monolithic lithium disilicate ceramic crowns?* Que se propõe a caracterizar o efeito da região de contato oclusal no desempenho à fadiga mecânica e na localização da região de fratura de coroas monolíticas de dissilicato de lítio, considerando 3 regiões de aplicação de carga (restrito às pontas das cúspides; restrito ao plano inclinado das cúspides; associando ponta de cúspide e plano inclinado das cúspides).

ARTIGO 3: *Fatigue behavior, failure mode and stress distribution of occlusal veneers: influence of the prosthetic preparation cusp inclinations and the type of restorative material.* Que teve como objetivo avaliar o efeito da inclinação das cúspides na superfície oclusal do preparo protético no comportamento à fadiga, modo de falha e distribuição de tensões em laminados oclusais de dissilicato de lítio e de resina composta.

2 ARTIGO 1: EFFECTS OF MATERIAL AND PISTON DIAMETER ON THE FATIGUE BEHAVIOR, FAILURE MODE, AND STRESS DISTRIBUTION OF FELDSPATHIC CERAMIC SIMPLIFIED RESTORATIONS

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Effects of material and piston diameter on the fatigue behavior, failure mode, and stress distribution of feldspathic ceramic simplified restorations

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Abstract

Objective: This study evaluated the influence of the piston material (glass fiber-reinforced epoxy resin or stainless steel) and the piston tip diameter (6 or 40 mm) on the fatigue mechanical behavior, failure mode, and stress distribution of feldspathic ceramic simplified restorations.

Materials and methods: Pistons were machined in glass fiber-reinforced epoxy resin (ER) and in stainless steel (SS), with active tips simulating the curvature radius of 6- or 40-mm diameter spheres. A total of sixty (N= 60) feldspathic ceramic discs (Ø= 10 mm; thickness= 1.0 mm) were adhesively cemented onto supporting substrate discs (Ø= 10 mm; thickness= 2.5 mm) and allocated into 4 groups (n= 15) according to the piston used for fatigue testing: ER_6, ER_40, SS_6, SS_40. Afterward, the specimens were submitted to the stepwise fatigue test (20 Hz frequency; initial load= 100 N; step= 50 N; 10,000 cycles/step, upon specimen failure detection). The collected data were analyzed by two-way ANOVA (α= 0.05) to verify differences by considering 'piston material' and 'piston diameter' as factors, and their association. In addition, a survival analysis (Kaplan Meier with Mantel-Cox log-rank post-hoc tests) was conducted (α= 0.05). Fractographic and finite element (FEA) analyzes were also performed.

Results: 'Piston material' (p= 0.040, F= 4.43) and 'piston diameter' (p< 0.000, F= 563.21) had a significant influence on the fatigue failure load (FFL) and the number of cycles for failure (CFF) values. Feldspathic restorations showed higher FFL and CFF (p< 0.05) when tested with a 40 mm diameter piston compared to a 6 mm diameter piston (ER_40 and SS_40 > ER_6 > SS_6). In relation to the piston material, ER and SS pistons with 40 mm diameter promoted similar fatigue performance (ER_40: 946.67 N/179,333 cycles = SS_40: 936.67 N/177,333 cycles), while 6 mm diameter groups presented different fatigue performance (ER_6: 440 N; 78,000 cycles > SS_6: 353.3 N; 60,667 cycles). Hertzian cone crack failures were only observed in the groups tested with 6 mm pistons, regardless of piston material. Higher stress concentration on the ceramic surface was observed when using 6 mm diameter pistons, whereas the SS_6 group showed a slight increase in stress concentration in comparison to the ER_6 group.

Conclusion: The piston diameter showed an influence on the fatigue behavior, failure mode, and stress distribution of feldspathic ceramic simplified restorations. However, the influence of piston material is only observed when 6 mm diameter pistons are used. Considering that the 6 mm diameter pistons tend to underestimate fatigue performance and to induce Hertzian cone crack failures, they should be avoided in fatigue testing of simplified feldspathic ceramic restorations with a thickness ≤ 1mm.

Keywords: Fatigue test. Glass-ceramic. Loading piston.

Highlights:

- Piston diameter affects the fatigue behavior of feldspathic ceramics.
- Stress distribution on feldspathic ceramic is mainly influenced by the piston diameter.
- 6 mm pistons predispose hertzian cone crack failures in feldspathic ceramics.
- Material has no influence on the fatigue behavior for 40 mm diameter pistons.

1. Introduction

The constant improvements in dental ceramics seen in the last decades have allowed excellent aesthetic properties (translucency, chromatic stability), biocompatibility, chemical inertia, and better mechanical properties (flexural strength, fracture toughness and wear resistance) (Fu et al., 2020). Ceramic materials in the oral environment are exposed to degradation of the mechanical properties associated with cyclic fatigue. Thus, cyclic fatigue tests can be important tools to assess the long-term behavior of ceramic materials (Kelly et al., 2017; Zhang and Lawn, 2004). For this, it is important that the different aspects to which restorations are subjected to in the oral cavity are well reproduced by these fatigue laboratory tests so that the results can reproduce situations and clinical outcomes (Kelly et al., 2010).

However, it is important to emphasize that there is no standardization of the choice of testing parameters in the cyclic fatigue experiments, such as piston material and tip shape and diameter. In fact, the choice of the piston (load applicator) used for mechanical loading presents great variability among studies (Velho et al., 2022). Pistons of different materials, dimensions, and shape tip (spherical or flat) are used in fatigue tests, i.e. fiber-reinforced epoxy resin with 2 or 3 mm diameter flat tip (Kelly et al., 2010; Lodi et al., 2018; May et al., 2015; Venturini et al., 2018a, 2018b), tungsten with 6.25 mm diameter spherical tip (Alessandretti et al., 2020) or stainless steel with 40 mm diameter spherical tip (Guilardi et al., 2020; Venturini et al., 2019).

The material, dimensions, and tip shape of the piston are factors which can influence the failure mode and stress distribution during loading (Kelly, 1999). Regarding the piston material, although stainless steel is the most used material (Velho et al., 2022), it is still not clear which material would be more suitable. Epoxy resin pistons would be able to generate a more homogeneous stress distribution in feldspathic specimens and failures in a shorter period when compared to tungsten and stainless steel pistons (Miranda et al., 2019). In addition, it would present an increased failure load compared to stainless steel, ceramic, and human teeth pistons (Weber et al., 2018). On the other hand, Lorenzoni et al., (2020) observed similar veneered crown fatigue reliability and failure modes when using high stiffness tungsten carbide vs. enamel like steatite piston. Regarding the tip shape of the piston, spherical tip pistons are known to be the most suitable (Kelly et al., 2010). However, the tip dimensions should be able to reproduce the clinical occlusal contact area for which large spheres (> 40 mm) should be used, and this would be possible by machining these large radii onto the end/tip of pistons (Kelly, 1999).

From this viewpoint, the present study aims to evaluate whether the piston material (glass fiber-reinforced epoxy resin and stainless steel) and the piston tip diameter (6 or 40 mm) influence the fatigue mechanical behavior, failure mode, and stress distribution of feldspathic ceramic simplified restorations. The hypothesis tested was that the fatigue behavior, failure mode and stress distribution of simplified feldspathic ceramic restorations would present the worst performance with the stainless-steel piston and the smaller diameter piston.

2. Material and methods

The materials used in this study, their chemical composition and manufacturers are described in Table 1.

2.1 Piston fabrication

Pistons were machined on a precision lathe (Diplomat 3001, Nardini; Americana, Brazil) from glass fiber-reinforced epoxy resin and stainless-steel rods (Table 2), with active tips simulating the curvature radius of 6 mm and 40 mm in diameter spheres (Figure 1), since these piston diameters are commonly reported in a recent scoping review (Velho et al., 2022).

2.2 Simplified ceramic restoration preparation

Simplified three-layer restorations (ceramic, resin cement and glass fiber-reinforced epoxy resin) (Chen et al., 2014) were performed, simulating a posterior tooth restoration with final thickness of 3.5 mm (Sulieman et al., 2005), including a 1 mm of ceramic thickness, commonly used in occlusal veneers (Ladino et al., 2021), and a 2.5 mm thickness of the supporting substrate.

To do so, feldspathic ceramic blocks (Vitablocs Mark II, VITA Zahnfabrik, Bad Sackingen, Germany) were shaped into cylinders using a diamond drill (internal diameter= 10 mm; Diamant Boart, Brussels, Belgium) coupled to a bench drill (SBE 1010 Plus, Metabo; Nürtingen, Germany) under refrigeration. Afterwards, the cylinders were sectioned under water-cooling in a cutting machine (Isomet 1000, Buehler, Lake Bluff, USA), resulting in 60 discs with an initial thickness of 1.1 mm and 10 mm of diameter. Both disc surfaces were ground (EcoMet/AutoMet 250, Buehler) with grit SiC papers (#400, #600 and #1200, 3M;

Sumaré, Brazil), in order to standardize the surfaces, removing possible cutting scratches and defects to obtain a final thickness of 1 mm ± 0.02.

The supporting substrate discs (N= 60) were obtained from 10 mm diameter glass fiber-reinforced epoxy resin rods (Protec, São Paulo, Brazil), which were sectioned under cooling in a cutting machine (Isomet 1000; Buehler) into slices of 2.5 mm thickness. The ceramic discs were cleaned in an ultrasonic bath with isopropyl alcohol (5 min) prior to the surface treatment, while the glass fiber-reinforced epoxy resin discs were cleaned with distilled water (5 min).

The bonding surfaces of the ceramic discs were etched using 5% hydrofluoric acid (Condac Porcelana, FGM, Joinville, Brazil) for 60 s, rinsed with air-water spray for 30 s, and cleaned in an ultrasonic bath with distilled water for 5 min. Afterwards, the coupling agent (Monobond N, Ivoclar, Schaan, Liechtenstein) was applied for 15 s on the treated ceramic surface, reacting for up to 45 s.

The glass fiber-reinforced epoxy resin discs had the cementation surfaces etched with 10% hydrofluoric acid (Condac Porcelana, FGM) for 60 s, rinsed with air-water spray for 30 s, and cleaned in an ultrasonic bath (5min) with distilled water. Then, Primers Multilink A and B (Ivoclar) were mixed at a ratio of 1:1, actively applied to the treated surface (30 s) and air-dried until a thin film of material was obtained.

Each ceramic disc was adhesively cemented to the glass fiber-reinforced epoxy resin disc with resin cement (Multilink N, Ivoclar). A 2.5 N load was applied to the discs to standardize the resin cement thickness. Resin cement excess was removed, and light curing was carried out for five exposures of 20 s (0º, 90º, 180º, 270º and occlusal surface) (1200 mW/cm^2, Radii-Cal, SDI, Bayswater, Australia). All specimens were stored in distilled water (37°C) for at least 24 h up to a maximum period of 7 days before performing the mechanical fatigue tests.

After the cementation procedures, the specimens were randomly allocated (www.randomizer.org) into 4 groups (n= 15) according to the piston material (glass fiber-reinforced epoxy resin or stainless steel) and simulated sphere diameter at the end of the piston (6 or 40 mm) (Table 2).

2.3 Stepwise fatigue test

The specimens were submitted to stepwise fatigue testing in an electrodynamic testing machine (Instron ElectroPuls E3000, InstronCorp, Norwood, USA) according to the piston groups (Table 2) in distilled water at a loading frequency of 20 Hz. The stepwise test parameters adopted were initial load of 100 N, 10,000 cycles per step and a fixed increment of 50 N. Thus, the test was paused every 10,000 cycles and the specimen checked for the presence of failures (radial or hertzian cone cracks) by oblique transillumination (Dibner and Kelly, 2016). In case of no failure, the load was increased by a fixed increment (50 N) and the test continued until failure was detected. The fatigue failure load (FFL) and the number of failure cycles for failure (CFF) at the time of failure were recorded for further statistical analysis.

Glass fiber-reinforced epoxy resin pistons were replaced after testing five specimens in order to avoid changes in the load contact area between the piston and the ceramic surface due to piston tip deformation.

2.4 Finite element analysis (FEA)

Models consisting of ceramic discs, resin cement, fiber-reinforced epoxy resin and pistons with different materials and tested diameters were performed. Tridimensional models were created using a software program (Rhinoceros version 5.0 SR8, McNeel North America, Seattle, USA), while the analysis was performed using the ANSYS CAE software program (ANSYS 19.3, ANSYS Inc., Houston, USA). A static structural analysis was applied according to the fatigue experimental set-up. All materials were considered isotropic, linear, and homogeneous. Young's moduli (GPa) (E) and Poisson's ratios (v) for feldspathic ceramic (E= 48.7 GPa; v= 0.23), fiber-reinforced epoxy resin (E= 14.9 GPa; v= 0.31), resin cement (E= 7.5 GPa, v= 0.3) and stainless steel (E= 190 GPa; v= 0.27) were obtained from previous studies (Kelly et al., 2010; Machry et al., 2021; Ramos et al., 2016). The models for the ER_6 and SS_6 groups were composed of 413,006 tetrahedron (piston) or hexahedron (restoration) solid elements with 101,792 nodes, and the ER_40 and SS_40 groups were composed of 415,926 tetrahedron (piston) or hexahedron (restoration) solid elements with 103,496 nodes. The connections among base, restoration and resin cement were considered perfectly bonded and frictional (0.12) to the piston and restoration. The models were loaded (100 N) at the top of the piston and constrained at the bottom surface of the base. Convergence test was applied increasing the quantity of elements until the mesh do not influence the results at the Center of the disc. The first data was default of the software (12556 elements), after that, in each step we increase about 50% the quantity of elements until the MPS did not variate more than 5% (about

400528 elements). After the coherence and mesh convergence test, the maximum principal stress and minimal principal stress were used as failure criteria to compare the groups.

2.5 Calculation contact radii and contact pressures between piston and ceramic surface

The created contact radii (**a**) between piston and ceramic surface was calculated following relationship described by Lawn (1993):

$$a = \left(\frac{4\kappa Pr}{3E}\right)^{1/3} \qquad (1)$$

In which: **E** is the elastic modulus of the ceramic, **P** the load, **r** the piston radius and **κ** is a dimensionless constant given by:

$$\kappa = \frac{9}{16}\left[(1 - \nu^2) + (1 - \nu_s^2)\frac{E}{E_s}\right] \qquad (2)$$

with $\boldsymbol{\nu}$ and $\boldsymbol{\nu_s}$ being Poisson's ratio for the ceramic and piston and $\mathbf{E_s}$ the modulus of the piston.

The created contact pressures (**p**) between piston and ceramic surface were calculated following relationship described by Lawn (1993):

$$p = \left(\frac{3E}{4\pi\kappa}\right)\left(\frac{a}{r}\right) \qquad (3)$$

In which: a is the contact radius given in Eq. (1), and all other variables have the same meanings as in Eqs. (1) and (2).

2.6 Fractographic analysis

All failed specimens were analyzed in a stereomicroscope (Discovery V20, Carl Zeiss, Gottingen, Germany) and the failures were classified as radial or hertzian cone cracks. Then, the ceramic fragments were detached to expose the fractured surface. Next, one representative specimen of each type of failure was selected and submitted to Scanning Electron Microscopy (SEM, JSM-6360, JEOL, Tokyo, Japan) at 150× and 1000× magnifications to access and illustrate the failure origin and characterize the fractographic features.

2.7 Data analysis

The IBM SPSS software program (IBM, Armonk, USA) was used for statistical analysis. Data were diagnosed as non-normal by the Shapiro-Wilk test ($p < 0.001$) and non-homoscedastic by the Levene test ($p = 0.001$). Two-way ANOVA was performed ($\alpha = 0.05$) to access the influence of the 'piston material' and 'piston diameter' factors, as well as to access the interaction between such factors. Due to non-normal data, bootstrapping procedures (1000 re-samplings; 95% CI, BCa) were implemented to obtain greater reliability of the results, to correct deviations from the normality of the sample distribution and differences among the sizes of the groups, and also to present a 95% CI for differences among means (Haukoos, 2005). Additionally, FFL and CFF data were subjected to a survival analysis using Kaplan Meier and Mantel-Cox (log-rank) post-hoc tests ($\alpha = 0.05$). The FEA and fractographic data were qualitatively analyzed.

3. Results

Two-way ANOVA revealed significant influence of the 'piston material' (FFL: $p = 0.040$, $F = 4.43$; CFF: $p = 0.040$, $F = 4.43$) and 'piston diameter' (FFL: $p < 0.000$, $F = 563.21$; CFF: $p < 0.000$, $F = 563.21$) factors, but not statistically significant for the interaction between factors (FFL: $p = 0.101$, $F = 2.79$; CFF: $p = 0.101$, $F = 2.79$).

FFL and CFF of feldspathic simplified ceramic restorations were influenced by piston diameter, showing higher values when tested with a 40 mm diameter piston; however, the piston material only had an influence in restorations tested with 6 mm diameter pistons (Table 3). The survival analysis showed that specimens tested with 40 mm diameter pistons lasted longer until failure, while SS_6 group samples failed earlier (lower survival rates – Figure 2).

FEA images demonstrated that 40 mm diameter pistons promoted a reduced Maximum Principal Stress and Minimum Principal Stress concentration when compared to 6 mm diameter pistons (Figure 3). Furthermore, it is possible to notice a slight increase in Maximum Principal Stress when the 6 mm stainless steel piston was used when compared to the glass fiber-reinforced epoxy resin.

The piston material and piston diameter directly influenced the radii contact and contact area created between piston and ceramic surface (Table 4). Stainless steel pistons with 6 mm diameter created smaller radii contact and higher contact pressure between piston and ceramic surface, while glass fiber-reinforced epoxy resin pistons with 40 mm diameter created wider radii contact and a reduced contact pressure between piston and ceramic surface. In addition, it was possible to observe an increase in the radii contact and contact pressure between piston and ceramic surface in all groups related to the increase in load applied.

All specimens tested with 40 mm diameter pistons presented radial cracks (failures originated at cementation surface from defects present at the ceramic surface, and then propagated to occlusal surface) (Table 3; Figure 4). On other hand, all specimens tested with 6 mm diameter stainless steel piston presented hertzian cone cracks (cone-shaped crack originating from the loading area - piston contact zone), and then propagated to cementation surface). The specimens tested with 6 mm diameter glass fiber-reinforced epoxy resin piston presented a rate of 60% radial cracks and 40% hertzian cone cracks.

4. Discussion

Different fatigue test setups are adopted by the studies published in the literature, including different types of pistons for load application (Velho et al., 2022). The findings of our study showed that the fatigue mechanical behavior, failure mode, and stress distribution of simplified feldspathic ceramic restorations were influenced by piston diameter and piston material when using 6 mm diameter pistons; however, the piston material factor had no influence when using 40 mm diameter piston. Thus, the tested hypothesis was partially accepted.

Stainless steel and glass fiber-reinforced epoxy resin are materials which present a greater difference of elastic modulus: 14.9 GPa and 190 GPa (Kelly et al., 2010; Machry et al., 2021; Ramos et al., 2016). Thus, a difference in fatigue behavior, failure mode and stress distribution would be expected since a material with a higher elastic modulus would be able to induce higher contact pressure on the ceramic surface (Kelly et al., 2010). However, the effect of piston material was only statistically significant when 6 mm diameter pistons were used (Table 3, Figure 3). Stainless steel pistons showed lower FFL and CFF values, higher prevalence of Hertzian cone-crack failures, differences in the stress concentration according to FEA analysis and higher contact pressure on the ceramic surface (Table 4). Despite the differences in the elastic modulus of materials, the stress distribution on the ceramic surfaces became more homogeneous (Figure 3) and the difference in the elastic modulus between piston materials seems to lose its influence when 40 mm diameter pistons were applied.

Loaded contact area will influence subsurface stresses in ceramics cemented onto supporting substrate (Yi and Kelly, 2008). The contact area between piston and ceramic surface is related to piston diameter (the wider the diameter is, the wider the contact area will be; the smaller the diameter, the smaller the contact area). This fact justifies the lower FFL and CFF values presented by the 6 mm diameter piston groups. Thus, smaller contact areas tend to induce greater contact pressure on the ceramic surface (Kelly, 1999) and consequently higher stress concentration, as shown by the FEA analysis (Figure 3) and Table 4 data, causing failure in a shorter period (Table 3). On the other hand, the wider contact area between piston and ceramic surface created by 40 mm diameter pistons decreased contact pressure on the ceramic surface (Figure 3, Table 4), making the stress distribution more homogeneous.

Another relevant point to note about piston is the tip shape. Spherical tip pistons were used in the present study, which were different from 3 mm diameter flat tip pistons used by Weber et al. (2021, 2018). The main negative point of using flat tip pistons is the edge-loading that occurs when the ceramic deforms slightly beneath the piston (Kelly, 1999). This can be avoided by using spherical tip pistons. However, it is important that these tip spheres are able to simulate an occlusal contact facet which has been reported to be to the order of 1 to 4 mm in diameter (Piotrowski et al., 2001; Woda et al., 1987). From this standpoint, spheres bigger than 40 mm diameter are recommended (Kelly, 1999). According to our results, only 40 mm diameter pistons would approach the occlusal contact facet (Table 4). Thus, the large difference between 6 mm and 40 mm diameter pistons indicates that FFL and CFF values are underestimated when using 6 mm diameter pistons.

Cracks originating from the cementation surface (radial cracks) for the test geometry adopted in our study (simplified three-layer restorations) which simulates a posterior tooth restoration (Chen et al., 2014) are a clinically relevant failure mode (Kelly, 1999; Kelly et al., 2010; Yi and Kelly, 2008). This failure mode was observed in all specimens tested with 40 mm diameter pistons, regardless of piston material (Table 3). On the opposite side, Hertzian cone crack failures (cone-shaped crack originating from the loading area – piston contact zone) were more prevalent in specimens tested with 6 mm diameter pistons (100% - SS_6 and 40% - ER_6; Table 3), corroborating fatigue data (SS_6 < ER_6). Hertzian cone crack failures are associated with surface contact damage caused by increased contact pressure induced by smaller diameter pistons (Kelly, 1999). This behavior is corroborated by the FEA analysis (Figure 3), in which a higher compression stress concentration in the ceramic occlusal surface was observed for groups of 6 mm diameter pistons. However, Hertzian cone crack failures are not often observed in clinical failures (Kelly, 1999; Kelly et al., 2010; Yi and Kelly, 2008). Therefore, 6 mm diameter pistons are not appropriate for testing simplified three-layer restorations. However, glass fiber-reinforced epoxy resin pistons appear to be most suitable when the use of 6 mm diameter pistons is required (i.e., anatomical restorations), as they induce a lower percentage of Hertzian cone crack failures, even though the glass fiber-reinforced epoxy resin pistons with 40 mm work better mechanically.

Finally, we have to state that 40 mm diameter pistons seem to be the best choice for fatigue tests of simplified three-layer restorations using axial loading. As no differences were found in the FFL and CFF values between the piston materials (glass fiber-reinforced epoxy resin and stainless steel), the 40 mm diameter stainless steel piston would be a more viable alternative when considering the ease of machining and durability, with no need for replacement during the test. It is important to note that although simplified three-layer restorations are an alternative consolidated in the literature for simulating posterior restorations (Chen et al., 2014; Velho et al., 2022). However, distinct behaviors might be observed when more complex scenarios, including anatomical geometries, bilayer restorations and sliding loading, are evaluated. Thus, the effect of the material and piston diameter adopted for the fatigue testing and its related outcomes might be different in other scenarios.

5. Conclusion

- The piston diameter influences the fatigue mechanical behavior, failure mode, and stress distribution of feldspathic ceramic simplified restorations. However, the piston material presents an influence on the fatigue mechanical behavior, failure mode, and stress distribution of feldspathic ceramic simplified restorations only when 6 mm diameter pistons are used.

- Considering that the 6 mm diameter pistons tend to underestimate fatigue performance and create hertzian cone crack failures, its use should be avoided in fatigue testing of simplified feldspathic ceramic restorations with a thickness ≤ 1mm.

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TABLES

Table 1. Description of materials, commercial name, manufacturer, composition and batch number.

Material	Commercial name/manufacturer	Composition	Batch number
Feldspathic Ceramic	Vita Mark II, VITA Zahnfabrik, Bad Sackingen, Germany	SiO_2 56–64 wt%, Al_2O_3 20–23 wt%, K_2O 6–8 wt%, Na_2O 6–9 wt%, other and coloring oxide 0–0.6 wt%	45950
Ceramic primer coupling agent	Monobond N, Ivoclar, Schaan, Liechtenstein	Alcohol solution of silane methacrylate, phosphoric acid methacrylate and sulphide methacrylate.	Y19262
5% hydrofluoric acid	Condac Porcelana, FGM, Joinville, Brazil	< 5% hydrofluoric acid.	020819
10% hydrofluoric acid	Condac Porcelana, FGM	< 10% hydrofluoric acid.	211019
Dual cure resin cement	Multilink N, Ivoclar	Silicate glass, ytterbium trifluoride, highly dispersed silica, catalysts and stabilizer, pigments.	Y26001
Primer	Multilink Primer (A and B), Ivoclar	Primer A: water, initiators; primer B: phosphonic acid acrylate, hydroxyethyl methacrylate, methacrylate mod. polyacrylic acid, stabilizer.	Primer A: Y25800 Primer B: Y34727

*The chemical composition is described according to the manufacturers' information.

Table 2. Experimental design (n= 15).

Group	Piston material	Material description	Piston tip diameter (mm)
ER_6	Glass fiber-reinforced epoxy resin (ER)	Continuous filament woven fiberglass bonded with epoxy resin. E= 14.9 GPa; v= 0.31	6
ER_40			40
SS_6	Stainless steel (SS)	304 Stainless Steel – C 0.08%, C 1%, Mn 2%, P 0.045%, S0.030%, Cr 18-20%, Ni 8-11% E= 190 GPa; v= 0.27	6
SS_40			40

E: Young's moduli (GPa), v: Poisson's ratios

Table 3. Mean fatigue failure loads (FFL) in Newtons, number of cycles for failure (CFF) with 95% confidence intervals (CI) and failure mode of each evaluated condition.

Groups	FFL	CFF	Failure mode	
	Mean (CI)*	Mean (CI)*	Radial cracks	Hertzian cone cracks
ER_6	440.00 (410.99 – 469.01) ^{B}	78,000 (72,198 – 83,802) ^{B}	9 (60%)	6 (40%)
ER_40	946.67 (871.69 – 1021.65) ^{A}	179,333 (164,337 – 194,329) ^{A}	15 (100%)	0 (0%)
SS_6	353.33 (333.12 – 373.55) ^{C}	60,667 (54,036 – 65,964) ^{C}	0 (0%)	15 (100%)
SS_40	936.67 (901.57 – 971.76) ^{A}	177,333 (170,314 – 184,353) ^{A}	15 (100%)	0 (0%)

* Different letters indicate statistical differences on each column for each considered outcome considering Kaplan-Meier and Mantel-Cox (log-rank) tests.

Table 4. Contact radii (a) in mm and contact pressure (p) in MPa between piston and ceramic surface. The values were calculated considering an applied loaf of 100 N and the mean of FFL of each group.

Groups	100N		Mean FFL	
	a	p	a	p
ER_6	0.26	463.02	0.43	758.72
ER_40	0.49	130.71	1.04	276.45
SS_6	0.18	1024.87	0.27	1560.48
SS_40	0.33	289.33	0.70	609.75

FIGURES

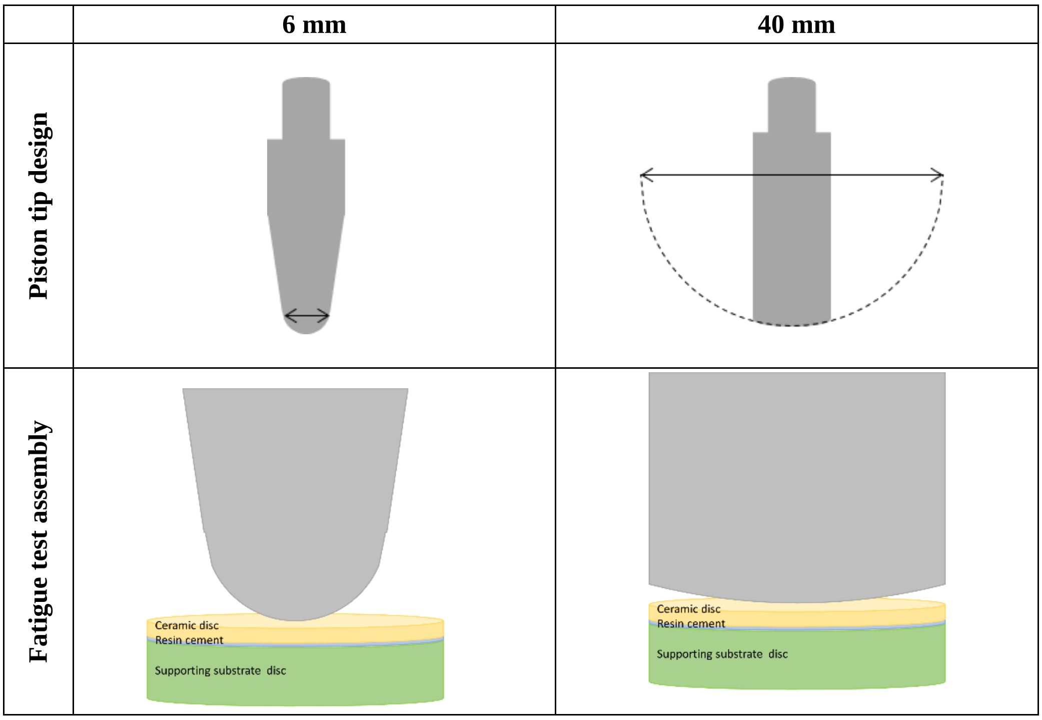


Figure 1. Illustrations of pistons and fatigue test assembly.

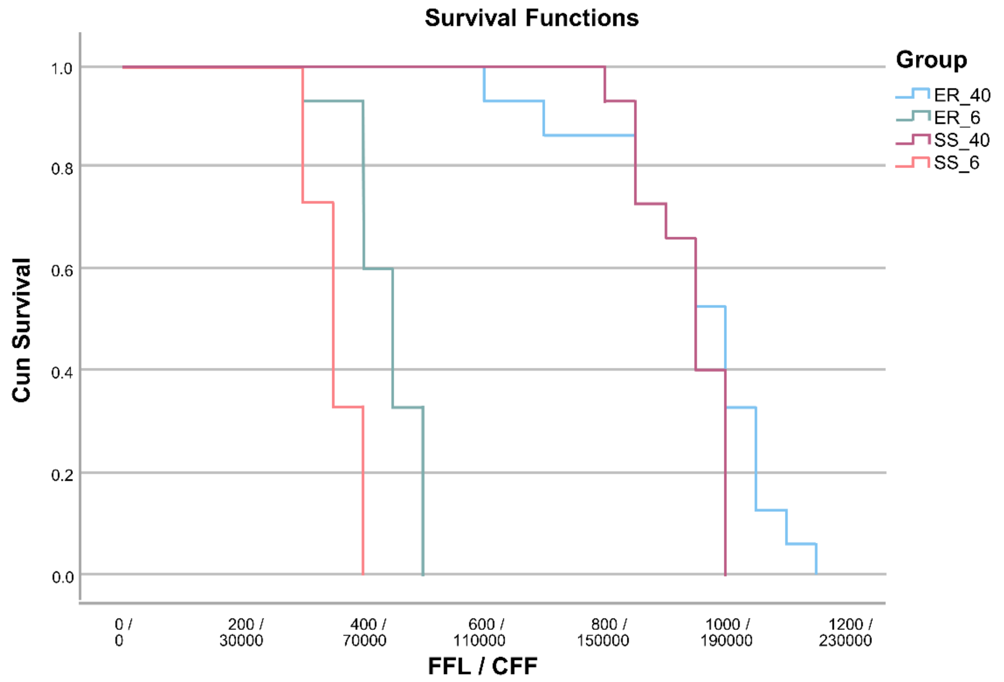


Figure 2. Survival rates according to the steps of FFL and CFF in which each disc failed, obtained by Kaplan–Meier and Log-rank tests.

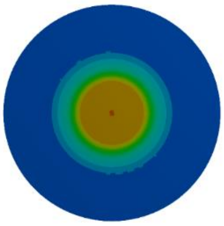
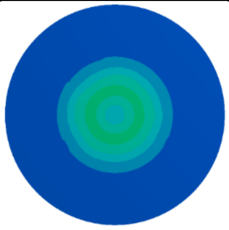
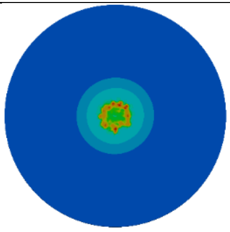
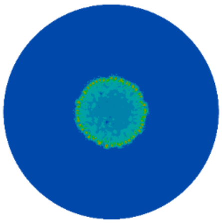
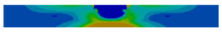
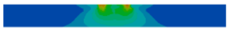
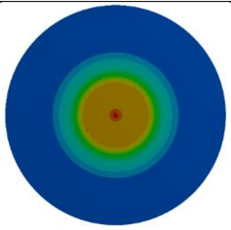
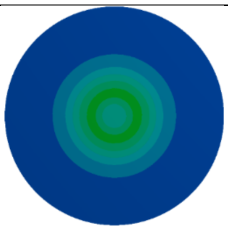
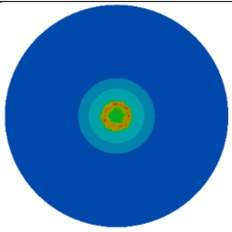
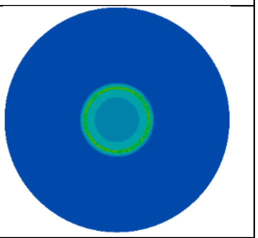
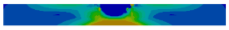
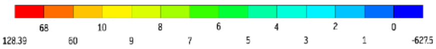
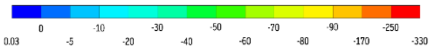
Material	**Diameter**		**Diameter**	
	6 mm	**40 mm**	**6 mm**	**40 mm**
Glass fiber-reinforced epoxy resin				
				
Stainless steel				
				
Scale				

Figure 3 - FEA analysis demonstrates the tensile stress in the inner surface of the restorations (Maximum Principal Stress) and compression stress in the occlusal surface of restorations (Minimum Principal Stress). The stress concentration on the inner surface or occlusal surface of ceramic discs decreased when 40 mm diameter pistons were used under the same applied load.

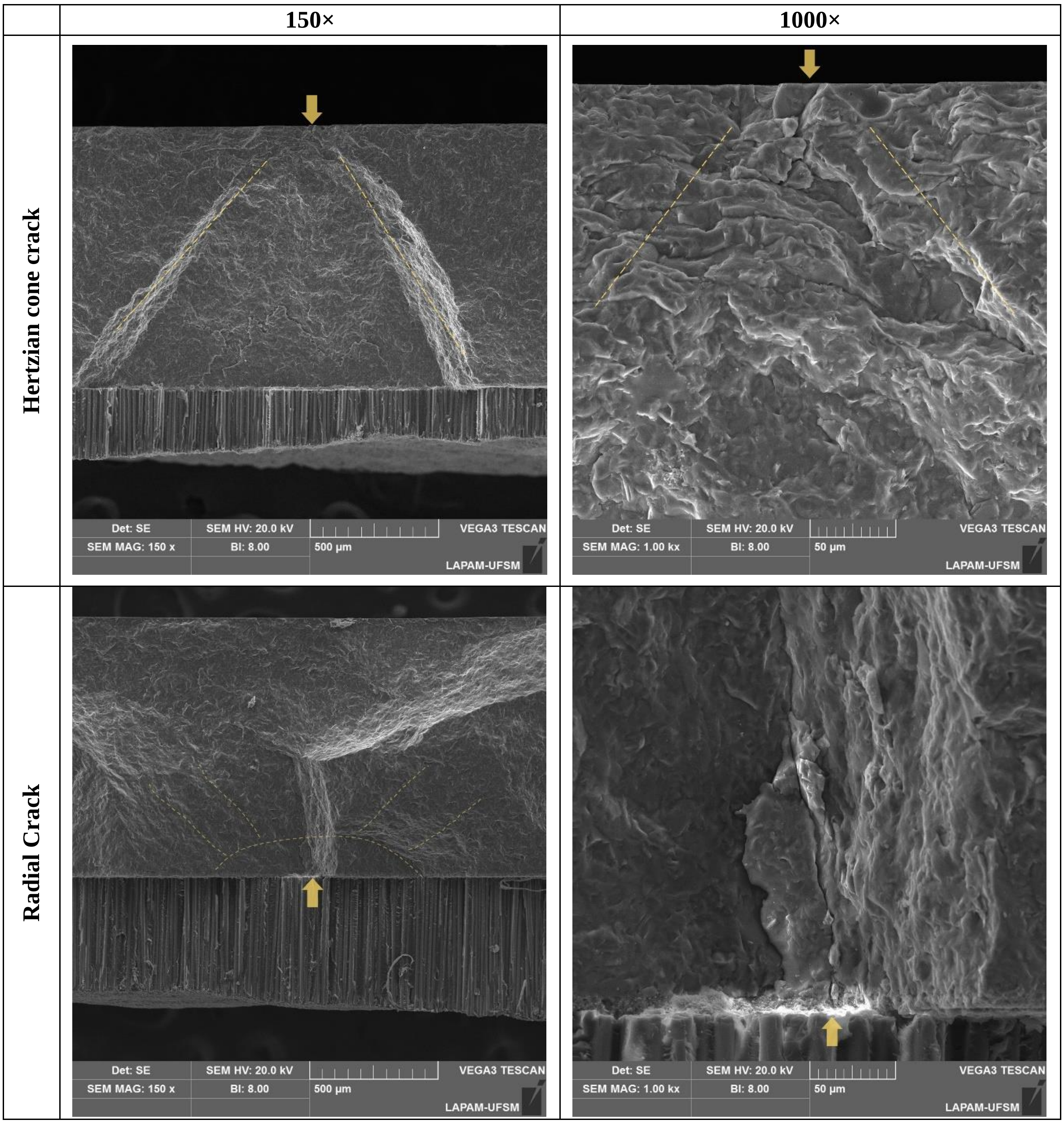


Figure 4 - Representative SEM images (150× and 1000×). **Hertzian cone crack** - cone-shaped crack originating from the loading area (piston contact zone), pointed by yellow arrows, and then propagated to the opposite side (cementation surface). The dashed lines demonstrate the propagation of the failure. **Radial crack** - failures originated at cementation surface from defects present at the ceramic surface, pointed out by yellow arrows, and then propagated to the opposite side (occlusal/top surface).

3 ARTIGO 2: HOW DOES THE OCCLUSAL CONTACT REGION INFLUENCE THE MECHANICAL FATIGUE PERFORMANCE AND FRACTURE REGION OF MONOLITHIC LITHIUM DISILICATE CERAMIC CROWNS?

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How does the occlusal contact region influence the mechanical fatigue performance and fracture region of monolithic lithium disilicate ceramic crowns?

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Abstract

Objective: To characterize the effect of the occlusal contact region on the mechanical fatigue performance and on the fracture region of monolithic lithium disilicate ceramic crowns.

Materials and methods: Monolithic lithium disilicate ceramic crowns were machined in a CAD/CAM system and adhesively luted onto glass-fiber reinforced epoxy resin preparations with resin cement. The crowns were divided into three groups (n= 16) according to load application region (cusp tip: restricted to cusp tips; cusp plane: restricted to cuspal inclined plane; or mixed: associating tip cusp and cuspal inclined plane). The specimens were submitted to a cyclic fatigue test (initial load: 200 N; step-size: 100 N; cycles/step: 20,000; loading frequency: 20 Hz; load applicator: 6mm or 40mm diameter stainless steel) until observing cracks (1st outcome) and fracture (2nd outcome). The data were analyzed by the Kaplan-Meier + Mantel-Cox post-hoc tests for both outcomes (cracks and fracture). Finite element analysis (FEA), occlusal contact region, contact radii measurements, and fractographic analyzes were performed.

Results: The mixed group presented worse fatigue mechanical behavior (550N / 85.000 cycles) compared to the cuspal inclined plane group (656N / 111,250 cycles) ($p< 0.05$) for the first crack outcome, while the cusp tip group was similar to both groups (588N / 97,500 cycles) ($p> 0.05$). The mixed group had the worst fatigue behavior (1413N / 253,029 cycles) in relation to the other groups (Cusp tip: 1644N / 293,312 cycles; Cuspal inclined plane: 1631N / 295,174 cycles) considering the crown fracture outcome ($p< 0.05$). FEA showed higher tensile stress concentration areas just below the load application region. In addition, loading on the cuspal inclined plane induced a higher tensile stress concentration in the groove region. The most prevalent type of crown fracture was the wall fracture. Groove fracture was observed in 50% of the loading specimens exclusively on the cuspal inclined plane.

Conclusion: Load application on distinct occlusal contact regions affects the stress distribution pattern and consequently the mechanical fatigue performance and fracture region of the monolithic lithium disilicate ceramic crowns. A combination of loading at distinct regions is recommended to promote better evaluation of the fatigue behavior of a restored set.

Keywords: Dental ceramics. Fatigue. Fracture. Loading pattern.

Highlights

- The loading pattern influences the fatigue performance of lithium disilicate crowns.
- The fracture region of monolithic crowns changes according to the adopted loading pattern.
- The combination of occlusal loading at distinct regions leads to better evaluation of the fatigue behavior of a restored set.

1. Introduction

Improvements in computer-aided design and manufacturing (CAD/CAM) technology and ceramic materials in recent decades have expanded the use of monolithic crowns in prosthetic dentistry (Bacchi and Cesar, 2022). CAD/CAM materials present less incorporation of pores and flaws within the material, which results in greater reliability and lower risk of clinical failure compared to traditional porcelain-veneered crowns (Belli et al., 2017; Zhang and Kelly, 2017). However, they still have limitations such as brittleness and are prone to mechanical failures that can lead to catastrophic fractures and chipping (Della Bona and Kelly, 2008; Mazza et al., 2022; Zahran et al., 2008).

An important factor to decrease the risk of failure in ceramic restorations is adequate occlusion between natural teeth, restorations and/or fixed prostheses, as inadequate occlusal contacts can generate stress concentration areas which consequently tend to lead to failures (Amin et al., 2019). Therefore, it is important to consider that the loading pattern (distribution of occlusal contacts) and the direction of applied forces may influence the performance of ceramic restorations (Corazza et al., 2015; Zhang et al., 2013). The way in which restorations are loaded will particularly depend on the design of the occlusal surface and consequently on the amount and location of occlusal contacts (Dittmer et al., 2011).

The first contact after the preparatory and crushing phases during the masticatory cycle occur in an eccentric position (cusp tip), followed by sliding until centric occlusion (central fossa) (DeLong and Douglas, 1983). Occlusal contact in the central fossa region is more stable and better distributes stress; on the other hand, contact in the cusp tip generates high stress at the contact area, where it contributes to the origin of failures (Corazza et al., 2015; Dittmer et al., 2011). However, most laboratory studies use a single load at the center of the occlusal surface (Velho et al., 2022b). Thus, axial loading on the cusp tips and central fossa also needs to be considered when aiming to approximate to the clinical scenario.

In view of the above, evaluating distinct in vitro occlusal contact modes which can reproduce or approximate clinically observed failure modes in ceramic monolithic crowns becomes a relevant topic. Therefore, this study aimed to characterize the effect of the occlusal contact region on the mechanical fatigue performance and on the fracture region of monolithic ceramic crowns considering 3 load application regions (restricted to cusp tips; restricted to cuspal inclined plane; associating tip cusp and cuspal inclined plane). The hypothesis tested was that the occlusal loading region influences the mechanical fatigue performance and the fracture region of monolithic lithium disilicate ceramic crowns.

2. Material and methods

2.1 Materials and study design

The materials used in this study, their chemical composition, manufacturers and batch number are described in Table 1. The study design is seen in Table 2, which shows that this study was composed of three experimental groups (n= 16), assigned taking into consideration the region of load application factor.

2.2 Specimen preparation

Glass-fiber reinforced epoxy resin bars (Perfil Pultrudado F, Protec, São Paulo, Brazil) were machined in a precision lathe (Diplomat 3001, Nardini, Americana, Brazil) to simulate a simplified posterior tooth prosthetic preparation (N= 48) with the following characteristics: rounded internal angles, 5.32 mm height, 16° occlusal convergence, 30° convergence between the cusps, and 1.0 mm thickness along its entire length (Figure 1).

Next, a randomized glass-fiber reinforced epoxy resin preparation was scanned to prepare the ceramic crown (AutoScan-DS-EX Pro, Shining 3D Tech. Co., Ltd., Zhejiang, China). In sequence, a simplified monolithic molar crown was designed using a computer software program (Exocad, Exocad GmbH, Darmstadt, Germany), considering a cementation space of 50 µm and 1.0 mm of final occlusal and axial thickness (Figure 2). Finally, the ceramic crowns (N= 48) were machined on a milling machine (CORiTEC 250i SERIES, Imes-icore GmbH, Eiterfeld, Germany) from lithium disilicate glass-ceramic CAD/CAM blocks (IPS e.max CAD, Ivoclar, Schaan, Liechtenstein). Each machined ceramic crown was seated in its respective glass-fiber reinforced epoxy resin preparation to assess the marginal fit.

The crowns were polished on the external surface using a multi-step finishing and polishing system (OptraFine, Ivoclar) coupled to an electric motor (Perfecta 300, W&H Dentalwerk Bürmoos GmbH, Bürmoos, Austria). The procedure was carried out in three steps: finishing (OptraFine F, Ivoclar), polishing (OptraFine P, Ivoclar) and high-gloss polishing with nylon brushes (OptraFine HP, Ivoclar) with diamond polishing paste. The crowns were subsequently cleaned in an ultrasonic bath in distilled water (1440 D, Odontobras Medical and Dental Equipment, Ltda., Ribeirão Preto, Brazil) for 5 min.

Finally, the crowns were fixed on crystallization pins (IPS e.max CAD Crystallization Pin, Ivoclar) using a crystallization paste (IPS Object Fix Flow, Ivoclar) applied inside the crown and the crowns were crystallized according to the manufacturer’s instructions (840°C, 7 min vacuum, Vacumat 6000 MP, VITA Zahnfabrik, Bad Säckingen, Germany).

2.3 Bonding procedures

Before the surface treatment, the ceramic crowns and glass-fiber reinforced epoxy resin preparations were cleaned in an ultrasonic bath (1440 D, Odontobras Medical and Dental Equipment, Ltda) for 5 min using 78% isopropyl alcohol and distilled water, respectively.

The bonding surfaces of ceramic crowns were etched with 5% hydrofluoric acid (Condac Porcelana, FGM, Joinville, Brazil) for 20 s, rinsed with air-water spray for 30 s, and air-dried. Then, the specimens were cleaned in an ultrasonic bath with distilled water for 5 min. In sequence, a layer of a silane-containing universal primer (Monobond N, Ivoclar) was applied on the etched ceramic surface for 15 s and kept untouched for 45 s.

The glass-fiber reinforced epoxy resin preparations were etched with 10% hydrofluoric acid (Condac Porcelana, FGM) for 60 s, rinsed with air-water spray for 30 s, and air-dried. Then, the specimens were cleaned in an ultrasonic bath with distilled water for 5 min. Multilink N Primer A and Multilink N Primer B were subsequently mixed in a 1:1 ratio and applied for 30 s actively and air-dried to obtain a thin and uniform layer.

Each ceramic crown was bonded to its respective glass-fiber reinforced epoxy resin preparation using a resin cement (Multilink N, Ivoclar). After the resin cement application onto the ceramic crown bonding surface, each ceramic crown was placed over the corresponding glass-fiber reinforced epoxy resin preparation under a constant load (7.5 N) on the occlusal surface of the ceramic crown. Then, the excess resin cement was removed, and the set was light-cured (1200 mW/cm², Radii-cal LED curing light, SDI, Bayswater, Australia) for five exposures of 20 s around the specimen (buccal, mesial, lingual, distal, and occlusal surfaces). After the bonding procedures, the specimens were randomly allocated (www.randomizer.org) into three experimental groups (Table 2).

2.4 Cyclic fatigue tests

The specimens were subjected to the cyclic fatigue test in an electrodynamic testing machine (Instron ElectroPuls E3000, InstronCorp, Norwood, USA). To do so, the following test parameters were adopted: initial load of 200 N, load increment of 100 N, number of cycles per step and load applicator (6mm or 40mm diameter stainless steel) (Velho et al., 2022a), as described in Table 2 and Figure 3 respectively, as well as a loading frequency of 20 Hz (Velho et al., 2020) and immersion in distilled water.

The fatigue test was paused at two times: (1) at the end of the number of cycles defined for the step, when the specimen was verified for the presence of the first crack by oblique transillumination (Dibner and Kelly, 2016); or (2) when the machine displacement limit was tripped (crown fracture). In the absence of any of the outcomes, the test was resumed with the load increased by a load increment. If first cracks were detected, the fatigue failure load (FFL) and the number of cycles for failure (CFF) were recorded, and the test continued until the crown fractured, and then the FFL and CFF values were re-registered.

2.5 Finite element analysis (FEA)

Models of ceramic crowns, resin cement, glass-fiber reinforced epoxy resin preparations and load applicators were performed. Three-dimensional models were created using a software program (Rhinoceros version 5.0 SR8, McNeel North America, Seattle, USA), and analyzed using the ANSYS CAE software program (ANSYS 19.3, ANSYS Inc., Houston, USA). A static structural analysis was applied according to the fatigue test assembly of the cusp tip and cusp plane groups (Figure 3). All materials were considered isotropic, linear, and homogeneous. Young's moduli (GPa) and Poisson's ratios for lithium disilicate ceramic (E= 95; Gpa; ν= 0.25), glass-fiber reinforced epoxy resin (E= 14.9; GPa; ν= 0.31), resin cement (E = 7.5; GPa, ν= 0.3) and stainless steel (E= 190; GPa; ν= 0.27) were obtained from previous studies (Kelly et al., 2010; Machry et al., 2021; Ramos et al., 2016). The cusp tip group model was composed of 158,312 tetrahedron solid elements with 233,766 nodes and the cusp plane group was composed of 156,189 tetrahedron solid elements with 230,228 nodes. The connections among the base, restoration and resin cement were considered perfectly bonded and frictional (0.12) to the load applicator and restoration. The models were loaded (100 N) at the top of the load applicator and constrained at the bottom surface of the base. After the coherence and mesh convergence test, the maximum principal stress was used as the failure criteria to compare the groups.

2.6 Contact point region and contact radii measurements

In aiming to determine the contact point region for the two loading patterns adopted before the fatigue test, all specimens had their contact point between the load applicator and the ceramic crown highlighted using a carbon paper (Contacto paper, 100 µm, Angelus, Londrina, Brazil) and a compressive load application of 10 N was applied in the testing machine. Then, the contact radii were measured in a stereomicroscope (Discovery V20, Carl Zeiss, Gottingen, Germany), and the mean values were calculated.

2.7 Fractographic analysis

Two failures modes were considered: first crack (initial crack, identified by transillumination) and crown fracture (final failure, involving crown breaking into one or more fragments). In addition, the crown fractures were classified as wall fracture (when displacement of a fragment of the crown wall occurred) or groove fracture (when the failure involved fracture in the central groove region of the crown and cohesive failure of the substrate). Next, one representative specimen of each type of failure was selected and submitted to Scanning Electron Microscopy (SEM, JSM-6360, JEOL, Tokyo, Japan) at 30× and 100× magnifications to access and illustrate the failure origin and characterize the fractographic features.

2.8 Data analysis

The FFL and CFF data for both outcomes (first crack and crown fracture) were statistically analyzed through a survival analysis using Kaplan Meier and Mantel-Cox (log-rank) post-hoc tests ($\alpha = 0.05$) in the IBM SPSS software program (IBM, Armonk, USA). The FEA, contact point measurements and fractographic data were qualitatively analyzed.

3. Results

The mixed loading group presented lower fatigue behavior considering the first crack outcome in relation to the cuspal inclined plane group, while the cusp tip group was similar to both groups (Table 3, Figure 4). In addition, the mixed group showed the worst fatigue behavior regarding the crown fracture outcome compared to the other groups (Table 3, Figure 4).

FEA data showed higher tensile stress concentrations areas just below the load application region. In addition, loading on the cuspal inclined plane induced a higher tensile stress concentration in the central groove region compared to cusp tip loading (Figure 5). The occlusal contact point regions as well as the mean values of the contact radii are shown in Figure 6.

The most prevalent type of crown fracture was the wall fracture (Table 3). The fractographic analysis (Figure 7) showed the failure starting from the bonding surface as a radial crack and then the failure also starts to propagate with increasing load from the occlusal surface (hertzian-cone crack) towards the bonding surface until reaching the radial crack (the fracture moment). On the other hand, the groove fracture occurred in 50% of the loading specimens exclusively on the cuspal inclined plane. These failures involved the displacement of ceramic and supporting substrate in the groove region. It is not possible to identify an apparent failure origin in the ceramic – the failure possibly occurred from the shear rupture of the supporting substrate (Figure 7).

4. Discussion

Overall, this study showed that load application at distinct occlusal contact regions affected the stress distribution pattern owing to different tip sizes, and consequently the mechanical fatigue performance and fracture region, since the group with combined loading induced the worst behavior. Thus, the assumed hypothesis was accepted.

In addition, worse mechanical fatigue performance was observed considering the loading associating the cusp tip and cuspal inclined plane contacts (mixed group) (Table 3, Figure 4). This can be associated with the loading pattern adopted, which consequently generated tensile stress concentration in two regions (cusp tip and cuspal inclined plane – Figure 5), contributing to a faster fatigue mechanical degradation in relation to the other groups. Furthermore, the predominant failure mode was wall fracture (Table 3), which is similar to the total fractures of clinical crowns reported by Skjold et al. (2022). From the fractographic analysis (Figure 7), it is possible to observe that the failure happened from radial and hertzian-cone cracks, which are competing failure modes (Deng et al., 2002; Lawn et al., 2004; Zhang et al., 2009). A radial crack initially forms and progresses towards the occlusal surface (Zhang et al., 2009). As the applied load and number of cycles increase, contact damage to the occlusal surface tends to occur (Zhang et al., 2005), which contributes to the formation of hertzian-cone cracks.

On the other hand, the groups with exclusive loading in one region (cusp tip or cuspal inclined plane) presented similar mechanical fatigue behavior to each other and performed better than the mixed loading group (Table 3, Figure 4) for the crown fracture outcome. The main difference between these two groups was the failure mode observed. While all failures in the cusp tip loading group were wall fractures, half of the specimens presented fractures in the central groove region when considering the cuspal inclined plane group (Table 3). This may be associated with a higher tensile stress concentration in the central groove region generated by inclined cuspal plane loading compared to the loading restricted to the cusp tips, as demonstrated by the FEA (Figure 5). Furthermore, the present results are similar to those shown by Shahmoradi et al. (2020) when adopting loading in the cuspal inclined plane.

The occlusal region (intaglio surface) (Kelly et al., 2010) and the crown margin (Øilo et al., 2014; Skjold et al., 2022) stand out among the failure origin regions of monolithic crowns;

however, the failure origin region of the restorations in our study was the occlusal surface, as can be seen in the fractographic analysis (Figure 7). This fracture pattern was similar to that found by Yamaguchi et al., (2020) and it is associated with the higher tensile stress concentration in the occlusal region (Figure 5) generated by the adopted loading pattern. Although exclusive loading at the cusp tip generated subtle stress concentration in the crown margin region (Figure 5), this would not have been enough to initiate a fracture. In addition, the thickness in the margin region of the crowns used in this study was greater than that commonly reported in clinical crowns (Skjold et al., 2022), which can contribute to strengthening in the region.

The occlusal contact regions adopted in this study were the cusp tips (simulating a cusp-cusp contact) and the cuspal inclined plane (simulating a cusp-fossa contact), and the fatigue mechanical behavior was similar in both scenarios (Table 3). However, these contacts do not happen in an isolated way during the masticatory cycle (Corazza et al., 2015). The first contact after the preparatory and the crushing phases occur in an eccentric position (cusp tip), followed by sliding until centric occlusion (central fossa) (DeLong and Douglas, 1983). Thus, the present study used loading associating tip cusp and cuspal inclined plane in aiming to approach this masticatory cycle. In this sense, considering that the mechanical fatigue behavior of this group was worse (Table 3), the loading being restricted to only one region may overestimate the mechanical behavior of the restoration. However, the mixed scenario explored herein should not be considered as a perfect simulation to what is seen in clinical function, since sliding movements are not reproduced.

The specimen model used in this study was designed to increase the complexity of the model in relation to simplified non-anatomic crowns (occlusal plane) (Velho et al., 2022b). Therefore, restoration design parameters – such as the thickness, preparation height, margin geometry, cusp inclination, presence of surface irregularities and sharp features of the occlusal anatomy – can influence the restoration's mechanical behavior (Shahmoradi et al., 2020; Sornsuwan and Swain, 2011). While the model still enables isolating factors such as ceramic thickness, the loading pattern and the stress distribution in the restoration, it tends to be more complex in relation to simplified non-anatomic crowns. However, there are still limitations such as the origin of failure restricted to the occlusal surface and the absence of sliding movements. Therefore, the data must be interpreted with caution. New studies proposing changes in the model of restorations in which the load is taken towards the crown margin are suggested, so that the model can better approximate what happens clinically. Lastly, sliding movements approaching the occlusal surface are encouraged.

5. Conclusion

Load application at the distinct occlusal contact regions influenced the stress distribution pattern, and consequently the mechanical fatigue performance and the fracture region of monolithic lithium disilicate crowns. Although there are limitations in relation to simulating the clinical loading pattern, the association of *in vitro* loading regions is recommended to promote better evaluation of the fatigue behavior of a restored set.

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TABLES

Table 1. Description of materials, commercial name, manufacturer, composition, and batch number.

Material	**Commercial name/manufacturer**	**Composition***	**Batch number**
Lithium disilicate glass ceramic	IPS e.max CAD, Ivoclar, Schaan, Liechtenstein	SiO_2 57–80 %wt, Li_2O 11–19 %wt, K_2O 0–13 %wt, P_2O_5 0–11 %wt, ZrO_2 0–8 %wt, ZnO 0–8 %wt, Al_2O_3 0–5 %wt, MgO 0–5 %wt, colouring oxides 0–8 %wt.	X27104
Silane-containing universal primer	Monobond N, Ivoclar	Alcohol solution of silane methacrylate, phosphoric acid methacrylate and sulphide methacrylate.	Y29207
5% hydrofluoric acid	Condac Porcelana, FGM, Joinville, Brazil	< 5% hydrofluoric acid.	020819
10% hydrofluoric acid	Condac Porcelana, FGM	< 10% hydrofluoric acid.	220920
Dual cure resin cement	Multilink N, Ivoclar	Base: Dimethacrylates and HEMA 33.1 %wt, Barium glass filler 37.4 %wt, ytterbium trifluoride, 23 %wt, highly dispersed silica 5.4 %wt, catalysts and stabilizer 1 %wt, pigments <0.03 %wt	Y26001
		Catalyst: Dimethacrylates and HEMA 32.4 %wt, Barium glass filler 37.4 %wt, ytterbium trifluoride, 23 %wt, highly dispersed silica 5.4 %wt, catalysts and stabilizer 1.8 %wt	
Primer	Multilink Primer (A and B), Ivoclar	Primer A: water 85.7 %wt, initiators 14.3 %wt.	Z000M9
		Primer B: phosphonic acid acrylate 48.1 %wt, (hydroxyethyl methacrylate, methacrylate mod. polyacrylic acid) 51.9 %wt, stabilizer <0.02 %wt.	Z003PX
Glass-fiber reinforced epoxy resin	Perfil Pultrudado F, Protec, São Paulo, Brazil	Continuous filament woven fiberglass bonded with epoxy resin.	-

*The chemical composition is described according to the manufacturers' information.
%wt - % in weight

Table 2. Experimental design (n= 16).

Group Code	**Load application region**	**Number of cycles per step for fatigue testing**	**Load applicator**
Cusp tip	Restricted to cusp tips	20,000	40 mm diameter stainless steel
Cusp plane	Restricted to inclined cuspal plane	20,000	6 mm diameter stainless steel
Mixed	Cusp tips and inclined cuspal plane	10,000 (cusp tip loading) 10,000 (inclined cuspal plane loading)	40 mm diameter stainless steel and 6 mm diameter stainless steel

Table 3. Mean fatigue failure loads (FFL) in Newtons, number of cycles for failure (CFF) with their respective 95% confidence interval (CI) of each evaluated outcome and region of fracture.

Groups	**First crack**		**Crown fracture**			
	FFL Mean 95% CI	**CFF Mean 95% CI**	**FFL Mean 95% CI**	**CFF Mean 95% CI**	**Region of fracture**	
					Wall n (%)	**Groove n (%)**
Cusp tip	588^{AB} 514-661	$97{,}500^{AB}$ 82,800-112,200	1644^{A} 1468-1820	$293{,}312^{A}$ 260,005-326,619	16 (100)	0 (0)
Cuspal inclined plane	656^{A} 583-730	$111{,}250^{A}$ 96,509-125,990	1631^{A} 1560-1702	$295{,}174^{A}$ 282,603-307,744	8 (50)	8 (50)
Mixed	550^{B} 503-597	$85{,}000^{B}$ 74,414-95,585	1413^{B} 1325-1500	$253{,}029^{B}$ 235,991-270,067	15 (94)	1 (6)

Different letters indicate statistical differences on each column for each considered outcome.
* Kaplan-Meier and Mantel-Cox (log-rank) tests for FFL and CFF.

FIGURES

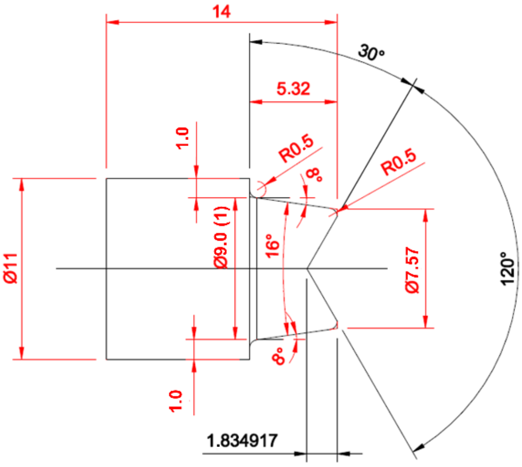


Figure 1 - Representation of the dimensions (in mm) and characteristics of the glass-fiber reinforced epoxy resin preparations.

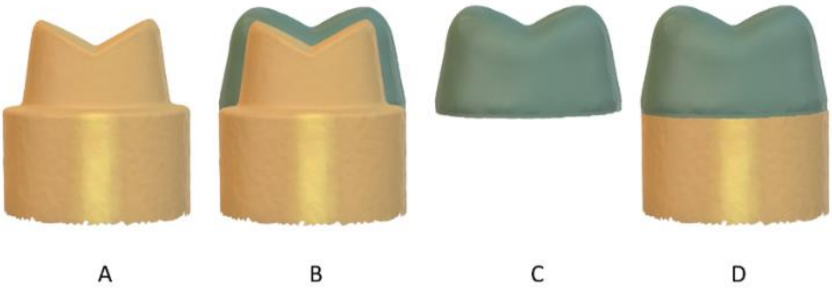


Figure 2 – Illustrations of glass-fiber reinforced epoxy resin preparations and ceramic crown planning. **A -** Scanning of glass-fiber reinforced epoxy resin preparations. **B** – Digital design of ceramic crowns with a thickness of 1 mm onto the preparation. **C –** Machined ceramic crowns. **D -** Verification of fit between glass fiber-reinforced epoxy resin preparations/ceramic crowns.

Cusp tip **Cuspal inclined plane** **Mixed**

Figure 3. Representations of the region of load application of each experimental group. Load application on the cusp tips was performed with a 40 mm diameter stainless steel load applicator. The load application in the cuspal inclined plane was obtained with a 6 mm diameter stainless steel load applicator, while the mixed group used both load regions and pistons.

First crack **Crown frature**

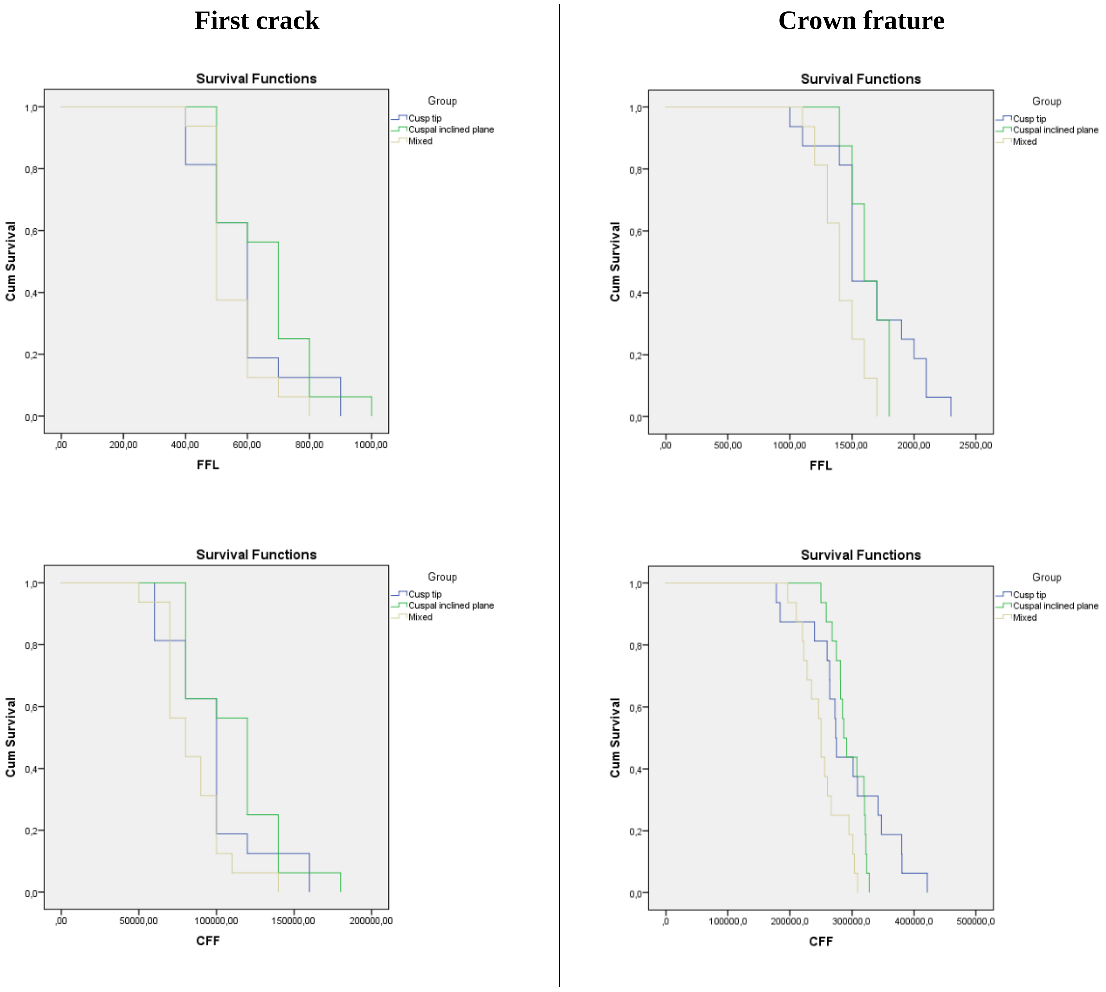


Figure 4. Survival curves according to the fatigue failure loads (FFL) and number of cycles for failure (CFF) steps in which each luted crown failed (according to first crack and crown fracture), obtained by the Kaplan–Meier and Log-rank tests.

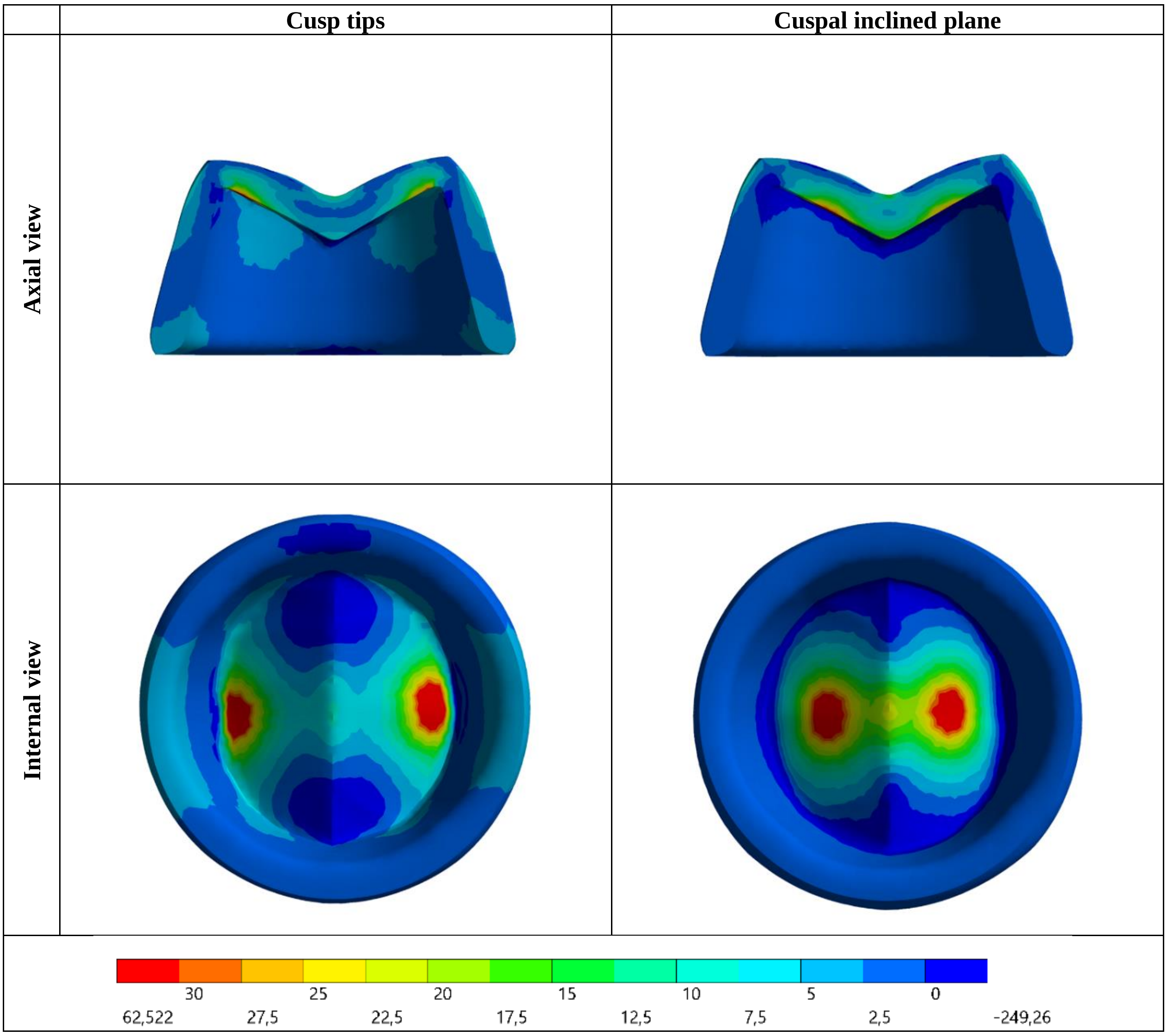


Figure 5. Finite elements analysis (FEA) demonstrates the tensile stress in the inner surface of the ceramic crowns (Maximum Principal Stress – MPS). The higher tensile stress concentration areas are located just below the load application region. In addition, loading on the inclined cuspal plane induced a higher tensile stress concentration in the central groove region compared to cusp tip loading.

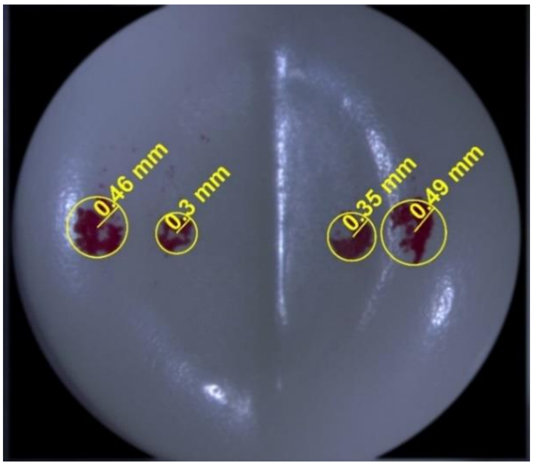
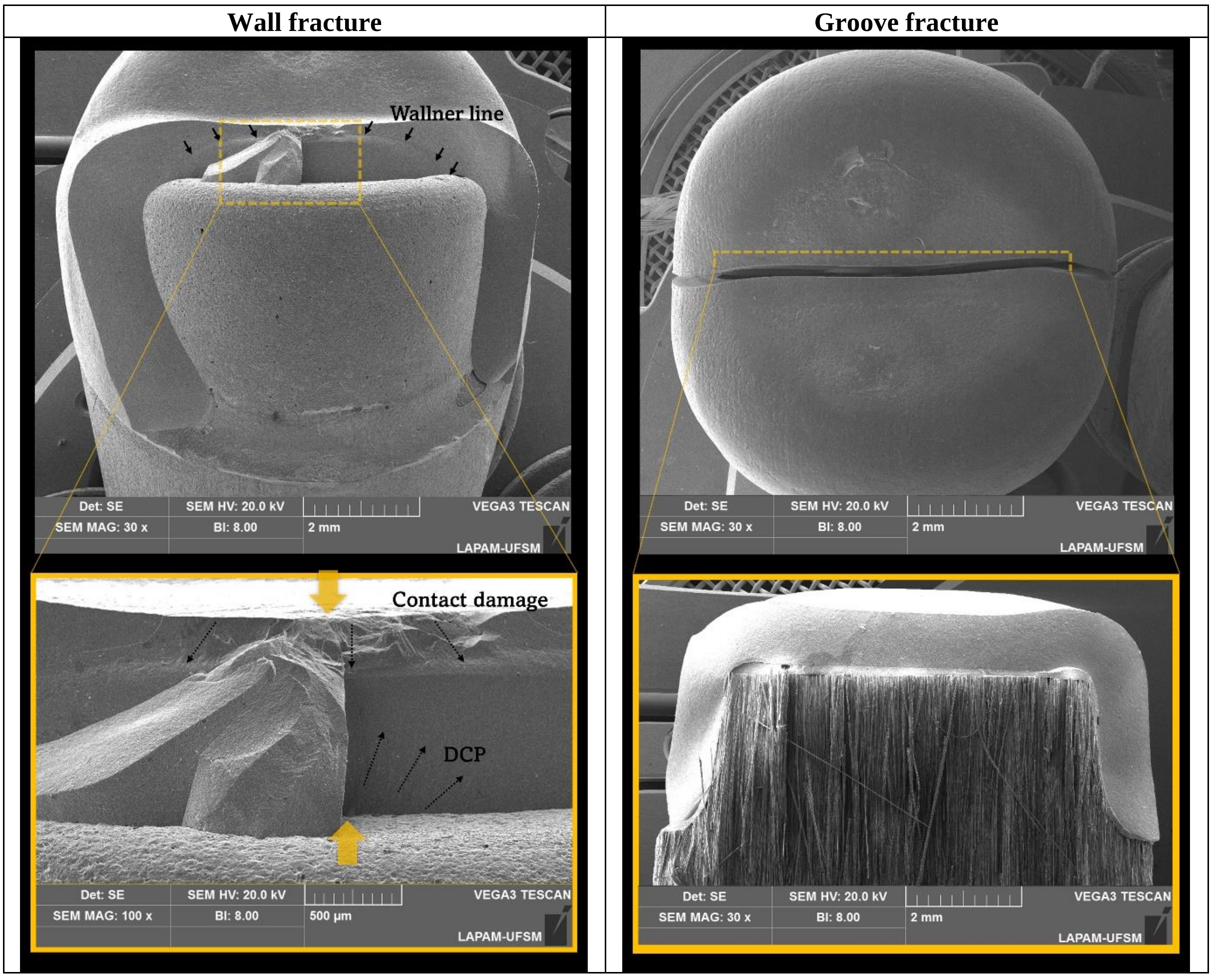


Figure 6. Representation of contact points between the ceramic crown and the load applicator with the mean values of contact point radii of a tested sample.



Wall fracture

Groove fracture

Figure 7. SEM micrographs (30× and 100× magnification) illustrating the fractographic pattern accessed at one representative specimen of each crown fracture region (Wall and Groove fracture). The yellow arrows indicate the origin of failure, while the dashed black arrows indicate the direction of the crack propagation (DCP); and the filled arrows indicate the Wallner lines. **Wall fracture:** The failure starting from the bonding surface as a radial crack and with increasing loading the failure also starts to propagate from the occlusal surface (hertzian-cone crack) towards the bonding surface until reaching the radial crack - moment of fracture. **Groove fracture:** Involved the displacement of ceramic and supporting substrate in the region of groove. It is not possible to identify an apparent failure origin in the ceramic, the failure possibly occurred from the shear rupture of the supporting substrate.

4 ARTIGO 3: FATIGUE BEHAVIOR, FAILURE MODE AND STRESS DISTRIBUTION OF OCCLUSAL VENEERS: INFLUENCE OF THE PROSTHETIC PREPARATION CUSP INCLINATIONS AND THE TYPE OF RESTORATIVE MATERIAL.

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Fatigue behavior, failure mode and stress distribution of occlusal veneers: influence of the prosthetic preparation cusp inclinations and the type of restorative material

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Fatigue behavior, failure mode and stress distribution of occlusal veneers: influence of the prosthetic preparation cusp inclinations and the type of restorative material

Abstract

Objective: To evaluate the effects of cusp inclination of the prosthetic preparation's occlusal surface and type of restorative material on the fatigue behavior, failure mode and stress distribution of occlusal veneers. *Materials and methods*: Glass-fiber reinforced epoxy resin prosthetic preparations for occlusal veneers with three different occlusal surface cusp inclination degrees (0°, 15 and 30°) were produced and assigned into six testing groups (n= 11) according to the cusp inclination (0°, 15 or 30°) and type of restorative material (lithium disilicate – LD or resin composite - RC). Despite different substrate preparation cusp inclination degrees, the restorations were designed maintaining 30° inclination between the cusps at the occlusal surface and a thickness of 0.7mm at the central groove region of the restorations to be machined in a CAD/CAM system. After cementation, the specimens were stored for about 7 days (under water at 37°C), and subsequently submitted to a load to failure test (n= 2) and an intermittent cyclic fatigue test (n= 9) (initial load: 100 N; step-size: 50 N; cycles/step: 10,000; loading frequency: 20 Hz; loading piston: 6 mm diameter stainless steel) until observing cracks. The data were analyzed by two-way ANOVA, Kaplan-Meier, and Mantel-Cox post-hoc tests. Finite element analysis (FEA) and fractographic analyzes were performed. *Results:* The fatigue performance of LD and RC occlusal veneers was evaluated based on different prosthetic preparation cusp inclinations. The 0° inclination showed the best fatigue performance for both materials (LD: 944 N, RC: 861 N), while the 15° and 30° inclinations had lower values (LD: 800 N and 533 N, RC: 739 N and 717 N, respectively). The study also found that for a 0° inclination, LD occlusal veneers performed better than RC ones (LD: 944 N > RC: 861 N), while for a 30° inclination, RC occlusal veneers had better fatigue performance than LD ones (LD: 533 N < RC: 717 N). No significant difference was observed between the materials for a 15° inclination (LD: 800 N = RC: 739 N). The FEA results showed a higher tensile stress concentration on lithium disilicate than on resin composite occlusal veneers. All lithium disilicate occlusal veneers showed radial crack failures, while resin composite occlusal veneers showed Hertzian cone cracks and radial cracks combined. *Conclusion:* Considering mechanical perspective only, RC occlusal veneers should be indicated when prosthetic preparation cusps inclinations are 30°. When 0° prosthetic preparation cusps inclinations are observed, LD occlusal veneers will behave mechanically better. When a 15° cusp inclination is preserved, both restorative materials behave similarly.

Keywords: Occlusal veneer. Preparation design. Ceramic. Resin composite. Fatigue. Failure

1. Introduction

Loss of dental hard tissue caused by wear and erosion has increased significantly in recent years [1]. This is a multifactorial and progressive process that is related to eating and parafunctional habits, occupation, oral hygiene, systemic problems and occlusal pattern [2, 3]. The restoration of the lost occlusal vertical dimension is important to prevent progression of the deleterious effects caused by the pathological wear of the tooth structure [4]. Treatment of tooth wear varies according to the severity of dental tissue loss. Conservative treatment may include removing damaged tooth tissue, restoring the tooth surface, and modifying the occlusion [5].

Occlusal veneers are currently indicated as less invasive restorative techniques compared to conventional treatments with metal-ceramic or all-ceramic crowns [6, 7]. Its use has been driven by advances and developments in reliable adhesive bonding techniques [8] since they are extracoronal restorations with a non-retentive design and their retention will exclusively depend on adhesion mechanisms.

Occlusal veneers are primarily indicated to restore the function and morphology of an occlusal surface affected by tooth wear [9]. The tooth wear mainly affects the occlusal surface and functional cusps [10]. In order to preserve tooth structure, occlusal veneer restorations will require a simpler and more intuitive preparation guided by interocclusal clearance and anatomical considerations [7]. Thus, the design and thickness of the restoration will change depending on the severity of tooth wear.

Another important topic is the choice of restorative material, computer-aided design and computer-aided manufacturing (CAD/CAM) materials such as ceramics and resin composite are widely used materials for occlusal veneer restorations [11]. Both materials presented a high patient satisfaction rate and provided favorable performance (survival rate: 100% for ceramic and 84.7% for resin composite) after 3 years of follow-up [12]; however, these materials have different mechanical properties, since they have different microstructures [13, 14] which tend to influence their load-bearing capacity in the posterior region [15].

Resin composite for CAD/CAM have a low abrasiveness for the antagonists [16] and a low elastic modulus, enabling better absorption of functional stresses by deformation [17]. In contrast, this material presents limitations such as wear, discoloration, and low fracture strength [18, 19]. Moreover, reinforced glass ceramics, such as lithium disilicate, present good adhesion capacity, increasing the material use of this material for adhesive bonded restorations as occlusal veneers [20]. When comparing with resin composites, lithium disilicate ceramic has less potential for material wear and color instability [21]; however, lithium disilicate still has the limitations of ceramics such as brittleness which can lead to catastrophic failures, and which are not always subject to intraoral repair as resin composite restorations [12].

In addition, the experiments applying intermittent cyclical loading for mechanical fatigue induction of samples designed with anatomical geometry as close as possible to dental crowns reach more relevant test conditions and methodological approach when intending to clinically attain more realistic biomechanical outcomes [22–29]. Given that the different degrees of tooth wear severity can be observed clinically, and that this factor will consequently determine the final design of the restoration necessary to reestablish the loss of tooth structure, the substrate preparation design [30] and type of restorative material [11] might influence the mechanical performance of occlusal veneers. Thus, this study aims to evaluate if the cusp inclination of the substrate occlusal surface and type of restorative material influence the fatigue behavior, failure mode and stress distribution of occlusal veneers. The hypothesis tested was that the cusp inclination of the substrate occlusal surface and type of restorative material would influence the fatigue behavior, failure mode and stress distribution of occlusal veneers.

2. Materials and methods

2.1 Experimental design

The materials used in this study, their chemical composition and manufacturers are described in Table 1. The study sample size was determined by convenience. As the specimen geometry used involves high acquisition costs, based on previous studies [7, 31, 32] that normally use a sample size of 10 per group to explore such themes, we defined a sample size of 11 specimens per group. The study design is presented in Table 2, where it can be observed that this study was composed of six experimental groups (n= 11), divided according to the cusp inclination degree and restorative material factors.

2.2 Specimen preparation

Three-dimensional models of the glass-fiber reinforced epoxy resin preparations were created using a software program (Rhinoceros version 5.0 SR8, McNeel North America, Seattle, USA) to simulate a simplified posterior tooth preparation for occlusal veneer (N= 66) with 0°, 15° or 30° convergence between the cusps (Figure 1). Next, the glass-fiber reinforced epoxy resin preparations were machined in a precision lathe (Diplomat 3001, Nardini, Americana, Brazil) from the glass-fiber reinforced epoxy resin bars (Perfil Pultrudado F, Protec).

The occlusal veneers were subsequently designed using a software program (Rhinoceros version 5.0 SR8, McNeel North America) from the three-dimensional models of the glass-fiber reinforced epoxy resin preparations, considering 30° convergence between the cusps in the occlusal surface and a thickness of 0.7mm in the central groove region (Figure 1). Then, the three-dimensional models of the occlusal veneers were exported to a CAD software program (CEREC in-Lab 3D, v4.1, Dentsply, Sirona Dental Systems GmbH) and machined (CEREC inLab MC XL, Dentsply Sirona Dental Systems GmbH) using diamond rotary instruments (step bur 12S a cylinder-pointed bur 12S) under water-cooling from lithium disilicate glass ceramic CAD/CAM blocks (IPS e.max CAD, Ivoclar) or resin composite CAD/CAM blocks (Tetric CAD, Ivoclar). After machining, the lithium disilicate occlusal veneers were crystallized according to the manufacturer’s instructions (840°C, 7 min vacuum, Vacumat 6000 MP, VITA Zahnfabrik, Bad Säckingen, Germany).

2.3 Bonding procedures

Before the surface treatment, the occlusal veneers and glass-fiber reinforced epoxy resin preparations were cleaned in a sonic bath (1440 D, Odontobras Medical and Dental Equipment, Ltda, Ribeirão Preto, SP, Brazil) for 5 min using 78% isopropyl alcohol [33] and distillated water, respectively.

The bonding surfaces of lithium disilicate occlusal veneers were etched with 5% hydrofluoric acid (Condac Porcelana, FGM) for 20 s, rinsed with air-water spray for 30 s, cleaned in a sonic bath with distilled water for 5 min and air-dried. Next, a layer of a silane-containing universal primer (Monobond N, Ivoclar) was applied on the etched ceramic surface for 15 s and kept untouched for 45 s.

The bonding surfaces of resin composite occlusal veneers were air abraded with aluminum oxide particles (50 µm, 10 mm distance, 1.5 bar) for 10 s [34], rinsed with air-water spray for 30 s, cleaned in an sonic bath with distilled water for 5 min and air-dried. Then, the abraded resin composite surface received a layer of primer (Multilink N Primer A and Multilink N Primer B - 1:1 ratio, Ivoclar), actively applied for 30 s and air-dried to obtain a thin and uniform layer.

The glass-fiber reinforced epoxy resin preparations were etched with 10% hydrofluoric acid (Condac Porcelana, FGM) for 60 s [35], rinsed with air-water spray for 30 s, cleaned in an sonic bath with distilled water for 5 min and air-dried. Multilink N Primer A and Multilink N Primer B were then mixed in a 1:1 ratio and applied for 30 s actively and air-dried to obtain a thin and uniform layer.

Each occlusal veneer was bonded to its respective glass-fiber reinforced epoxy resin preparation using a resin cement (Multilink N, Ivoclar). After the resin cement application onto the occlusal veneer bonding surface, each occlusal veneer was placed over the corresponding glass-fiber reinforced epoxy resin preparation under a constant load (2.5 N). Next, the excess of resin cement was removed, and the set was light-cured (1200 mW/cm², Radii-cal LED curing light, SDI, Bayswater, Australia) for five exposures of 20 s around the specimen (buccal, mesial, lingual, distal, and occlusal surfaces). The samples were then stored for about 7 days (under water at 37°C).

2.4 Load to failure test

In aiming to define the load fatigue test parameters and evaluate the mechanical degradation generated by cyclic fatigue, two specimens (n= 2) of each group were randomly selected for a load to failure monotonic test using a 6 mm diameter stainless steel loading piston (Figure 2) in a universal testing machine (EMIC DL1000, São José dos Pinhais, Brazil) at a crosshead speed of 1 mm/min.

2.5 Cyclic fatigue tests

Nine specimens (n= 9) per group were subjected to the cyclic fatigue test in an electrodynamic testing machine (Instron ElectroPuls E3000, InstronCorp, Norwood, USA). Thus, the following test parameters were adopted: initial load of 100 N, load increment of 50 N, 10,000 cycles per step, 6 mm diameter stainless steel loading piston [36], loading frequency of 20 Hz [37] and immersion in distilled water.

The fatigue test was paused at the end of the number of cycles defined for the step when the specimen was removed from the fatigue machine, air-dried and verified for the presence of the cracks by oblique transillumination [38]. Both the specimen and the fatigue device received marks with a permanent marker, to ensure that the loading always occurred at the same region when repositioning the sample to run the fatigue test. In the absence of cracks, the test was resumed with the load increased by a load increment. If cracks were detected, the fatigue failure load (FFL) and the number of cycles for failure (CFF) were recorded, and the test was ended.

2.6 Finite element analysis (FEA)

Three-dimensional models of occlusal veneers, resin cement, glass-fiber reinforced epoxy resin preparations and loading piston were created using a software program (Rhinoceros version 5.0 SR8, McNeel North America) and analyzed using the ANSYS CAE software program (ANSYS 19.3, ANSYS Inc., Houston, USA). A static structural analysis was applied according to the test assembly (Figure 2). All materials were considered isotropic, linear, and homogeneous. Young’s moduli (GPa) and Poisson’s ratios for lithium disilicate ceramic

(E= 95; Gpa; ν= 0.25), resin composite (E= 11; Gpa; ν= 0.28), glass-fiber reinforced epoxy resin (E= 14.9; GPa; ν= 0.31), resin cement (E = 7.5; GPa, ν= 0.3) and stainless steel (E= 190; GPa; ν= 0.27) were obtained from previous studies [35, 39, 40]. The models of 0° substrate preparation groups were composed of 106,980 tetrahedron solid elements with 168,677 nodes; the 15° substrate preparations groups were composed of 177,855 tetrahedron solid elements with 244,403 nodes; and 30° substrate preparations groups were composed of 223,686 tetrahedron solid elements with 329,530 nodes. The connections among base, restoration and resin cement were considered perfectly bonded and frictional (0.12) to the loading piston and restoration. The models were loaded (100 N) at the top of the loading piston and constrained at the bottom surface of the base. After the coherence and mesh convergence test, the maximum principal stress was used as failure criteria to compare the groups.

2.7 Fractographic analysis

After the fatigue test the specimens were analyzed in a stereomicroscope (Discovery V20, Carl Zeiss, Gottingen, Germany) to identify the failure types. Then, the restorative material fragments were detached to expose the fractured surface. Next, representative specimens of each restorative material were selected and submitted to Scanning Electron Microscopy (SEM, JSM-6360, JEOL, Tokyo, Japan) 35×, 50×, 100× and 120× magnifications to access and illustrate the failure origin and characterize the fractographic features.

2.8 Data analysis

A power estimation of the statistical analysis was performed in the G*Power software [41] using a post-hoc power analyzes based on α= 0.05, sample size= 66 and effect size= 1.59, where the effect size was calculated based on FFL means and the mean standard deviation of the experimental groups. The data were analyzed using a statistical software program (IBM SPSS Software, IBM, Armonk, United States) considering a significance level of 0.05. The data were diagnosed as normal by the Shapiro-Wilk test (FFL: p= 0.442; CFF: p= 0.370) and homoscedastic by the Levene’s test (FFL: p= 0.312; CFF: p= 0.369). Two-way ANOVA was performed (α= 0.05) to access the influence of the ‘substrate preparation’ and ‘restorative material’ factors, as well as to access the interaction between such factors. Additionally, FFL and CFF data were statistically analyzed through a survival analysis using Kaplan Meier and Mantel-Cox (log-rank) post-hoc tests (α= 0.05). The FEA, and fractographic data were qualitatively analyzed.

3. Results

A power estimation of the statistical analysis was determined as 100%. Two-way ANOVA revealed significant influence of the ‘substrate preparation’ (FFL: p= 0.000, F= 59.556; CFF: p= 0.000, F= 50.097) factor, and for the interaction between factors ‘substrate preparation*restorative material’ (FFL: p= 0.000, F= 16.889; CFF: p= 0.000, F= 14.969), but not statistically significant for ‘restorative material’ (FFL: p< 0.536, F= 0.389; CFF: p= 0.323, F= 0.997).

Considering load to failure data, all groups showed a decrease in load values from the monotonic test to the fatigue test. The greatest decreases in load values were observed in the resin composite occlusal veneer groups (Table 3).

The fatigue performance of lithium disilicate and resin composite occlusal veneers was evaluated based on different prosthetic preparation cusp inclinations. The 0° inclination showed the best fatigue performance for both materials, while the 15° and 30° inclinations had lower values, with the 15° inclination condition presenting intermediate behavior (Table 3, Figure 3). The study also found that for a 0° inclination, lithium disilicate occlusal veneers performed better than resin composite ones, while for a 30° inclination, resin composite occlusal veneers had better fatigue performance than lithium disilicate ones. No significant difference was observed between the materials for a 15° inclination.

FEA data showed a higher concentration of tensile stress on the lithium disilicate than on resin composite occlusal veneers. In addition, it is possible to observe that the areas of greatest concentration of tensile stresses are located on the intaglio surface of the lithium disilicate occlusal veneers, while they are concentrated on the occlusal surface in the resin composite ones (Figure 4).

The fractographic analysis showed different failure patterns according to the restorative material. The lithium disilicate occlusal veneers presented radial cracks – failures starting from defects present at the bonding surface that propagated to the opposite side (occlusal surface) (Figure 5). White resin composite occlusal veneers showed both radial and hertzian-cone cracks. The failure starts from the bonding surface as a radial crack, and then with increasing load the failure also starts to propagate from the occlusal surface (Hertzian-cone crack) towards the bonding surface until it reaches the radial crack, which is the moment of failure detection (Figure 5).

4. Discussion

Occlusal veneers have been indicated as more conservative treatments for teeth affected by wear and loss of occlusal vertical dimension. In this perspective, the substrate preparation's cusp inclination and restoration design will vary according to the severity degree of tooth wear. Our results showed different behavior of restorations depending on substrate preparation and restorative material used. Thus, the hypothesis was accepted.

The substrate preparations adopted in our study considered the different severity degrees of tooth wear observed clinically, which progresses from functional cusps [10]. In this perspective, all restorations presented thickness of 0.7 mm in the central groove, but different thickness in load application area depending on the type of substrate preparation (Figure 1). Therefore, different material thickness in loading area could justify the wide range on the fatigue behavior values according to substrate preparation (Table 3).

The main point of discussion is the interaction between the substrate preparation and the restorative material. Considering tooth remnants which maintain cusp inclinations of 30°, a 0.7 mm thickness occlusal veneer (Figure 1) was necessary to reconstruct the lost tooth structure. In this scenario, the resin composite occlusal veneers presented better fatigue behavior than lithium disilicate occlusal veneers (Table 3), constituting similar results to those found by Schlichting et al. [6]. This fact can be explained by the relatively similar elastic modulus of the resin composite (≅11 GPa) and supporting substrate (≅14.9 GPa) which enable better absorption of functional stresses by deformation [17]. A lower concentration of tensile stress was found on the resin composite occlusal veneers than on lithium disilicate occlusal veneers (Figure 4) which contributed to better fatigue performance.

As tooth wear increases, the inclination of the cusps decreases until complete removal of the cuspal anatomy (0°). With this, there is also an increase in the restoration thickness (Figure 1) needed to reconstruct the lost tooth structure. However, no statistically significant differences were found between restorative materials in relation to 15° substrate preparations (Table 3). This means that both restorative materials presented similar load-bearing capacity for the intermediate load values (Table 3) and demonstrate a balance between the greater resistance of the lithium disilicate and the better capacity to absorb the resin composite loads for intermediate thicknesses [13].

Nevertheless, in a scenario when the tooth remnant has already lost its cusp inclinations and a 0° of cusp inclination is observed (and so a greater thickness of restorative material is required), lithium disilicate occlusal veneers showed the best fatigue behavior (Table 3).

Lithium disilicate restorations with greater material thickness tend to have better fatigue performance than resin composite [13]. As the thickness of the restorative material increases, a greater load is required to fracture the restorative material. Thus, considering that the intrinsic strength of lithium disilicate is higher than the composite resin [42], it is logical to expect better behavior for the ceramic material which is reinforced by crystals.

Next, the failure mode was different according to occlusal veneer restorative material (Figure 5). On the one hand, lithium disilicate occlusal veneers presented radial crack failures, which are characterized as starting from defects on the intaglio surface and progressing towards the occlusal surface [35]. On the other hand, resin composite occlusal veneers presented competitive failure modes originating from the occlusal indentation area (Hertzian-cone crack) and radial cracks of the intaglio surface [43, 44]. This failure mode demonstrates that there was an interaction between contact damage on the occlusal surface and tensile stresses at the intaglio interface [45].

Unlike other studies reported in the literature, the present study used glass-fiber reinforced epoxy resin simplified preparations, which is considered a material with properties like those of dentin [35]. While this geometry allows for greater sample standardization and better isolation of study factors, the intrinsic variability of natural teeth is no longer considered. It should also be noted that the tested restorations had different thicknesses, because of the different degrees of prosthetic preparation cusps inclinations, despite this being a limitation, the focus of the study was to evaluate the interaction between the preparation cusps inclinations and type of restorative material. Also, the long-term evaluation is a must when the retention condition of the restauration is diminished dramatically as in this current study, mainly when having a flat occlusal surface. Thus, the data must be interpreted with caution. Furthermore, the loading pattern was only axial, sliding movements were not considered, nor was degradation by aging the bonding surfaces using different restorative materials. In this sense, further studies considering tooth substrate and aging are suggested.

5. Conclusion

When considering the mechanical perspective, resin composite occlusal veneers should be indicated when prosthetic preparation cusps inclinations are 30°. When 0° prosthetic preparation cusps inclinations are observed, lithium disilicate occlusal veneers will behave mechanically better. When a 15° cusp inclination is preserved, both restorative materials behave similarly.

Author contribution

Helder Callegaro Velho: conceptualization, data curation, formal analysis, methodology, writing—original draft. Kiara Serafini Dapieve: methodology, formal analysis, data curation, writing—review and editing. Elisa Donária Aboucauch Grassi: methodology, formal analysis, data curation, writing—review and editing. Alexandre Luis Souto Borges: data curation, methodology, writing—review and editing. Renata Marques de Melo Marinho: data curation, methodology, writing—review and editing. Gabriel Kalil Rocha Pereira: supervision, formal analysis, data curation, writing—review and editing, validation. Andressa Borin Venturini: conceptualization, supervision, formal analysis, data curation, writing—review and editing. Luiz Felipe Valandro: conceptualization, supervision, funding acquisition, formal analysis, data curation, writing—review and editing.

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Declarations

Informed consent: Written informed consent was taken from all participants included in the study.

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TABLES

Table 1. Description of materials, commercial name, manufacturer, composition, and batch number.

Material	Commercial name/manufacturer	Composition*	Batch number
Lithium disilicate glass ceramic	IPS e.Max CAD LT A2/C16, Ivoclar, Schaan, Liechtenstein	SiO_2 57–80 %wt, Li_2O 11–19 %wt, K_2O 0–13 %wt, P_2O_5 0–11 %wt, ZrO_2 0–8 %wt, ZnO 0–8 %wt, Al_2O_3 0–5 %wt, MgO 0–5 %wt, colouring oxides 0–8 %wt.	Z02NKF
Composite resin	Tetric CAD HT A2/C16, Ivoclar	Dimethacrylates monomers (Bis-GMA, Bis-EMA, TEGDMA and UDMA) 28%wt, barium aluminum silicate glass (particle size: < 1 µm) 64%wt, silicon dioxide (particle size: < 20 nm)7.1 %wt, additives and pigments 0.5%wt	Z03JD6
Aluminum Oxide	Polidental, Cotia, Brazil	Aluminum Oxide (particle size: 50 µm)	44493
Ceramic primer coupling agent	Monobond N, Ivoclar	Alcohol solution of silane methacrylate, phosphoric acid methacrylate and sulphide methacrylate.	Z02M0Y
5% hydrofluoric acid	Condac Porcelana, FGM, Joinville, Brazil	< 5% hydrofluoric acid.	051021
10% hydrofluoric acid	Condac Porcelana, FGM	< 10% hydrofluoric acid.	220322
Dual cure resin cement	Multilink N, Ivoclar	Base: Dimethacrylates and HEMA 33.1 %wt, Barium glass filler 37.4 %wt, ytterbium trifluoride, 23 %wt, highly dispersed silica 5.4 %wt, catalysts and stabilizer 1 %wt, pigments <0.03 %wt	Z01Y95
		Catalyst: Dimethacrylates and HEMA 32.4 %wt, Barium glass filler 37.4 %wt, ytterbium trifluoride, 23 %wt, highly dispersed silica 5.4 %wt, catalysts and stabilizer 1.8 %wt	
Primer	Multilink Primer (A and B), Ivoclar	Primer A: water 85.7 %wt, initiators 14.3 %wt.	Z01B1D
		Primer B: phosphonic acid acrylate 48.1 %wt, (hydroxyethyl methacrylate, methacrylate mod. polyacrylic acid) 51.9 %wt, stabilizer <0.02 %wt.	Z01YLS
Glass-fiber reinforced epoxy resin	Perfil Pultrudado F, Protec, São Paulo, Brazil	Continuous filament woven fiberglass bonded with epoxy resin.	-

*The chemical composition is described according to the manufacturers' information.
%wt – % in weight.

Table 2. Experimental design (n= 11).

Restorative material	Substrate preparation*	Group
Lithium disilicate	0°	LD_0
	15°	LD_15
	30°	LD_30
Resin composite	0°	RC_0
	15°	RC_15
	30°	RC_30

*Cusp inclination.

Table 3. Mean fatigue failure loads (FFL) in Newtons, number of cycles for failure (CFF) with their respective 95% confidence interval (CI) of each evaluated outcome and decrease percentage from monotonic to fatigue test.

Cusp preparation	Outcome	Restorative Material			
		Lithium disilicate		Resin composite	
		Mean 95% CI	Decrease from monotonic to fatigue test (%)	Mean 95% CI	Decrease from monotonic to fatigue test (%)
0°	**FFL**	944 ^{A} 897 – 992	53	861 ^{B} 819 – 904	77
	CFF	174,444 ^{A} 160,164 – 188,725		162,222 ^{AB} 153,718 – 170,726	
15°	**FFL**	800 ^{BC} 735 – 865	37	739 ^{CD} 683 – 795	81
	CFF	151,111 ^{BC} 138,696 – 163,526		137,778 ^{CD} 126,567 – 148,988	
30°	**FFL**	533 ^{E} 490 – 577	17	717 ^{D} 677 – 757	78
	CFF	94,444 ^{E} 86,369 – 102,519		133,333 ^{D} 125,332 – 141,335	

Different letters indicate statistical differences for each considered outcome.
* Kaplan-Meier and Mantel-Cox (log-rank) tests for FFL and CFF.

FIGURES

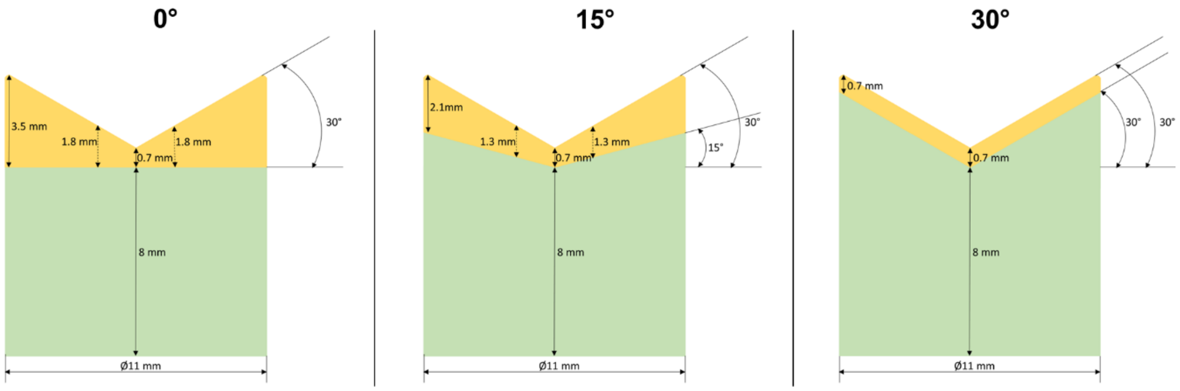

Figure 1. Representation of the dimensions and characteristics of the glass-fiber reinforced epoxy resin preparations and occlusal veneers. The glass-fiber reinforced epoxy resin preparations were designed considering different cusp inclination degrees (0°, 15° or 30°), while the occlusal veneers were designed maintaining 30° of cusp inclination on the occlusal surface and a thickness of 0.7 mm in the central groove region. The dashed arrows indicate the restoration thickness in the load application region.

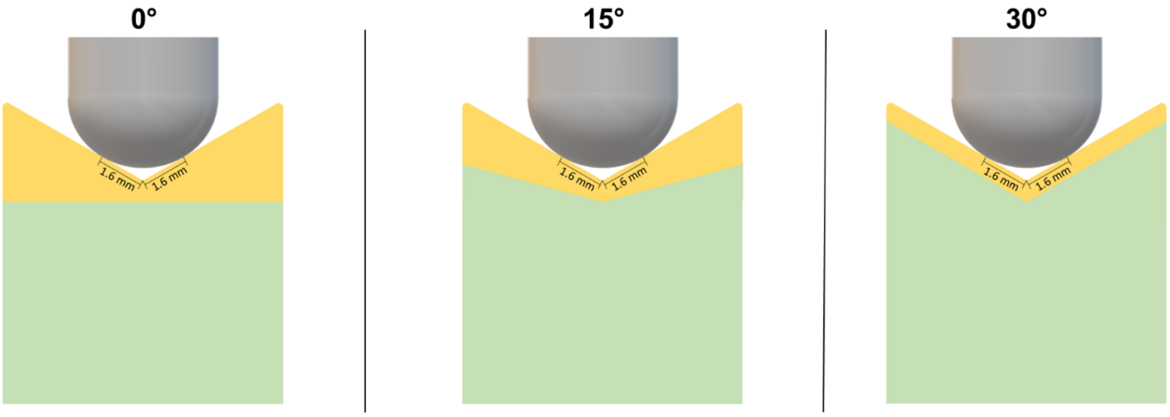

Figure 2. Representations of the load application region during the load to failure and fatigue tests. The load was applied on the inclined cuspal plane with a 6 mm diameter stainless steel loading piston.

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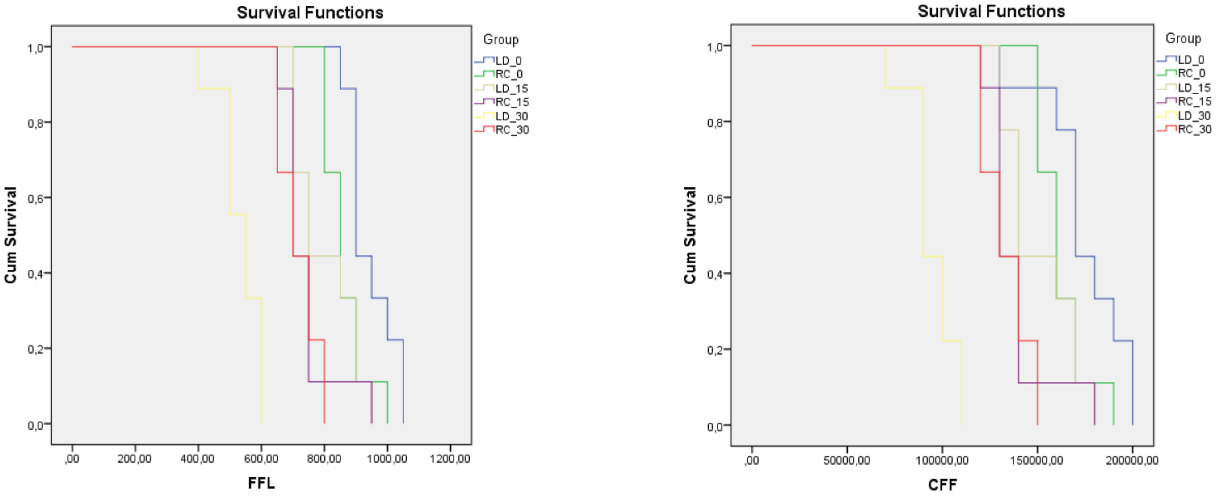

Figure 3. Survival curves according to the steps of FFL and CFF in which each specimen failed obtained by Kaplan–Meier and Log-rank tests.

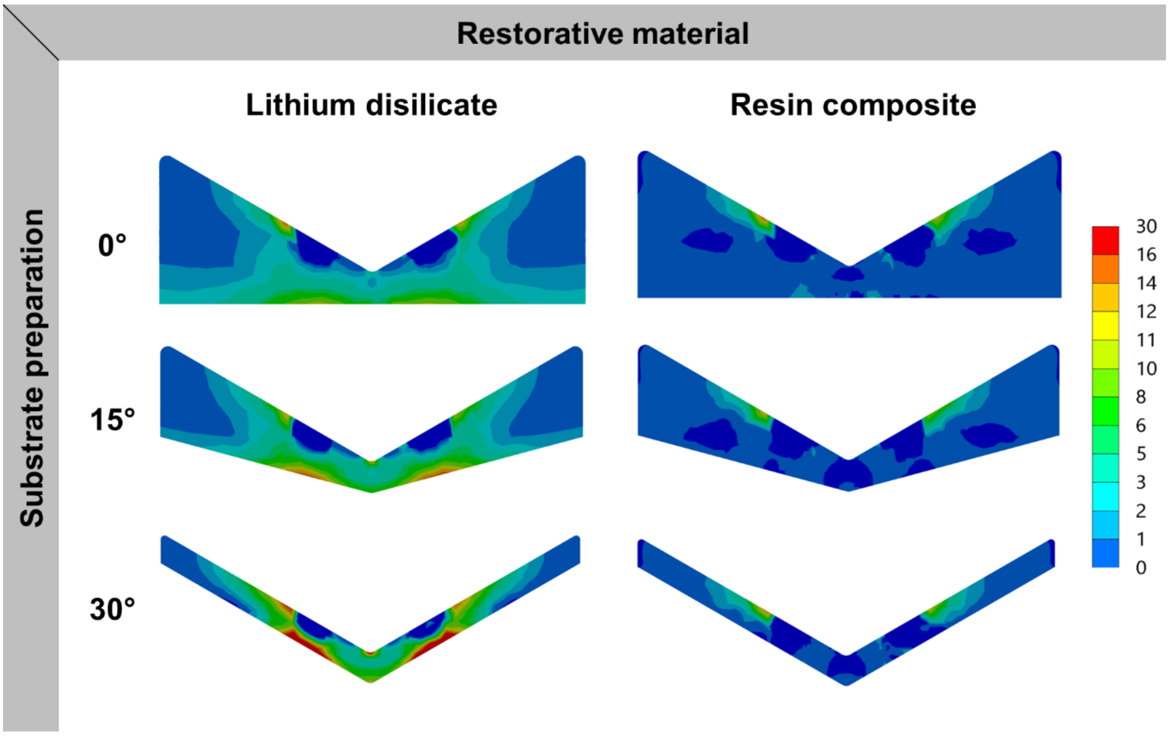

Figure 4. FEA demonstrates the tensile stress (Maximum Principal Stress – MPS) in an axial view of occlusal veneers. The higher tensile stress concentration areas are located just below the load application region. It is possible to see that the areas of greatest tensile stresses concentration are located on the intaglio surface in the lithium disilicate occlusal veneers, while they are concentrated on the occlusal surface in the resin composite occlusal veneers.

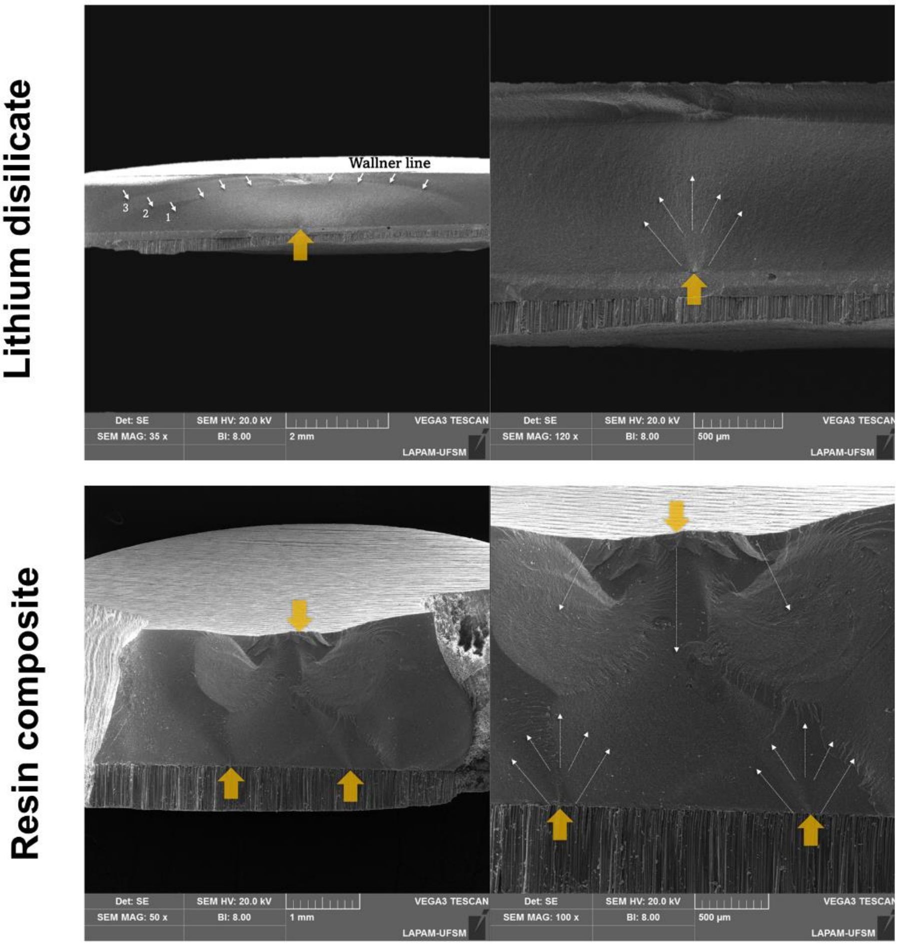

Figure 5. SEM micrographs (35×, 50×, 100× and 120× magnification) illustrating the fractographic pattern accessed at one representative specimen of each restorative material. The yellow arrows indicate the failure origin region, while the dashed white arrows indicate the crack propagation direction; and the filled arrows indicate the Wallner lines. **Lithium disilicate –** Fractographic pattern of a radial crack - failure starting from the defects present at the bonding surface that propagated to the opposite side (occlusal surface). White arrows 1, 2 and 3 indicate crack propagation at three different times until crack detection. **Resin composite –** The fractographic pattern shows radial and Hertzian-cone cracks. The failure starting from the bonding surface as a radial crack and the failure also starts to propagate from the occlusal surface (Hertzian-cone crack) towards the bonding surface with increasing loading until reaching the radial crack - moment of failure detection.

5 CONSIDERAÇÕES FINAIS

As diferentes variáveis exploradas nos estudos da presente tese tiveram influência no comportamento em fadiga e modo de falha de restaurações CAD/CAM.

O diâmetro do pistão teve influência no comportamento em fadiga, o modo de falha e a distribuição de tensões de restaurações simplificadas de cerâmica feldspática. No entanto, o material do pistão apresentou influência no comportamento à fadiga, no modo de falha e na distribuição de tensões de restaurações simplificadas de cerâmica feldspática apenas quando foram usados pistões de 6 mm de diâmetro. Pistões de 6 mm de diâmetro tendem a subestimar o desempenho à fadiga e criar falhas do tipo *Hertzian cone-crack*. Assim, seu uso deve ser evitado em testes de fadiga de restaurações simplificadas de cerâmica feldspática com espessura ≤ 1 mm.

As diferentes regiões de aplicação de carga tiveram influência no padrão de distribuição de tensão e, consequentemente, no desempenho à fadiga e na região de fratura de coroas monolíticas de dissilicato de lítio. Embora existam limitações em relação à simulação do padrão de carregamento clínico, a associação de regiões de carregamento *in vitro* é recomendada para promover melhor avaliação do comportamento à fadiga de um conjunto restaurado.

Em relação as restaurações do tipo laminados oclusais, considerando apenas uma perspectiva mecânica, os laminados oclusais em resina composta comportam-se melhor que os de dissilicato de lítio quando as inclinações das cúspides do preparo protético são de 30°, enquanto os laminados oclusais de dissilicato de lítio apresentam melhor performance do que os laminados de resina composta para inclinações de cúspides do preparo protético de 0°. Para uma inclinação de cúspides do preparo protético de 15°, ambos os materiais se comportam de forma semelhante.

Portanto, a partir dos pontos discutidos na presente tese, destaca-se importância de um adequado planejamento dos estudos laboratoriais, tendo em vista que as diferentes variáveis exploradas tiveram influência no comportamento em fadiga e modo de falha das restaurações. Além disso, comparações de diferentes variáveis (metodologias de teste, parâmetros de teste e relacionamento oclusal) seguem sendo sugeridas, numa perspectiva de tornar os estudos laboratoriais mais precisos e condizentes com os desafios pelos quais as restaurações estarão expostas quando em função na cavidade bucal.

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- Preferred fonts: Arial (or Helvetica), Times New Roman (or Times), Symbol, Courier.

- Number the illustrations according to their sequence in the text.

- Use a logical naming convention for your artwork files.

- Indicate per figure if it is a single, 1.5 or 2-column fitting image.

- For Word submissions only, you may still provide figures and their captions, and tables within a single file at the revision stage.

- Please note that individual figure files larger than 10 MB must be provided in separate source files.

A detailed guide on electronic artwork is available.

You are urged to visit this site; some excerpts from the detailed information are given here.

Formats

Regardless of the application used, when your electronic artwork is finalized, please 'save as' or convert the images to one of the following formats (note the resolution requirements for line drawings, halftones, and line/halftone combinations given below):

EPS (or PDF): Vector drawings. Embed the font or save the text as 'graphics'.

TIFF (or JPG): Color or grayscale photographs (halftones): always use a minimum of 300 dpi.

TIFF (or JPG): Bitmapped line drawings: use a minimum of 1000 dpi.

TIFF (or JPG): Combinations bitmapped line/half-tone (color or grayscale): a minimum of 500 dpi is required.

Please do not:

- Supply files that are optimized for screen use (e.g., GIF, BMP, PICT, WPG); the resolution is too low.

- Supply files that are too low in resolution.

- Submit graphics that are disproportionately large for the content.

Color artwork

Please make sure that artwork files are in an acceptable format (TIFF (or JPEG), EPS (or PDF) or MS Office files) and with the correct resolution. If, together with your accepted article, you submit usable color figures then Elsevier will ensure, at no additional charge, that these figures will appear in color online (e.g., ScienceDirect and other sites) in addition to color reproduction in print. Further information on the preparation of electronic artwork.

Figure captions

Ensure that each illustration has a caption. A caption should comprise a brief title (not on the figure itself) and a description of the illustration. Keep text in the illustrations themselves to a minimum but explain all symbols and abbreviations used.

Tables

Please submit tables as editable text and not as images. Tables can be placed either next to the relevant text in the article, or on separate page(s) at the end. Number tables consecutively in accordance with their appearance in the text and place any table notes below the table body. Be sparing in the use of tables and ensure that the data presented in them do not duplicate results described elsewhere in the article. Please avoid using vertical rules and shading in table cells.

References

Citation in text

Please ensure that every reference cited in the text is also present in the reference list (and vice versa). Any references cited in the abstract must be given in full. Unpublished results and personal communications are not recommended in the reference list, but may be mentioned in the text. If these references are included in the reference list they should follow the standard reference style of the journal and should include a substitution of the publication date with either 'Unpublished results' or 'Personal communication'. Citation of a reference as 'in press' implies that the item has been accepted for publication.

Reference links

Increased discoverability of research and high quality peer review are ensured by online links to the sources cited. In order to allow us to create links to abstracting and indexing services, such as Scopus, CrossRef and PubMed, please ensure that data provided in the references are correct. Please note that incorrect surnames, journal/book titles, publication year and pagination may prevent link creation. When copying references, please be careful as they may already contain errors. Use of the DOI is highly encouraged.

A DOI is guaranteed never to change, so you can use it as a permanent link to any electronic article. An example of a citation using DOI for an article not yet in an issue is: VanDecar J.C., Russo R.M., James D.E., Ambeh W.B., Franke M. (2003). Aseismic continuation of the Lesser Antilles slab beneath northeastern Venezuela. Journal of Geophysical Research, https://doi.org/10.1029/2001JB000884. Please note the format of such citations should be in the same style as all other references in the paper.

Web references

As a minimum, the full URL should be given and the date when the reference was last accessed. 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.

Data references

This journal encourages you to cite underlying or relevant datasets in your manuscript by citing them in your text and including a data reference in your Reference List. Data references should include the following elements: author name(s), dataset title, data repository, version (where available), year, and global persistent identifier. Add [dataset] immediately before the reference so we can properly identify it as a data reference. The [dataset] identifier will not appear in your published article.

References in a special issue

Please ensure that the words 'this issue' are added to any references in the list (and any citations in the text) to other articles in the same Special Issue.

Reference management software

Most Elsevier journals have their reference template available in many of the most popular reference management software products. These include all products that support Citation Style Language styles, such as Mendeley. Using citation plug-ins from these products, authors only need to select the appropriate journal template when preparing their article, after which citations and bibliographies will be automatically formatted in the journal's style. If no template is yet available for this journal, please follow the format of the sample references and citations as shown in this Guide. If you use reference management software, please ensure that you remove all field codes before submitting the electronic manuscript. More information on how to remove field codes from different reference management software.

Users of Mendeley Desktop can easily install the reference style for this journal by clicking the following link: http://open.mendeley.com/use-citation-style/journal-of-the-mechanical-behavior-of-biomedical-materials

When preparing your manuscript, you will then be able to select this style using the Mendeley plugins for Microsoft Word or LibreOffice.

Reference formatting

There are no strict requirements on reference formatting at submission. References can be in any style or format as long as the style is consistent. Where applicable, author(s) name(s), journal title/book title, chapter title/article title, year of publication, volume number/book chapter and the article number or pagination must be present. Use of DOI is highly encouraged. The reference style used by the journal will be applied to the accepted article by Elsevier at the proof stage. Note that missing data will be highlighted at proof stage for the author to correct. If you do wish to format the references yourself they should be arranged according to the following examples:

Reference style

Text: All citations in the text should refer to:

1. Single author: the author's name (without initials, unless there is ambiguity) and the year of publication;

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 can be listed either first alphabetically, then chronologically, or vice versa.

Examples: 'as demonstrated (Allan, 2000a, 2000b, 1999; Allan and Jones, 1999)…. Or, as demonstrated (Jones, 1999; Allan, 2000)… Kramer et al. (2010) 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., 2010. The art of writing a scientific article. J. Sci. Commun. 163, 51–59. https://doi.org/10.1016/j.Sc.2010.00372.

Reference to a journal publication with an article number: Van der Geer, J., Hanraads, J.A.J., Lupton, R.A., 2018. The art of writing a scientific article. Heliyon. 19, e00205. https://doi.org/10.1016/j.heliyon.2018.e00205.

Reference to a book:

Strunk Jr., W., White, E.B., 2000. The Elements of Style, fourth ed. Longman, New York.

Reference to a chapter in an edited book:

Mettam, G.R., Adams, L.B., 2009. 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.

Reference to a website:

Cancer Research UK, 1975. Cancer statistics reports for the UK. http://www.cancerresearchuk.org/ aboutcancer/statistics/cancerstatsreport/ (accessed 13 March 2003).

Reference to a dataset:

[dataset] Oguro, M., Imahiro, S., Saito, S., Nakashizuka, T., 2015. Mortality data for Japanese oak wilt disease and surrounding forest compositions. Mendeley Data, v1. https://doi.org/10.17632/ xwj98nb39r.1.

Journal abbreviations source

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

Video

Elsevier accepts video material and animation sequences to support and enhance your scientific research. Authors who have video or animation files that they wish to submit with their article are strongly encouraged to include links to these within the body of the article. This can be done in the same way as a figure or table by referring to the video or animation content and noting in the body text where it should be placed. All submitted files should be properly labeled so that they directly relate to the video file's content. In order to ensure that your video or animation material is directly usable, please provide the file in one of our recommended file formats with a preferred maximum size of 150 MB per file, 1 GB in total. Video and animation files supplied will be published online in the electronic version of your article in Elsevier Web products, including ScienceDirect. Please supply 'stills' with your files: you can choose any frame from the video or animation or make a separate image. These will be used instead of standard icons and will personalize the link to your video data. For more detailed instructions please visit our video instruction pages. Note: since video and animation cannot be embedded in the print version of the journal, please provide text for both the electronic and the print version for the portions of the article that refer to this content.

Data visualization

Include interactive data visualizations in your publication and let your readers interact and engage more closely with your research. Follow the instructions here to find out about available data visualization options and how to include them with your article.

Supplementary material

Supplementary material such as applications, images and sound clips, can be published with your article to enhance it. Submitted supplementary items are published exactly as they are received (Excel or PowerPoint files will appear as such online). Please submit your material together with the article and supply a concise, descriptive caption for each supplementary file. If you wish to make changes to supplementary material during any stage of the process, please make sure to provide an updated file. Do not annotate any corrections on a previous version. Please switch off the 'Track Changes' option in Microsoft Office files as these will appear in the published version.

Research data

This journal encourages and enables you to share data that supports your research publication where appropriate, and enables you to interlink the data with your published articles. Research data refers to the results of observations or experimentation that validate research findings. To facilitate reproducibility and data reuse, this journal also encourages you to share your software, code, models, algorithms, protocols, methods and other useful materials related to the project.

Below are a number of ways in which you can associate data with your article or make a statement about the availability of your data when submitting your manuscript. If you are sharing data in one of these ways, you are encouraged to cite the data in your manuscript and reference list. Please refer to the "References" section for more information about data citation. For more information on depositing, sharing and using research data and other relevant research materials, visit the research data page.

Data linking

If you have made your research data available in a data repository, you can link your article directly to the dataset. Elsevier collaborates with a number of repositories to link articles on ScienceDirect with relevant repositories, giving readers access to underlying data that gives them a better understanding of the research described.

There are different ways to link your datasets to your article. When available, you can directly link your dataset to your article by providing the relevant information in the submission system. For more information, visit the database linking page.

For supported data repositories a repository banner will automatically appear next to your published article on ScienceDirect.

In addition, you can link to relevant data or entities through identifiers within the text of your manuscript, using the following format: Database: xxxx (e.g., TAIR: AT1G01020; CCDC: 734053; PDB: 1XFN).

Mendeley Data

This journal supports Mendeley Data, enabling you to deposit any research data (including raw and processed data, video, code, software, algorithms, protocols, and methods) associated with your manuscript in a free-to-use, open access repository. During the submission process, after uploading your manuscript, you will have the opportunity to upload your relevant datasets directly to Mendeley Data. The datasets will be listed and directly accessible to readers next to your published article online.

For more information, visit the Mendeley Data for journals page.

Data in Brief

You have the option of converting any or all parts of your supplementary or additional raw data into one or multiple data articles, a new kind of article that houses and describes your data. Data articles ensure that your data is actively reviewed, curated, formatted, indexed, given a DOI and publicly available to all upon publication. You are encouraged to submit your article for Data in Brief as an additional item directly alongside the revised version of your manuscript. If your research article is accepted, your data article will automatically be transferred over to Data in Brief where it will be editorially reviewed and published in the open access data journal, Data in Brief. Please note an open access fee of 600 USD is payable for publication in Data in Brief. Full details can be found on the Data in Brief website. Please use this template to write your Data in Brief.

MethodsX

You have the option of converting relevant protocols and methods into one or multiple MethodsX articles, a new kind of article that describes the details of customized research methods. Many researchers spend a significant amount of time on developing methods to fit their specific needs or setting, but often without getting credit for this part of their work. MethodsX, an open access journal, now publishes this information in order to make it searchable, peer reviewed, citable and reproducible. Authors are encouraged to submit their MethodsX article as an additional item directly alongside the revised version of their manuscript. If your research article is accepted, your methods article will automatically be transferred over to MethodsX where it will be editorially reviewed. Please note an open access fee is payable for publication in MethodsX. Full details can be found on the MethodsX website. Please use this template to prepare your MethodsX article.

Data statement

To foster transparency, we encourage you to state the availability of your data in your submission. This may be a requirement of your funding body or institution. If your data is unavailable to access or unsuitable to post, you will have the opportunity to indicate why during the submission process, for example by stating that the research data is confidential. The statement will appear with your published article on ScienceDirect. For more information, visit the Data Statement page.

ANEXO B– NORMAS PARA PUBLICAÇÃO NO PERIÓDICO CLINICAL ORAL INVESTIGATIONS

Instructions for Authors

Types of papers

Papers may be submitted for the following sections:

- Research Article
- Reviews
- Brief Report – with up to 2000 words and up to two figures and/or tables
- Correspondence (Discussion paper)
- Debate (Letter to the Editor)
- Perspective (by Editor invitation only)
  Perspective articles are focused articles on topics of interest to a broad audience, but are written from a personal viewpoint. They are intended to provide a forum to be more speculative than Reviews, but should remain balanced and are intended to cover timely and relevant topics. These articles are peer reviewed.
  Limited to 1,500-3,000 words (excluding abstract, references and figure legends); Unstructured abstract 200 words; 4 tables/figures; 60 references
  It is the general policy of this journal not to accept case reports and pilot studies.

Editorial Procedure

Clinical Oral Investigations operates a single-blind peer-review system, where the reviewers are aware of the names and affiliations of the authors, but the reviewer reports provided to authors are anonymous.

Submitted manuscripts will generally be reviewed by two or more experts who will be asked to evaluate whether the manuscript is scientifically sound and coherent, whether it duplicates already published work, and whether or not the manuscript is sufficiently clear for publication. The Editors will reach a decision based on these reports and, where necessary, they will consult with members of the Editorial Board.

Summary of the editorial process

- The author submits a manuscript and the Editorial Office performs an initial quality check on the manuscript to ensure that the paper is formatted correctly
- The manuscript receives a tracking number and Manuscripts are assigned to an Editor-in-Chief or a Section Editor for an initial editorial assessment. If the decision is not to send the manuscript for review, the Editor contacts the author with the decision.
- If the Editor decides the paper is within the Journal's remit, peer reviewers are selected and assigned. This can take some time dependent on the responsiveness and availability of the reviewers selected.
- Reviewers are given 14 days from acceptance to submit their reports. Once the required reports are submitted, the Associate Editor will give a recommendation or the Editor-

in-Chief makes a final decision based on the comments received. The final decision is the sole responsibility of the Editors-in-Chief.

Manuscript Submission

Submission of a manuscript implies: that the work described has not been published before; that it is not under consideration for publication anywhere else; that its publication has been approved by all co-authors, if any, as well as by the responsible authorities – tacitly or explicitly – at the institute where the work has been carried out. The publisher will not be held legally responsible should there be any claims for compensation.

Permissions

Authors wishing to include figures, tables, or text passages that have already been published elsewhere are required to obtain permission from the copyright owner(s) for both the print and online format and to include evidence that such permission has been granted when submitting their papers. Any material received without such evidence will be assumed to originate from the authors.

Online Submission

Please follow the hyperlink “Submit manuscript” and upload all of your manuscript files following the instructions given on the screen.

Source Files

Please ensure you provide all relevant editable source files at every submission and revision. Failing to submit a complete set of editable source files will result in your article not being considered for review. For your manuscript text please always submit in common word processing formats such as .docx or LaTeX.

The Springer Author Academy is a set of comprehensive online training pages mainly geared towards first-time authors. At this point, more than 50 pages offer advice to authors on how to write and publish a journal article.

Title Page

The title page should include:

- The name(s) of the author(s)
- A concise and informative title
- The affiliation(s) and address(es) of the author(s)
- The e-mail address, telephone and fax numbers of the corresponding author

Abstract

Please provide a structured abstract of 150 to 250 words which should be divided into the following sections:

- Objectives (stating the main purposes and research question)
- Materials and Methods
- Results

- Conclusions
- Clinical Relevance
- These headings must appear in the abstract.

Keywords

Please provide 4 to 6 keywords which can be used for indexing purposes.

Text Formatting

Manuscripts should be submitted in Word.

- Use a normal, plain font (e.g., 10-point Times Roman) for text.
- Use italics for emphasis.
- Use the automatic page numbering function to number the pages.
- Do not use field functions.
- Use tab stops or other commands for indents, not the space bar.
- Use the table function, not spreadsheets, to make tables.
- Use the equation editor or MathType for equations.
- Save your file in docx format (Word 2007 or higher) or doc format (older Word versions).
- Manuscripts with mathematical content can also be submitted in LaTeX. We recommend using Springer Nature's LaTeX template.

Headings

Please use no more than three levels of displayed headings.

Abbreviations

Abbreviations should be defined at first mention and used consistently thereafter.

Footnotes

Footnotes can be used to give additional information, which may include the citation of a reference included in the reference list. They should not consist solely of a reference citation, and they should never include the bibliographic details of a reference. They should also not contain any figures or tables.

Footnotes to the text are numbered consecutively; those to tables should be indicated by superscript lower-case letters (or asterisks for significance values and other statistical data). Footnotes to the title or the authors of the article are not given reference symbols.

Always use footnotes instead of endnotes.

Acknowledgments

Acknowledgments of people, grants, funds, etc. should be placed in a separate section on the title page. The names of funding organizations should be written in full.

References

Citation

Reference citations in the text should be identified by numbers in square brackets. Some examples:

1. Negotiation research spans many disciplines [3].
2. This result was later contradicted by Becker and Seligman [5].
3. This effect has been widely studied [1-3, 7].

Reference list

The list of references should only include works that are cited in the text and that have been published or accepted for publication. Personal communications and unpublished works should only be mentioned in the text.

The entries in the list should be numbered consecutively.

If available, please always include DOIs as full DOI links in your reference list (e.g. “https://doi.org/abc”).

Journal article

Gamelin FX, Baquet G, Berthoin S, Thevenet D, Nourry C, Nottin S, Bosquet L (2009) Effect of high intensity intermittent training on heart rate variability in prepubescent children. Eur J Appl Physiol 105:731-738. https://doi.org/10.1007/s00421-008-0955-8.

Ideally, the names of all authors should be provided, but the usage of “et al” in long author lists will also be accepted:

Smith J, Jones M Jr, Houghton L et al (1999) Future of health insurance. N Engl J Med 965:325–329

Article by DOI

Slifka MK, Whitton JL (2000) Clinical implications of dysregulated cytokine production. J Mol Med. https://doi.org/10.1007/s001090000086

Book

South J, Blass B (2001) The future of modern genomics. Blackwell, London

Book chapter

Brown B, Aaron M (2001) The politics of nature. In: Smith J (ed) The rise of modern genomics, 3rd edn. Wiley, New York, pp 230-257

Online document

Cartwright J (2007) Big stars have weather too. IOP Publishing PhysicsWeb. http://physicsweb.org/articles/news/11/6/16/1. Accessed 26 June 2007

Dissertation

Trent JW (1975) Experimental acute renal failure. Dissertation, University of California

Always use the standard abbreviation of a journal's name according to the ISSN List of Title Word Abbreviations, see ISSN.org LTWA

If you are unsure, please use the full journal title.

Authors preparing their manuscript in LaTeX can use the bibliography style file sn-basic.bst which is included in the Springer Nature Article Template.

Tables

- All tables are to be numbered using Arabic numerals.
- Tables should always be cited in text in consecutive numerical order.
- For each table, please supply a table caption (title) explaining the components of the table.
- Identify any previously published material by giving the original source in the form of a reference at the end of the table caption.
- Footnotes to tables should be indicated by superscript lower-case letters (or asterisks for significance values and other statistical data) and included beneath the table body.

Artwork and Illustrations Guidelines

Electronic Figure Submission

- Supply all figures electronically.
- Indicate what graphics program was used to create the artwork.
- For vector graphics, the preferred format is EPS; for halftones, please use TIFF format. MSOffice files are also acceptable.
- Vector graphics containing fonts must have the fonts embedded in the files.
- Name your figure files with "Fig" and the figure number, e.g., Fig1.eps.

Line Art

- Definition: Black and white graphic with no shading.
- Do not use faint lines and/or lettering and check that all lines and lettering within the figures are legible at final size.
- All lines should be at least 0.1 mm (0.3 pt) wide.
- Scanned line drawings and line drawings in bitmap format should have a minimum resolution of 1200 dpi.
- Vector graphics containing fonts must have the fonts embedded in the files.

Halftone Art

- Definition: Photographs, drawings, or paintings with fine shading, etc.
- If any magnification is used in the photographs, indicate this by using scale bars within the figures themselves.
- Halftones should have a minimum resolution of 300 dpi.

Combination Art

- Definition: a combination of halftone and line art, e.g., halftones containing line drawing, extensive lettering, color diagrams, etc.
- Combination artwork should have a minimum resolution of 600 dpi.

Color Art

- Color art is free of charge for online publication.
- If black and white will be shown in the print version, make sure that the main information will still be visible. Many colors are not distinguishable from one another when converted to black and white. A simple way to check this is to make a xerographic copy to see if the necessary distinctions between the different colors are still apparent.
- If the figures will be printed in black and white, do not refer to color in the captions.
- Color illustrations should be submitted as RGB (8 bits per channel).

Figure Lettering

- To add lettering, it is best to use Helvetica or Arial (sans serif fonts).
- Keep lettering consistently sized throughout your final-sized artwork, usually about 2–3 mm (8–12 pt).
- Variance of type size within an illustration should be minimal, e.g., do not use 8-pt type on an axis and 20-pt type for the axis label.
- Avoid effects such as shading, outline letters, etc.
- Do not include titles or captions within your illustrations.

Figure Numbering

- All figures are to be numbered using Arabic numerals.
- Figures should always be cited in text in consecutive numerical order.
- Figure parts should be denoted by lowercase letters (a, b, c, etc.).
- If an appendix appears in your article and it contains one or more figures, continue the consecutive numbering of the main text. Do not number the appendix figures,"A1, A2, A3, etc." Figures in online appendices [Supplementary Information (SI)] should, however, be numbered separately.

Figure Captions

- Each figure should have a concise caption describing accurately what the figure depicts. Include the captions in the text file of the manuscript, not in the figure file.
- Figure captions begin with the term Fig. in bold type, followed by the figure number, also in bold type.
- No punctuation is to be included after the number, nor is any punctuation to be placed at the end of the caption.
- Identify all elements found in the figure in the figure caption; and use boxes, circles, etc., as coordinate points in graphs.
- Identify previously published material by giving the original source in the form of a reference citation at the end of the figure caption.

Figure Placement and Size

- Figures should be submitted within the body of the text. Only if the file size of the manuscript causes problems in uploading it, the large figures should be submitted separately from the text.
- When preparing your figures, size figures to fit in the column width.
- For large-sized journals the figures should be 84 mm (for double-column text areas), or 174 mm (for single-column text areas) wide and not higher than 234 mm.
- For small-sized journals, the figures should be 119 mm wide and not higher than 195 mm.

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If you include figures that have already been published elsewhere, you must obtain permission from the copyright owner(s) for both the print and online format. Please be aware that some publishers do not grant electronic rights for free and that Springer will not be able to refund any costs that may have occurred to receive these permissions. In such cases, material from other sources should be used.

Accessibility

In order to give people of all abilities and disabilities access to the content of your figures, please make sure that

- All figures have descriptive captions (blind users could then use a text-to-speech software or a text-to-Braille hardware)
- Patterns are used instead of or in addition to colors for conveying information (colorblind users would then be able to distinguish the visual elements)
- Any figure lettering has a contrast ratio of at least 4.5:1

Supplementary Information (SI)

Springer accepts electronic multimedia files (animations, movies, audio, etc.) and other supplementary files to be published online along with an article or a book chapter. This feature can add dimension to the author's article, as certain information cannot be printed or is more convenient in electronic form.

Before submitting research datasets as Supplementary Information, authors should read the journal’s Research data policy. We encourage research data to be archived in data repositories wherever possible.

Submission

- Supply all supplementary material in standard file formats.
- Please include in each file the following information: article title, journal name, author names; affiliation and e-mail address of the corresponding author.
- To accommodate user downloads, please keep in mind that larger-sized files may require very long download times and that some users may experience other problems during downloading.

  - High resolution (streamable quality) videos can be submitted up to a maximum of 25GB; low resolution videos should not be larger than 5GB.

Audio, Video, and Animations

- Aspect ratio: 16:9 or 4:3
- Maximum file size: 25 GB for high resolution files; 5 GB for low resolution files
- Minimum video duration: 1 sec
- Supported file formats: avi, wmv, mp4, mov, m2p, mp2, mpg, mpeg, flv, mxf, mts, m4v, 3gp

Text and Presentations

- Submit your material in PDF format; .doc or .ppt files are not suitable for long-term viability.
- A collection of figures may also be combined in a PDF file.

Spreadsheets

- Spreadsheets should be submitted as .csv or .xlsx files (MS Excel).

Specialized Formats

- Specialized format such as .pdb (chemical), .wrl (VRML), .nb (Mathematica notebook), and .tex can also be supplied.
- Collecting Multiple Files
- It is possible to collect multiple files in a .zip or .gz file.

Numbering

- If supplying any supplementary material, the text must make specific mention of the material as a citation, similar to that of figures and tables.
- Refer to the supplementary files as “Online Resource”, e.g., "... as shown in the animation (Online Resource 3)", “... additional data are given in Online Resource 4”.
- Name the files consecutively, e.g. “ESM_3.mpg”, “ESM_4.pdf”.

Captions

- For each supplementary material, please supply a concise caption describing the content of the file.

Processing of supplementary files

- Supplementary Information (SI) will be published as received from the author without any conversion, editing, or reformatting.

Accessibility

- In order to give people of all abilities and disabilities access to the content of your supplementary files, please make sure that
- The manuscript contains a descriptive caption for each supplementary material
- Video files do not contain anything that flashes more than three times per second (so that users prone to seizures caused by such effects are not put at risk)

Clinical Trial Registration

Clinical trials must be registered prior to submission of manuscripts. The registration site must be publicly available in English.

Recommended sites are: https://www.isrctn.com; https://www.clinicaltrialsregister.eu; https://clinicaltrials.gov or similar.

The registration number is required for the submission and must appear on the title page.

Ethical Responsibilities of Authors

This journal is committed to upholding the integrity of the scientific record. As a member of the Committee on Publication Ethics (COPE) the journal will follow the COPE guidelines on how to deal with potential acts of misconduct.

Authors should refrain from misrepresenting research results which could damage the trust in the journal, the professionalism of scientific authorship, and ultimately the entire scientific endeavour. Maintaining integrity of the research and its presentation is helped by following the rules of good scientific practice, which include*:

- The manuscript should not be submitted to more than one journal for simultaneous consideration.
- The submitted work should be original and should not have been published elsewhere in any form or language (partially or in full), unless the new work concerns an expansion of previous work. (Please provide transparency on the re-use of material to avoid the concerns about text-recycling ('self-plagiarism').
- A single study should not be split up into several parts to increase the quantity of submissions and submitted to various journals or to one journal over time (i.e. 'salami-slicing/publishing').
- Concurrent or secondary publication is sometimes justifiable, provided certain conditions are met. Examples include: translations or a manuscript that is intended for a different group of readers.
- Results should be presented clearly, honestly, and without fabrication, falsification or inappropriate data manipulation (including image based manipulation). Authors should adhere to discipline-specific rules for acquiring, selecting and processing data.
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