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Faculdade de Odontologia

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Tese de Doutorado

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Deise Cruz Oliveira

**Propriedades físicas e desenvolvimento de formação de biofilme em fios
ortodônticos estéticos**

Tese apresentada ao Programa de Pós-Graduação em Odontologia da Faculdade de Odontologia da Universidade Federal de Pelotas, como requisito parcial para obtenção do título de Doutora em Odontologia, área de concentração: Materiais Odontológicos.

Orientador: Prof. Dr. Rafael Guerra Lund

Co-Orientador: Prof. Dr. Douver Michelon

Pelotas, 2018

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Resumo

Oliveira, D. C. **Propriedades físicas e desenvolvimento de formação de biofilme em fios ortodônticos estéticos**. 2018. 6f. Projeto de Tese (Doutorado em Odontologia, Área de Concentração: Materiais Odontológicos) - Programa de Pós-Graduação em Odontologia, Faculdade de Odontologia, Universidade Federal de Pelotas, Pelotas, 2018.

A humanidade constantemente encontra-se em busca de novos padrões estéticos. Em Ortodontia, isso se reflete pelo uso de arcos ortodônticos estéticos. No entanto, tais materiais possuem limitada evidencia científica. Dessa forma, o objetivo deste estudo foi avaliar a formação de biofilme bacteriano, a superfície topográfica e a rugosidade antes e após ciclos de escovação em fios ortodônticos estéticos. Foram utilizadas 90 amostras de arcos de liga de Niquel-Titânio, de seção transversal 016 x .022 polegadas (n=10): Nitanium® Super Elastic (Ortho Organizers, Inc., EUA), Tooth Tone® Coated Archwire (Ortho Technology, BR), NT3™ SE (American Orthodontics, EUA), Niti Micro Dental White (Acme Monaco Co, EUA), NiTi Dental White S (Acme Monaco Co, EUA), FLI® Wire (RMO, Inc., EUA), Dany Coated Archwire (Dany BMT Co., Ltd., Coréia do Sul), Bio-active RC (GC Orthodontics, Japan) e Copper Ni-Ti® (Ormco Corporation, EUA). Grupos controle incluíram dois fios metálicos convencionais (Ormco, American Ortho). Fios estéticos incluíram: dois fios revestidos por resina epóxica (Ortho Technology, Ortho Organizer), dois por teflon (RMO, Monaco MD), dois fios revestidos por ródio (GC, Monaco DWS) e um fio revestido por prata+polímero (Dany). A avaliação de adesão do biofilme foi verificada por meio de técnica de coloração com cristal violeta e a viabilidade celular bacteriana por contagem de Unidades Formadoras de Colônia (UFC). A bactéria selecionada para os ensaios microbianos foi a *Streptococcus mutans* (ATCC® 25175™). Quanto a avaliação topográfica, os materiais avaliados foram preparados sobre stubs de alumínio, cobertos com uma fina camada de ouro-paládio (MED 010, Balzers Union, Liechtenstein) e observados em Microscopia Eletrônica de Varredura - MEV (JSM 5600LV, Jeol) operando a 15 kV em aumentos de 50 a 5.000X. A rugosidade superficial foi analisada através de um aparelho rugosímetro (Surf-Corder SE 1700, Kosaka Laboratory Ltd., Japan), com detector de 5µm e força de medição de 4mN, previamente calibrado e ajustado para uma leitura 0.05 mm, com 1.25 MM de comprimento e cut-off 0,025mm. As medidas foram realizadas antes e após simulação de ciclos de escovação. A simulação utilizou escovas dentais macias (Sorriso, Colgate) montadas junto à máquina de escovação (MEV 3Y-XT; Odeme, Luzerna, SC, Brazil). As amostras foram avaliadas após

1200, 2400 e 3600 ciclos, usando a proporção de 1:3 wt% água/pasta dental (Oral-B® Pro-Saúde, Procter & Gamble, São Paulo, SP, Brazil). Os dados de biofilme foram tabulados e submetidos a análise de variância e teste de Tukey; e os dados de rugosidade e escovação foram analisados por One-way ANOVA. Os resultados da análise microbiológica, indicaram que o grupo A8 (teflon) foi estatisticamente diferente do grupo A7 (controle). Houve diferença quanto à formação de biomassa, mas o ensaio de contagem bacteriana por colônia não apresentou diferença estatística significativa. Os resultados do MEV e rugosidade, antes e após escovação, visualmente demonstraram que o grupo A3 (teflon) apresentou maior irregularidade superficial. Quanto à rugosidade, houve um aumento na rugosidade no grupo A7 (controle) e A1 (Epóxica) de 40% e uma redução nos grupos A8, A3 (teflon), A5 (silver+polímero) e A9 (ródio) de 50% após escovação. Em conclusão, fios de Teflon demonstraram uma maior formação de biofilme, irregularidade superficial; no entanto, 50% de sua rugosidade foi reduzida após 3 meses de escovação.

Palavras-chave: Ortodontia. Fios ortodônticos estéticos. Materiais dentários ortodônticos. Propriedades de superfície. Ensaios mecânicos.

Abstract

OLIVEIRA, D. C. **Evaluation of mechanical and optical properties of aesthetic archwires**. 2018. 66f. Thesis Project (PhD Degree in Dentistry – Dental Materials) – Programa de Pós-Graduação em Odontologia, Faculdade de Odontologia, Universidade Federal de Pelotas, Pelotas, 2018.

Humanity is constantly in search of new aesthetic standards. In Orthodontics, this is reflected by the use of orthodontic archwires. Therefore, these materials have limited scientific evidence. Thus, the objective of this study was to evaluate the bacterial biofilm formation, topography surface and roughness before and after brushing cycles in aesthetic archwires. Nitinium® Super Elastic (Ortho Organizers, Inc., USA), Tooth Tone® Coated Archwire (Ortho Technology, BR), NiTi Dental White S (Acme Monaco Co, USA), FLI® Wire (RMO, Inc., USA), NT3™ SE (American Orthodontics, USA), Niti Micro Dental White (Acme Monaco Co, USA), Dany Coated Archwire (Dany BMT Co., Ltd., South Korea), Bio-active RC (GC Orthodontics, Japan) and Copper Ni-Ti (Ormco Corporation, USA). Conventional metal wires were control groups (Ormco, American Ortho). Aesthetic archwires were: two wires were coated by epoxy resin (Ortho Technology, Ortho Technology), two Teflon (RMO, Monaco MD), two Rhodium (GC, Monaco DWS) and one silver+polymer (Dany). The evaluation of biofilm formation was verified by means of a violet crystal staining technique and bacterial cell viability by counting of colony forming units (CFU). The bacterium selected for the bacterial count assay was *Streptococcus mutans* (ATCC® 25175™). Regarding the topographic evaluation, the wires were prepared on aluminum stubs, covered with a thin layer of gold-palladium (MED 010, Balzers Union, Liechtenstein) and observed in MEV (JSM 5600LV, Jeol) operating at 15 kV in increments of 50 to 5,000X. The surface roughness was analyzed using a Surf-Corder SE 1700 rugosimeter, Kosaka Laboratory Ltd., Japan, with a 5µm detector and 4mN measuring force, previously calibrated and adjusted for a reading of 0.05 mm, 1.25 mm length and a cut-off of 0.025 mm. The measurements were performed before and after the simulation of brushing cycles. The simulation used soft toothbrushes (Sorriso, Colgate) mounted into the brushing machine (MEV 3Y-XT; Odeme, Luzerna, SC, Brazil). The samples were evaluated after 1200, 2400 and 3600 cycles, using a ratio of 1: 3 wt% water / toothpaste (Oral-B® Pro-Health, Procter & Gamble, São Paulo, SP, Brazil). The biofilm data were tabulated and submitted to analysis of variance and Tukey's test; and the roughness and brushing data were analyzed by One-way ANOVA. According to the Article #1 results, microbiological and bacterial analysis, indicated that the group A8 (Teflon) was

statistically different from group A7 (control) regarding the biofilm formation. However, the colony count did not present statistically significant difference. Article #2, SEM and roughness before and after brushing, visually demonstrated that the groups A3 (Teflon) had the highest superficial irregularity. For roughness, there was an increase in roughness in the group A7 (control) and A1 (Epoxy) of 40% and a reduction in groups A8, A3 (Teflon), A5 (Silver + Polymer) and A9 (Rhodium) about 50% after brushing. In conclusion, Teflon wires demonstrated a greater adhesion of biofilm and superficial irregularity; however, 50% of its roughness was reduced after 3 months of brushing.

Keywords: Orthodontics. Aesthetic orthodontic archwires. Dental materials. Surface properties. Mechanical tests.

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Lista de abreviaturas e Siglas

BK	Bio-Kinetix® Thermally Activated Nitanium® (Ortho® Organizers, USA)
TT	Tooth Tone Coated Archwire (Ortho Technology, Brazil)
BA	Bio-active RC Tomy (GC Orthodontics, Japan)
NT3	NT3 SE Niti (American Orthodontics, USA)
AMC-MDW	AMC Niti Micro Dental White (Acme Monaco, USA)
AMC-DWS	AMC Niti Dental White S (Acme Monaco, USA)
Ormco-CN	Copper Niti Superelastic (Ormco, USA)
FLI	FLI® Nickel-Titanium (Rock Mountain Orthodontics, USA)
DC	Dany Coated Archwire (Dany BMT, South Korea)
ERC	Epoxy Resin-Coated
PTFE	Polytetrafluorethylene

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1. INTRODUÇÃO

1.1 Justificativa

Pouco se sabe sobre fios ortodônticos estéticos e suas propriedades mecânicas e biológicas, quando comparados com fios ortodônticos metálicos convencionais. Mesmo assim, a demanda desses materiais tem aumentado nas práticas odontológicas. Acredita-se que tal aumento não seja ainda mais significativo devido à falta de evidências científicas que suportem a sua utilização. Sendo assim, o propósito desta tese é prover evidência científica biológica através de ensaios comparativos de atividade bacteriana e formação de biofilme, os quais serão abordados no capítulo 1. O capítulo 2 analisará as propriedades mecânicas no que tange às características topográficas por MEV e rugosidade superficial antes e após ciclos de escovação.

1.2 Revisão da Literatura

1.2.1 Materiais Ortodônticos

A humanidade está constantemente buscando novas formas de aumentar os padrões estéticos de beleza, o que impacta a forma com a qual indivíduos são aceitos na sociedade. No entanto, padrões estéticos sofrem modificações com o passar dos anos. Na odontologia, isso se resume a demanda insaciável de pacientes que buscam o “sorriso perfeito”. A Odontologia cosmética refere-se a procedimentos odontológicos que buscam de uma melhora na aparência dos elementos dentários, gengivas e/ou oclusão do paciente (AACD, 2016). Essa prática odontológica se concentra, principalmente, na alteração da cor dos dentes, posição, forma, alinhamento e aparência geral do sorriso. Sendo assim, existem basicamente duas especialidades da Odontologia que predominantemente se dedicam a estética dental: Ortodontia e Odontologia Restauradora (ADA, 2016).

Em resposta a demanda estética, a Ortodontia aprimorou seus fios ortodôntico para fios estéticos recobertos por resina epóxica, polímeros ou teflon. Além disso, desenvolveu bráquetes estéticos feitos a partir de materiais cerâmicos ou compósitos para melhorar a apresentação estética de aparelhos ortodônticos (LEE, 2008; YU; LEE, 2011).

Em vista de satisfazer a constante demanda estética dos pacientes atuais, a Ortodontia como especialidade buscou o desenvolvimento de diferentes tipos de aparelhos ortodônticos, introduzindo transparentes removíveis conhecidos por Clear Aligners, como exemplo: Invisalign. Tais aparelhos removíveis satisfazem amplamente o desejo do consumidor por estética sendo que os mesmos se caracterizam por serem moldeiras transparentes dando ao paciente autonomia para remove-lo durante refeições, higiene oral e importantes eventos sociais. No entanto, essa autonomia ao consumidor resulta em mais de 60% do sucesso de tratamentos dependendo da colaboração e participação do paciente, o que na maioria das vezes leva a resultados comprometidos ou continuação do tratamento com aparelho ortodôntico fixo convencional (GRABER, 2016). Além disso, os chamados “Clear Aligner” apresentam limitações de tratamento devido a suas características estruturais e mecânicas. Por exemplo, Proffit e colaboradores (2016) reportam seis limitações de tratamentos com aparelhos removíveis as quais incluem: dentes impactados, dentes permanentes não erupcionados, coroas clinicas pequenas, excessiva overbite especialmente em presença de espaços, múltipla perda de dentes posteriores, e severa assimetria facial e discrepâncias no esqueleto.

Sendo assim, em busca de excelentes resultados de tratamento e estética simultaneamente, a Ortodontia vem desenvolvendo uma completa nova linha de aparelhos fixos a qual inclui bráquetes cerâmicos, fios recobertos estéticos, elásticos ortodônticos (translúcidos), implantes e outros (SAVAGE, 2015). Em resumo, diversos materiais estão sendo incorporados na composição dos dispositivos terapêuticos, tornando essencial a realização de novas pesquisas aplicadas a constante atualização do profissional desta área.

1.2.2 Fios ortodônticos estéticos

Considerando os avanços estéticos ocasionados pela introdução de bráquetes ortodônticos cerâmicos, o uso de arcos ortodônticos estéticos tem aumentado, mas não na mesma proporção. Fios ortodônticos feitos de ligas diferentes, como: aço inoxidável, cobalto-cromo-níquel, cobalto-cromo níquel-titânio são algumas das alternativas de fios ortodônticos disponíveis durante todas as fases do tratamento (O'BRIEN, 2008; ELIADES, 2012). Embora estes fios sejam fortes e duráveis, eles não são esteticamente agradáveis por causa de sua cor metálica, além disso, eles têm o potencial de causar reações alérgicas ou tóxicas devido ao contato com os tecidos moles ou duros da boca (ELIADES; ATHANASIOU, 2002; HENSTEN-PETTERSEN; JACOBSEN, 2003).

Apesar da contribuição ao ganho em estética desses arcos (IIJIMA et al., 2012), o seu uso não tem se traduzido na prática odontológica. A literatura reporta relativamente poucas evidências científicas sobre esses materiais estéticos devido a sua recente introdução, o que leva a insegurança de muitos ortodontistas ao utilizar tais materiais. Além disso, a possível fragmentação do revestimento estético e aumento da resistência ao atrito desses materiais, pode gerar incerteza quanto ao seu uso na prática clínica.

Atualmente, arcos estéticos ortodônticos podem ser divididos em duas categorias: [1] metálicos revestidos e [2] translúcidos (não-metálicos). Cada fio passa por um processo de fabricação próprio, resultando em propriedades específicas. Fios de liga revestidos de polímero foram primeiramente desenvolvidos como fios ortodônticos estéticos nos anos 2000 (ELAYYAN et al., 2008; ZEGAN et al., 2012; DA SILVA et al., 2013).

Por exemplo, materiais resinosos reforçados com fibra de vidro têm sido amplamente utilizados em Odontologia por décadas, porém sua aplicação como alternativa aos fios ortodônticos metálicos (CACCIAFESTA et al., 2008) tem sido recente. Atualmente, arcos metálicos podem ser revestidos com materiais metálicos como o ródio para um melhor efeito estético, ou com polímeros como o Poli-tetra-fluoroetileno [PTFE] (Teflon®) ou resina epóxica [dietileno glicol monobutil éter (1-5 %), dióxido de titânio (1-5 %), etilenoglicol 2-metil-hexil éter, acetato de metila,

cetona (0,1-1 %)]. O PTFE (politetrafluoretileno), popularmente conhecido pela marca comercial “Teflon®”, é um polímero sintético que consiste, basicamente, de carbono e flúor. Devido à força das ligações carbono-flúor, o PTFE tem o terceiro menor coeficiente de atrito de qualquer sólido conhecido, resistente ao calor, e hidrófobo (KRAVITZ, 2013).

Alguns fios metálicos de Cu-NiTi revestidos por PTFE (por exemplo, FLI®, Rocky Mountain Orthodontics) estão disponíveis no mercado. Já a resina epóxica é, basicamente, uma resina sintética composta pela combinação de epóxido e outros polímeros, entre as suas propriedades, podemos salientar sua aderência, resistência a solubilidade, isolamento elétrico, e estabilidade dimensional. Os fios revestidos por resina epóxica (por exemplo, Ultraesthetic™, G&H® Orthodontics), comumente, são revestidos em todas as superfícies do fio ou somente a face vestibular do fio para minimizar problemas de atrito, em geral até o segundo molar. A adesão entre o arco e revestimento segue sendo um problema, pois o revestimento pode se separar do arco metálico devido às forças mastigatórias e ativação de enzimas digestivas. Por exemplo, cerca de 25% do revestimento de epóxi é perdido durante o primeiro mês de tratamento *in vivo* (KRAVITZ, 2013), devido a essa baixa adesão.

Para arcos translúcidos (não-metálicos) podemos encontrar, também, fios de resina reforçados com fibras de vidro (FRP). Esses arcos (por exemplo, SimpliClear™, BioMers Products LLC) apresentam pouca deformação plástica/elástica, podendo ser considerados materiais friáveis quando comparados aos fios metálicos convencionais (KRAVITZ, 2013). Seu uso limita-se a pacientes Classe I, com leve a moderado apinhamento dentário, sendo contra-indicados para pacientes que necessitam de correções ântero-posteriores. No entanto, arcos de resina reforçados por fibra de vidro (arcos translúcidos) apresentam propriedades mecânicas, como resistência flexural, algo similares aos arcos metálicos de NiTi (BALLARD et al., 2012). Esses arcos apresentam alta translucidez se comparados aos arcos metálicos (KRAVITZ, 2013) e são indicados para o uso com bráquetes cerâmicos.

1.2.3 Avaliação de propriedades mecânicas da superfície de fios ortodônticos

Os valores de deflexão para fios recobertos quando comparados aos fios metálicos convencionais na literatura são controversos. Alguns autores afirmam que arcos revestidos apresentam menor carga de deflexão, o que poderia ser explicado por uma redução da espessura do fio metálico. Os arcos revestidos apresentam carga de deflexão menor se comparados aos arcos translúcidos de mesmo tamanho (KRAVITZ, 2013), enquanto arcos recobertos apresentam maiores forças de deflexão quando comparados a arcos sem recobrimento (IIJIMA et al., 2012).

Além disso, a fragmentação do revestimento pode levar a uma maior resistência causada pelo atrito, diminuindo o benefício estético (RYU et al., 2015). Arcos de NiTi recobertos por resina epóxica e Teflon apresentam rugosidade superficial menor que arcos de NiTi sem recobrimento (CHOI et al., 2015). Quando estes materiais são recobertos por prata ou ouro, os mesmos apresentam maior rugosidade superficial quando comparados a arcos recobertos somente por resina epóxica e Teflon, ou fios sem recobrimento. Arcos de NiTi recobertos por materiais contendo platina e prata apresentam uma maior rugosidade superficial quando comparado com outros fios ortodônticos recobertos (RYU et al., 2015).

Condições ambientais, tais como estocagem em água, afetam mais a superfície dos arcos estéticos de polímeros reforçados por fibra quando comparados aos arcos ortodônticos metálicos (CHANG et al., 2014). Arcos metálicos possuem valores de resistência flexural mais constantes que arcos estéticos de resina reforçados por fibra de vidro (CHANG et al., 2014). Apesar das desvantagens do uso de arcos ortodônticos recobertos atualmente disponíveis no mercado, esses são, provavelmente, o futuro de fios ortodônticos estéticos.

Ortodontistas devem considerar a incorporação do uso de arcos estéticos em seus procedimentos com objetivo de melhorar a experiência estética dos pacientes. Estudos demonstrando as diferentes propriedades destes materiais, in vitro e in vivo, devem ser conduzidos de forma a consolidar o seu uso na prática odontológica. Dessa forma, a oferta de novos substratos com potencial estético por parte da indústria e as experiências do profissional na prática clínica em ortodontia estimulam, diariamente, a realização de novos estudos para avaliação,

desenvolvimento e evolução dos materiais dentários. Esta demanda leva a um aprimoramento das propriedades dos fios ortodônticos estéticos com o objetivo de melhorar seu uso prático em ortodontia.

1.3 Objetivos

1.3.1 Objetivo Geral

O objetivo geral do presente estudo foi a avaliação das propriedades antimicrobiana, topográfica e mecânica de fios ortodônticos estéticos utilizados na prática odontológica.

1.3.2 Objetivos Específicos

A) Investigar a formação de biofilme bacteriano à superfície dos fios ortodônticos estéticos;

B) Avaliar a atividade antimicrobiana presente nos fios ortodônticos estéticos após exposição à *Streptococcus mutans*;

C) Analisar a superfície topográfica dos fios ortodônticos estéticos;

D) Avaliar a rugosidade superficial de fios ortodônticos antes e após ciclos de escovação.

2. Capítulo 1: Evaluation of superficial biofilm formation and bacterial activity in orthodontic aesthetic archwires

2.1 Introduction

Humanity is constantly searching for new ways to increase its levels of esthetic standards and this impacts individual's acceptance into society. However, standards evolve as years go by. In dentistry, this can be summarized by the increasing demand of patients for the “perfect smile” focuses on procedures to improve dental elements appearance, gum, and/or patients' occlusion (AACD, 2016).

In Orthodontics, this has impact the specialty dental materials such as: ceramic brackets, Orthodontic elastic (translucent, coloured) and aesthetic archwires, implants and others (PROFFIT et al., 2016). Several materials can be employed in the composition of the therapeutic devices, making it essential to carry out new applied researches and the constant updating of the professionals in this area.

Orthodontics treatments by itself increase the biofilm accumulation due the retention of plaque on brackets, archwires, elastomers, powerchains and elastics. Thus, the combination of biofilm accumulation and poor oral hygiene can lead to white spot lesions and further into dental caries (GRABER, 2016; PROFFIT, 2017). Furthermore, biomaterials for restoration of oral function are prone to biofilm formation, affecting oral health. Oral bacteria adhere to hydrophobic and hydrophilic surfaces but, due to floating shear, little biofilm accumulates on hydrophobic surfaces in vivo. Oral biofilms consist primarily of various bacterial strains, and depending on the type of restorative material to which the oral bacteria adhere and form the biofilm, biofilm may cause surface deterioration, which increases biofilm formation. The release of residual monomers from composite materials, for example, may affect biofilm growth in vitro, but in vivo effects are less pronounced, probably due to the large volume of saliva in which the compounds are released and its continuous refreshment (BUSSCHER et al., 2010; ZHANG et al., 2017).

In addition, Choi et al. (2015) analyzed the superficial roughness of aesthetic archwires and found that Epoxy and Teflon wire groups exhibited less surface

roughness and Silver+Polymer had the highest irregularity. D' Anto et al. (2012) also presented that Teflon was less roughness than other coated wires.

Ryu and colleagues (2015) showed that Silver+Polymer presented the highest roughness among coated and uncoated wires.

Also, Da Silva et al. (2013) analyzed the superficial roughness of aesthetic archwires and demonstrated that Epoxy wires showed a greater surface roughness when compared with other coated wires.

As a consequence, the scientific evidence about aesthetic orthodontic archwires, specially biological and mechanical properties, has been pretty limited in the literature (GRABER et al., 2016). Thus, the purpose of this study is to evaluate the bacterial activity and antimicrobial tests of such materials.

2.2 Objectives

A) Investigate superficial biofilm formation characterized by *Streptococcus mutans* on aesthetic archwires.

B) Evaluate superficial antimicrobial activity present on aesthetics archwires exposed to *Streptococcus mutans*.

2.3 Hypotheses

Orthodontic aesthetic archwires will present higher superficial biofilm formation when compared to conventional metallic orthodontic archwires.

2.4 Methodology

In all analyses, conventional metallic archwire will be used as control (A2, A7). The donated orthodontics aesthetics archwires used on the experiments are described on Table 1.

Table 1 - Orthodontic Aesthetic Archwires used on experiments

GROUP CODE	PRODUCT	MANUFACTURER	SPECIFICATION	COATING TYPE	LOT#	QTY
A1	Tooth Tone® Coated Archwire	Ortho Technology	16x22 NiTi	Epoxi	PO19425	5
A2	NT3™ SE	American Orthodontics	16x22 NiTi	No Coating (Control)	G74366	50
	NT3™ SE	American Orthodontics	16x22 NiTi	No Coating (Control)	149464	20
	NT3™ SE	American Orthodontics	16x22 NiTi	No Coating (Control)	152960	20
A3	FLI® Wire	RMO, Inc.	16x22 NiTi	PTFE	WO-786152	5
	FLI® Wire	RMO, Inc.	16x22 NiTi	PTFE	WO-727181	10
A4	Nitanium® Super Elastic	Ortho Organizers, Inc.	16x22 NiTi	Epoxi	183877	5
A5	Dany Coated Archwire	Dany BMT Co., Ltd.	16x22 NiTi	Silver + Polymer	362412	4
A6	Bio-active RC	Gc Orthodontics Europe GmbH	16x22 NiTi RC	Rhodium	A346	5
A7	Copper Ni-Ti®	Ormco Corporation	16x22 NiTi Cu	No Coating (Control)	51768825	20
A8	Niti Micro Dental White	Acme Monaco Co	16x22 NiTi	PTFE		
A9	NiTi Dental White S	Acme Monaco Co	16x22 NiTi	Rhodium		

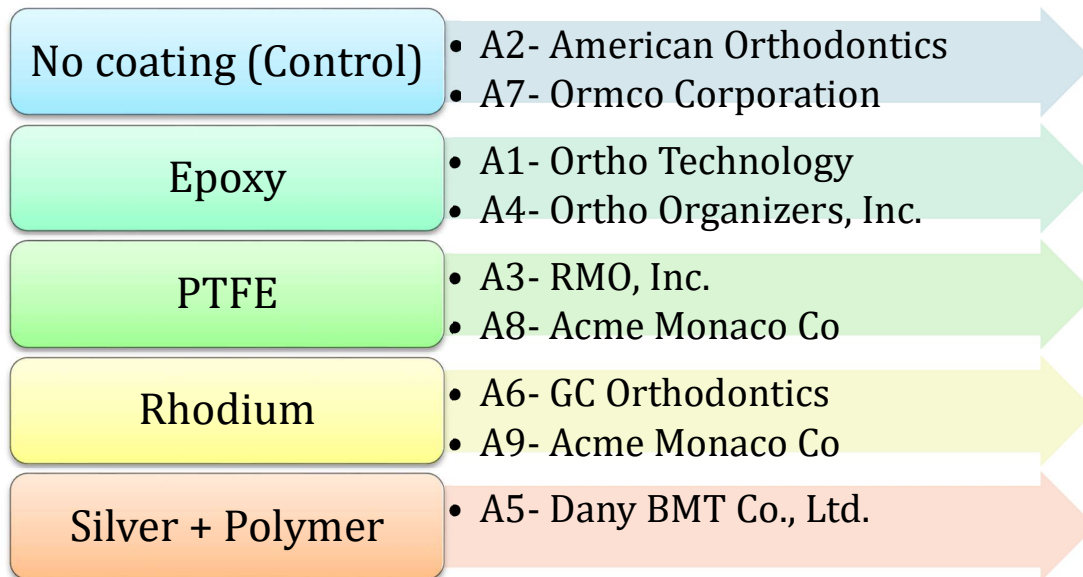


Figure 4 – Diagram illustrating material groups categories

2.4.1 Strains and culture methods

Streptococcus mutans (ATCC® 25175™) was used in all biofilm-formation studies. Bacterial stock was kept at -80°C in Brain Heart Infusion (BHI) Broth + 20%

glycerol. *Streptococcus mutans* was grown from stock on BHI agar for 48 hours at 37°C/5% CO₂. Overnight cultures were made in 5mL of BHI broth and grown statically for 16 hours at 37°C/5% CO₂.

2.4.2 Preparation of orthodontic archwires for biofilm assay

Orthodontic archwires were removed from the suppliers packaging material. Wires were bisected using wire cutters. The distal 1cm of each half of the wire was cut and discarded. Each half of the wire was then cut into three 2cm sections and placed into the wells of a 12-well polystyrene tissue culture plate (CytoOne). Wires were sterilized in the plates by subjecting to ultraviolet light for 30 minutes.

2.4.3 Experimental design to assess *Streptococcus mutans* biofilm accumulation

On the various commercial orthodontic archwires, the ends of archwires were cut off and discarded. To account for the variability in curvature of the wire, each archwire was first bisected and cut into three 2cm sections (A, B, C). Each half of the wire (3 sections) was placed in a column of a sterile, 12-well polystyrene tissue culture plate to assess one individual experimental condition. Two conditions were used: No sucrose, which results in little to no biofilm accumulation and 3% sucrose, which is necessary for *Streptococcus mutans* to make a biofilm. Three sections of wire (A, B, C) from each condition (-/+ sucrose) were subjected to biofilm staining using crystal violet. Similarly, sections of wire (A, B, C) from each condition were subjected to physical biofilm disruption by vortex and ultrasonic bath. Biofilm-associated *Streptococcus mutans* was subsequently determined by colony count. Two separate experiments were performed (n=6 for each condition, each biofilm assessment method).

Figure 1 shows the experimental design to assess *Streptococcus mutans* biofilm accumulation on various commercial orthodontic archwires. A) Ends of archwires were cut off and discarded. To account for the variability in curvature of the wire, each archwire was first bisected and cut into three 2cm sections (A, B, C).

Each half of the wire (3 sections) was placed in a column of a sterile, 12-well polystyrene tissue culture plate to assess one individual experimental condition. Two conditions were used: No sucrose, which results in little to no biofilm accumulation and 3% sucrose, which is necessary for *Streptococcus mutans* to make a biofilm. Three sections of wire (A, B, C) from each condition (-/+ sucrose) were subjected to biofilm staining using crystal violet. Similarly, sections of wire (A, B, C) from each condition were subjected to physical biofilm disruption by vortex and ultrasonic bath. Biofilm-associated *Streptococcus mutans* was subsequently determined by colony count. Two separate experiments were performed (n=6 for each condition, each biofilm assessment method).

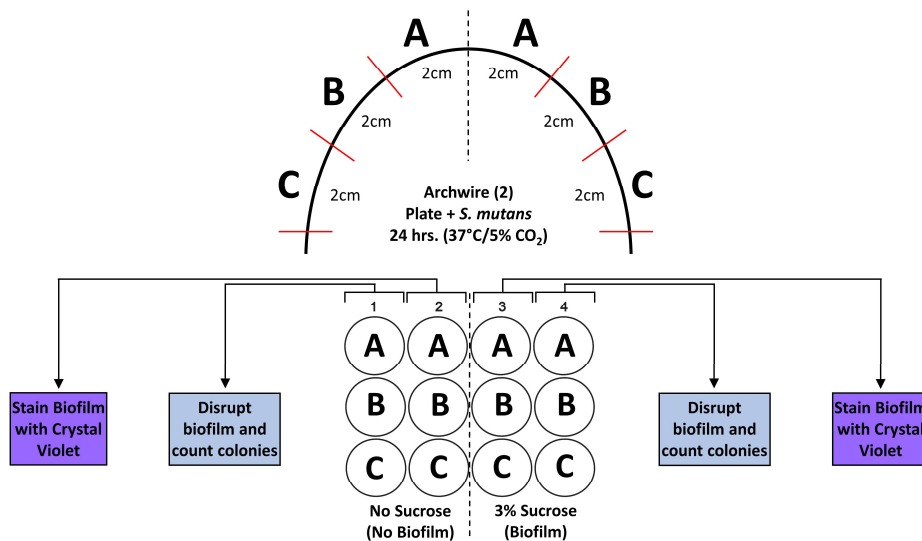


Figure 1 - Experimental design to assess *Streptococcus mutans* biofilm accumulation on various commercial orthodontic archwires

2.4.4 Biofilm Assay

Overnight cultures of *Streptococcus mutans* were normalized to OD₆₀₀ = 1.0 in fresh BHI broth. 500µL of the normalized culture (~1.5 x 10⁸ CFU) was added to each well containing sterilized wires. 440µL of BHI broth was added. To monitor the absence of biofilm, 60µL of sterile deionized water was added (No sucrose). To allow for biofilm accumulation, 60µL of 50% sucrose was added to each well (3% sucrose).

Plates were incubated statically for 24 hours at 37°C/5% CO₂. All wires were simultaneously analyzed on two separate experiments (n=6 sections, 2 half wires total) to control for variability in biofilm accumulation. After incubation, all supernatants were removed and wells were washed once with 1mL 1x PBS. Lastly 3 sections of wire were analyzed in each condition (-/+ sucrose) by crystal violet staining and colony counting.

2.4.5 Crystal Violet Biofilm Staining

After one wash of the wire in the well of the plate, wires were aseptically removed from the plate and placed into a microcentrifuge tube. Wires were washed with 1mL 1x PBS to remove excess planktonic *Streptococcus mutans*. Biofilm was heat-fixed at 80°C for 30 minutes. Then, 1mL of 0.5% crystal violet was added for 30 minutes. Crystal violet stain was aspirated and wires were washed 3 times with water, or until wash was clear of stain. Biofilm-associated stain was solubilized using 33% acetic acid for 10 minutes. Then, 100µL of solubilized crystal violet was added to wells of a clear, flat-bottomed plate in duplicate and absorbance was measured at 570nm. Average of the duplicates was used as the absorbance for each section of wire. Three wells in each condition (-/+ sucrose) were stained in the same way and absorbance readings were used as normalization factor for biofilm accumulation variation between experiments.

This part of the study evaluated crystal violet staining of *Streptococcus mutans* biofilm on orthodontic archwires. Sections of wire (2cm) were placed in 12-well plates with ~1.5x10⁸ CFU staining of *Streptococcus mutans* in BHI with and without the addition of 3% sucrose. Plates were incubated for 24 hours at 37°C/5%CO₂. Wells and wires were washed, heat-fixed, and then stained with 0.5% crystal violet. Crystal violet was solubilized from biofilm using 33% acetic acid. 100µL of stain was added in duplicate to a 96-well plate and absorbance was measured at 570nm. Each bar represents two separate experiments with 3 sections of wire (n=6). Biofilm on plates was measured each experiment to use for normalization of total biofilm. Plate biofilm was normalized to A₅₇₀ = 6.0 and crystal violet staining of wires was normalized using a factor derived from this normalization.

2.4.6 Colony Count Enumeration of Biofilm

After one wash of the wire in the well of the plate, wires were aseptically removed from the plate and placed into a microcentrifuge tube. Wires were washed 3 times with 1mL 1x PBS with gentle inversion to remove excess planktonic *Streptococcus mutans*. 1mL of 1x PBS was added to the wire and mixture was gently inverted. Immediately a 100µL sample was taken for serial dilution and plating on BHI agar for a baseline of free/unbound bacteria remaining after washes. Samples were then vortexed at max speed for 10 seconds. Samples were then placed in an ultrasonic bath for 5 minutes, followed by another 10-second vortex. Serial dilutions were plated on BHI agar and biofilm-associated CFU were enumerated by colony counting.

This section evaluates colony count of biofilm-associated staining of *Streptococcus mutans* on orthodontic archwires. Sections of wire (2cm) were placed in 12-well plates with $\sim 1.5 \times 10^8$ CFU *Streptococcus mutans* in BHI with and without the addition of 3% sucrose. Plates were incubated for 24 hours at 37°C/5%CO₂. Wells were washed once and wires were removed to a sterile tube and washed three times with 1x PBS and gentle inversion. 1mL of 1x PBS was added to the wire and mixture was gently inverted. Immediately a 100µL sample was taken for serial dilution and plating on BHI agar for a baseline of free/unbound bacteria remaining after washes. Samples were then vortex at max speed for 10 seconds. Samples were then placed in an ultrasonic bath for 5 minutes, followed by another 10-second vortex. Serial dilutions were plated on BHI agar and biofilm-associated CFU were enumerated by colony counting. In every case and for each condition (-/+ sucrose), physical disruption of biofilm resulted in an increase in CFU/mL compared to the baseline of free bacteria.

Each bar represents two separate experiments with 3 sections of wire (n=6). Statistical analysis was performed using one-way ANOVA with post-hoc Tukey HSD test. The p-value of the one-way ANOVA results was higher than 0.05 suggesting that wire composition did not affect biofilm accumulation using the colony count method. This was confirmed with the post-hoc Tukey HSD test.

2.4.7 Statistical Analysis

Biofilm accumulation on archwires for both crystal violet staining and colony count enumeration was analyzed using one-way ANOVA with the post-hoc Tukey HSD test. A p-value < 0.05 was considered as statistically significant.

2.5 Results

2.5.1 Crystal Violet Experiments

Tables 2-4 and the graph shown on Figure 2 demonstrate that there was a considerable increase in biofilm on 3% Sucrose solution when compared to No Sucrose experiment in the archwires tested in this study.

Table 2 - Crystal Violet Experiment - No Sucrose

Absorbance at 570nm								
Wire Number	Experiment 1			Experiment 2			Average	Standard Deviation
	A	B	C	A	B	C		
A1	0.0621	0.0633	0.0697	0.0611	0.0640	0.0703	0.0651	0.003923
A2	0.0498	0.0520	0.0534	0.0555	0.0642	0.0625	0.0562	0.005835
A3	0.0640	0.0588	0.0603	0.0619	0.0590	0.0665	0.0617	0.003048
A4	0.0720	0.0692	0.0783	0.1133	0.0789	0.1109	0.0871	0.019732
A5	0.0605	0.0635	0.0596	0.0726	0.0713	0.0706	0.0663	0.005824
A6	0.0615	0.0602	0.0626	0.0659	0.0610	0.0630	0.0624	0.001999
A7	0.0564	0.0596	0.0596	0.0542	0.0518	0.0553	0.0561	0.003074
A8	0.0596	0.0583	0.0644	0.0771	0.0602	0.0678	0.0645	0.007064
A9	0.0671	0.0725	0.0730	0.0581	0.0583	0.0553	0.0640	0.007832

[A1, A4]- Epoxi; [A3, A8]- PTFE; [A6, A9]- Rhodium; [A5]- Silver+Polymer; [A2, A7]- Control

Table 3 - Crystal Violet Experiment - 3% Sucrose

Absorbance at 570nm								
Wire Number	Experiment 1			Experiment 2			Average	Standard Deviation
	A	B	C	A	B	C		
A1	0.1980	0.1437	0.1626	0.2943	0.2807	0.2946	0.2290	0.069105
A2	0.2744	0.2119	0.2667	0.1719	0.3317	0.4177	0.2790	0.087369
A3	0.1344	0.1145	0.1710	0.4510	0.7142	0.5222	0.3512	0.247573
A4	0.1174	0.1224	0.1944	0.4852	0.4409	0.3838	0.2907	0.165343
A5	0.1804	0.1629	0.2344	0.4119	0.4285	0.6079	0.3376	0.174793
A6	0.1655	0.2431	0.1564	0.3706	0.2399	0.2147	0.2317	0.077235
A7	0.1161	0.1445	0.1070	0.4524	0.2306	0.2782	0.2215	0.131729
A8	0.3436	0.1666	0.3960	0.3954	0.6895	0.4586	0.4083	0.170000
A9	0.2042	0.2465	0.1541	0.2587	0.2386	0.3459	0.2413	0.063634

[A1, A4]- Epoxi; [A3, A8]- PTFE; [A6, A9]- Rhodium; [A5]- Silver+Polymer; [A2, A7]- Control

Table 4 - Crystal Violet Experiment - Normalized 3% Sucrose to No Sucrose

Absorbance at 570nm									
Wire #	Experiment 1			Experiment 2			Avg.	Std. Deviation	Fold Change over No Sucrose
	A	B	C	A	B	C			
A1	0.3265	0.2370	0.2682	0.2980	0.2842	0.2983	0.2854	0.030507	4.3867
A2	0.4525	0.3494	0.4398	0.1741	0.3359	0.4230	0.3625	0.104048	6.4494
A3	0.2217	0.1888	0.2820	0.4567	0.7232	0.5288	0.4002	0.207178	6.4828
A4	0.1935	0.2019	0.3205	0.4913	0.4464	0.3886	0.3404	0.124508	3.9084
A5	0.2974	0.2686	0.3865	0.4171	0.4339	0.6156	0.4032	0.123160	6.0796
A6	0.2729	0.4008	0.2579	0.3752	0.2429	0.2174	0.2945	0.075157	4.7238
A7	0.1915	0.2382	0.1765	0.4581	0.2335	0.2817	0.2632	0.102481	4.6918
A8	0.5667	0.2748	0.6531	0.4004	0.6982	0.4644	0.5096	0.160334	7.8963
A9	0.3368	0.4065	0.2541	0.2619	0.2416	0.3502	0.3085	0.065959	4.8201

[A1, A4]- Epoxi; [A3, A8]- PTFE; [A6, A9]- Rhodium; [A5]- Silver+Polymer; [A2, A7]- Control

Table 5 - Normalization to plate biofilm (OD = 6.0)

Dilution	Experiment	Well 1	Well 2	Well 3	AVERAGE	Factor
1:4 Dilution	Exp.1	1.4602	1.4510	1.5326	1.4812	1.01266962
1:4 Dilution	Exp. 2	0.9378	0.8280	0.9629	0.9095	1.64919739

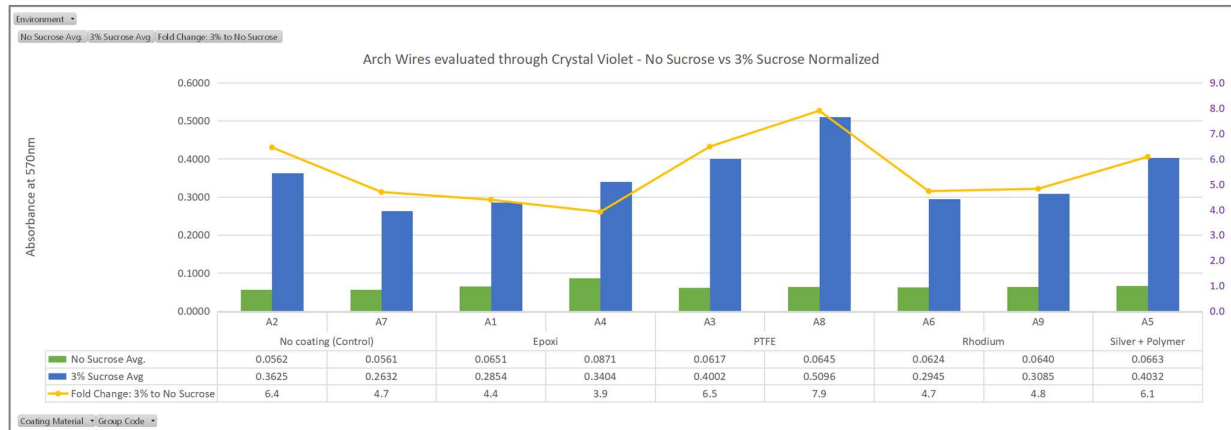


Figure 2 - Arch Wires Crystal Violet Comparison - No Sucrose to 3% Sucrose

Figure 3 shows that the statistical analysis was performed using one-way ANOVA with post-hoc Tukey HSD test. Wire A7 (control) and wire A8 (Teflon) (+ sucrose) were the only two wires with biofilm levels that differed by a statistically significant level (*, p-value < 0.05).

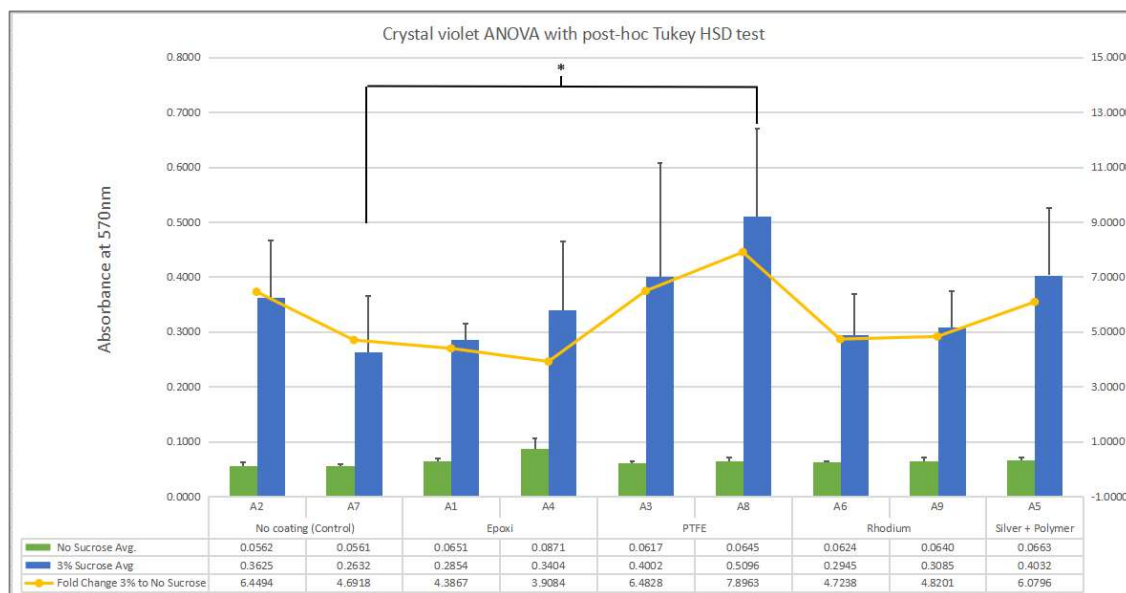


Figure 3 - Crystal violet ANOVA with post-hoc Tukey HSD test

Our Crystal Violet analyses have shown proportional fold increase from no sucrose to 3% normalized sucrose with most increase from wires A8, A3, A2 and A5 and least increase on wires A4, A1 and A7. In other words, Teflon (A3, A8) presented the highest biofilm formation and Epoxi Resin (A1, A4) the lowest biofilm. Statistical analysis show that wires A7 and A8 (+ sucrose) were the only two wires with biofilm levels that differed by a statistically significant level (*, p -value < 0.05) A7, A8, control and Teflon, respectively.

2.5.2 Colony Count

Tables 6 and 7 and Figure 4 show that groups A7 (control 0), A1 (Epoxi) and A9 (Rhodium) presented the highest colony counting numbers when compared with the other archwires; however, we did not find any significant statistical difference among the wires tested for the colony count analyses. Our study found that Epoxy, Control and Rhodium coated archwires presented the highest biofilm formation on 3% sucrose (between 305 to 386 CFU/mL), and the highest fold increase in biofilm (126, 138, 136 times) from no sucrose to 3% sucrose environment.

Table 6 - Colony Count Post-Vortex/Sonication (CFU/mL) – No Sucrose

NO SUCROSE Wire Number	Experiment 1			Experiment 2			Average	Standard Deviation
	A	B	C	A	B	C		
A1	1,500	500	900	2,200	1,100	8,300	2,416.67	2,939.67
A2	400	100	100	21,000	11,300	31,500	10,733.33	13,190.10
A3	100	400	600	49,000	33,300	31,200	19,100.00	21,424.29
A4	900	2,400	500	12,900	13,300	35,800	10,966.67	13,491.87
A5	700	7,100	1,300	16,400	4,800	22,500	8,800.00	8,788.63
A6	900	1,500	700	10,200	6,700	700	3,450.00	4,038.69
A7	400	200	500	5,500	4,000	5,100	2,616.67	2,515.09
A8	600	1,800	600	24,300	16,400	8,200	8,650.00	9,815.45
A9	900	400	600	5,600	8,600	1,000	2,850.00	3,432.64

[A1, A4]- Epoxi; [A3, A8]- PTFE; [A6, A9]- Rhodium; [A5]- Silver+Polymer; [A2, A7]- Control

Table 7 - Colony Count Post-Vortex/Sonication (CFU/mL) - 3% Sucrose

3% Sucrose Wire Number	Experiment 1			Experiment 2			Average	Standard Deviation	Fold Change over No Sucrose
	A	B	C	A	B	C			
A1	26,000	5,000	32,000	223,000	1,240,000	302,000	304,666.67	473,988.89	126.07
A2	10,000	1,000	0	590,000	198,000	324,000	187,166.67	237,568.03	17.44
A3	11,000	1,000	9,000	221,000	246,000	120,000	101,333.33	111,668.56	5.31
A4	20,000	12,000	6,000	231,000	392,000	436,000	182,833.33	198,561.24	16.67
A5	5,000	5,000	19,000	165,000	333,000	520,000	174,500.00	212,708.02	19.83
A6	8,000	32,000	51,000	155,000	68,000	690,00	63,833.33	50,300.76	18.50
A7	18,000	51,000	19,000	330,000	730,000	1,020,000	361,333.33	424,762.36	138.09
A8	10,000	3,000	4,000	191,000	324,000	207,000	123,166.67	136,682.72	14.24
A9	6,000	7,000	1,000	1,610,000	331,000	363,000	386,333.33	622,574.55	135.56

[A1, A4]- Epoxi; [A3, A8]- PTFE; [A6, A9]- Rhodium; [A5]- Silver+Polymer; [A2, A7]- Control



Figure 4 - Arch Wires Colony Count Comparison - No Sucrose to 3% Sucrose

Figure 5 is the logarithmic scale representation in order to visually illustrate the significant discrepancy among the results from 0% sucrose to 3% sucrose. For instance, group control A7 presented 2,617 colonies in 0% and 361,333 colonies in 3% sucrose.

On the ANOVA statistical analyses from Figure 5 the bar represents two separate experiments with 3 sections of wire (n=6). Statistical analysis was performed using one-way ANOVA with post-hoc Tukey HSD test. The p-value of the one-way ANOVA results was higher than 0.05 suggesting that wire composition did not affect biofilm accumulation using the colony count method. This was confirmed with the post-hoc Tukey HSD test.

Also on Figure 5, colony count analyses showed a very significant increase in colony formation from no sucrose condition to 3% sucrose. Groups A1, A7 and A9 were >120 times the baseline with a max of 138 from wire A7. The wire with the least amount of change over initial condition was wire A3 with only 5 times increase. Statistical analyses didn't demonstrate that there was a significant difference among the different archwires. Arch wires with PTFE coating demonstrated the least increase in colony count and the final colony count with 3% sucrose were between the 3 wires with the least values (~101-123).

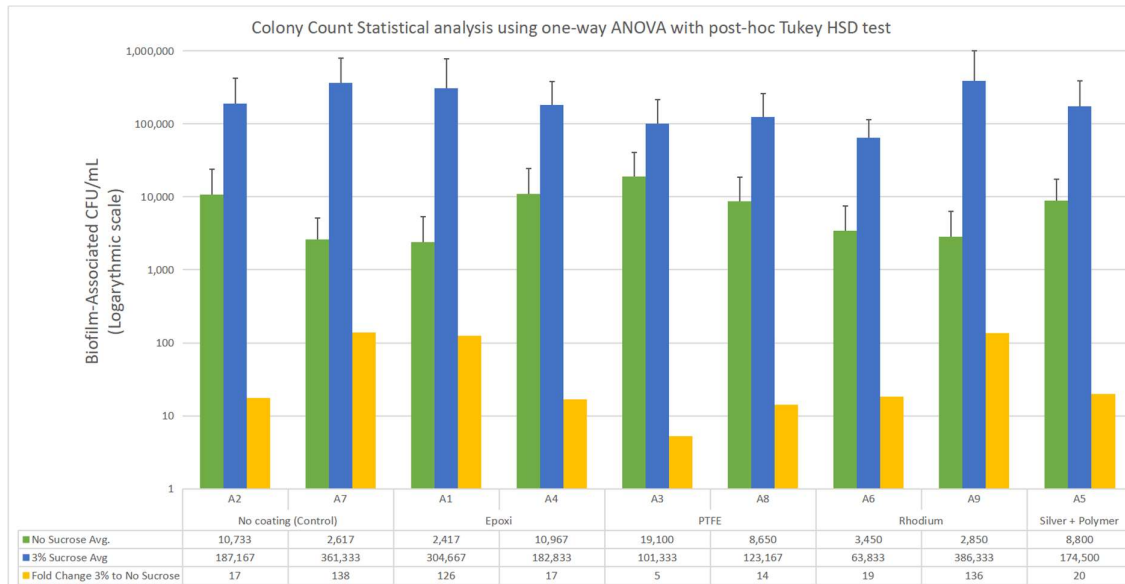


Figure 5 - Colony Count Statistical analysis using one-way ANOVA with post-hoc Tukey HSD test

2.6 Discussion

Aesthetic wires are commonly used in orthodontic treatments. Surface roughness is an important factor in the friction and bacterial formation in these wires. However, aesthetic wires have not been accessed clearly in the literature.

Epoxy resin is basically a synthetic resin composed of the combination of epoxy and other polymers. Among its properties, we can highlight its formation to the wire metal, resistance to solubility, electrical insulation, and dimensional stability (KRAVITZ, 2013).

Our study found that Epoxy, Control, Rhodium coated archwires presented the highest biofilm formation on 3% sucrose (between 305 to 386 CFU/mL) and the highest fold increase in biofilm (126, 138, 136 times) from no sucrose to 3% sucrose ambient.

Rongo et al., 2016, supports our results. They evaluated three fully coated, tooth-colored NiTi wires (BioCosmetic, Titanol Cosmetic, EverWhite), two ion-implanted wires (TMA Purple, Sentalloy High Aesthetic), five uncoated NiTi wires (BioStarter, BioTorque, Titanol Superelastic, Memory Wire Superelastic, and Sentalloy), one β -titanium wire (TMA), and one stainless steel wire (Stainless Steel) were considered for this study.

Under the experimental conditions, all of the NiTi esthetic archwires resulted in slight increase in bacterial counting, as did the respective uncoated wires. In other words, they found that the NiTi esthetic archwires Epoxy coated, resulted in slight cytotoxicity, as did the respective uncoated wires. For this reason, their clinical use may be considered to have similar risks to the uncoated archwires.

In contrast, Mousavi et al., 2017 compare the surface roughness of 4-coated esthetic wires with that of a conventional orthodontic wire. NiTi Memory wire (American Orthodontics, USA) as a control group; Orthocosmetic Elastinol (Ortho Organizers, USA); Perfect (Hubit, Korea); Imagination (Gestenco, Sweden); EverWhite (American Orthodontics, USA). The average range of the 4 parameters was the highest for the uncoated Ni-Ti Memory wire (control group) while the Perfect coated wire showed the lowest values. Significant differences existed between

uncoated and coated wires, being higher for the uncoated wires. Taking into account the study limitations, the surface roughness values of NiTi uncoated archwires were significantly higher than those of the coated wires. This study disagrees with our findings that Epoxy, Rhodium and no coated archwires presented similar property behavior.

Regarding PTFE coating archwires, PTFE (polytetrafluoroethylene), popularly known by the trademark "Teflon®", is a synthetic polymer consisting primarily of carbon and fluorine. Due to the strength of carbon-fluorine bonds, PTFE has the third lowest coefficient of friction of any known solid, heat-resistant, and hydrophobic (KRAVITZ, 2013). Some PTFE coated Cu-NiTi metal wires (for example, FLI®, Rocky Mountain Orthodontics) are available in the market.

Our Crystal Violet analyses has shown that proportional fold increase from no sucrose to 3% normalized sucrose with most increase from wires A8, A3, A2 and A5 and least increase on wires A4, A1 and A7. In other words, Teflon (A3, A8) presented the highest biofilm formation and Epoxi Resin (A1, A4) the lowest biofilme. Statistical analysis show that wires A7 and A8 (+ sucrose) were the only two wires with biofilm levels that differed by a statistically significant level (*, p-value < 0.05) A7, A8, control and Teflon, respectively.

According to the literature, Simion et al., 1994 found that the technique of guided tissue regeneration using expanded polytetrafluoroethylene (ePTFE) membranes has been shown to be effective in implant dentistry (bony defects, extremely thin alveolar ridges, and implants placed in fresh extraction sockets). Their study showed the oral bacteria formation to PTFE may contaminate ePTFE membranes exposed to the oral cavity. The specimens showed partial bacterial penetration after 2 and 3 weeks, however; after 4 weeks, all membrane specimens demonstrated bacterial contamination. These results agreed with our finds that Teflon (PTFE) presented the highest biofilm formation.

In addition, Gyo et al., 2008 evaluated experimental resin composites with incorporated polytetrafluoroethylene (PTFE) particles, which theoretically could improve the surface properties of the materials, including the inhibition of bacterial adherence. Results showed hydrophobicity improvement of the resin composite by

incorporation of PTFE fillers. However, surface resistance against biofilm formation was not improved. These results agreed with our finds that Teflon (PTFE) presented biofilm formation.

However, Demling et al., 2010 disagree with our find that Teflon presented more biofilm accumulation. The authors evaluated PTFE coated and uncoated stainless-steel brackets were bonded symmetrically on the first or second primary molars in 13 adolescent patients for 8 weeks. Their study revealed a significantly lower biofilm accumulation on PTFE coated brackets on all surfaces. The results indicate that PTFE coating of brackets reduces biofilm formation to a minimum and might have the potential to reduce iatrogenic side effects, e.g. decalcification during orthodontic treatment with fixed appliances.

Another study by Fuchslocher et al., 2013, which evaluated PTFE coated and stainless steel probes included 10 healthy volunteers (5 females and 5 males) with a mean age of 27.3 ± 3.7 years. Three different slabs (two PTFE coatings: one pure and one ceramic-reinforced polytetrafluoroethylene, and stainless steel) were placed in random order on a splint in the mandibular molar region. Their results indicate that the beneficial surface characteristics of PTFE coatings by reducing long-term biofilm, but they are not a result of inhibiting initial bacterial formaton.

The results from microbiological and bacterial counting experiments in this study indicate that there wasn't a significant difference among the distinct aesthetic archwires materials and coating and in comparison to conventional metallic archwires. These findings show us that the manufacturers are investing heavily in technology and they are focusing on material performance to deliver the most efficient product to the market. So, the esthetic archwires have similar behavior to the conventional metal archwires and the orthodontists can exchange materials without changing their treatment biomechanics.

However, giving a more detailed look into the microbiological analyses, the test of crystal violet indicated that the Teflon coating was the only aesthetic wire that presented a statistically significant difference in relation to the control group.

2.7 Conclusions

In conclusion, based on this *in vitro* study limitation, the clinician should not be concerned about bacterial colony counting properties of esthetic coated archwires when compared to conventional metallic archwires; however, the Teflon coated archwires was the only coated wire statistically different that presented higher biofilm accumulation than conventional archwires, which could contribute to increase the risk to develop dental caries in patients in treatment with such wires.

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3. Capítulo 2: Topographic Surface and Roughness of Aesthetic Orthodontic Archwires Before and After Brushing Cycles

3.1 Introduction

Orthodontic archwires are made of different alloys, such as: Stainless steel, cobalt-chromium-nickel, cobalt-chromium nickel-titanium are some of the alternatives of orthodontic wires available during all phases of treatment (O'BRIEN, 2008; ELIADES, 2012). Although these wires are strong and durable, they are not aesthetically pleasing because of their metallic color, in addition, they have the potential to cause allergic or toxic reactions due to contact with soft or hard tissues of the mouth (ELIADES, ATHANASIOU, 2002; HENSTEN-PETTERSEN, JACOBSEN, 2003).

Futhermore, the surface topography can affect the friction behavior between an orthodontic wire and brackets during clinical applications. A study by Huang (2007) investigates the influence of a fluoride-containing environment on the surface topography variations of different nickel-titanium (NiTi) orthodontic archwires. The NiTi commercial orthodontic archwires were immersed in fluoride mouthwashes and in artificial saliva with the addition of commercial fluoride toothpastes or prophylactic gels for a 28-day period. Atomic force microscopy (AFM) was used to measure the three-dimensional surface topography of NiTi archwires before and after the immersion tests. Regardless of the archwire, no statistically significant difference was observed on the tested NiTi archwires in lower fluoride-containing (<2500 ppm) environments, including the various fluoride mouthwashes and the artificial saliva added with fluoride toothpastes. In artificial saliva added with high fluoride prophylactic gel (around 17,000 ppm), a significant increase in the surface roughness, was observed on the tested NiTi archwires. Isac et al. (2015) evaluated the changes in surface topography and roughness of stainless steel (SS), nickel-titanium and beta-titanium (β -Ti) archwires after clinical use and sterilization. Scanning electron microscope images revealed an increase in surface irregularities in SS and nitinol wires after clinical use. There was a significant increase in Ra values of SS orthodontic wires after intra-oral exposure. In conclusion, the surface

roughness of SS wires increased significantly after clinical use. Autoclave sterilization did not affect considerably on surface characteristics of any archwire.

Also, Syed et al. (2015) evaluated stainless steel (Ormco, CA, USA), titanium molybdenum alloy (Ormco, CA, USA) and nickel-titanium (G and H Wire Company, USA) orthodontic wires with a rectangular cross-section dimension of 0.019"× 0.025", were selected. The wires were later coated with a uniform and smooth nanoparticle film using 100 ml nanocremls. The coating procedure was verified by comparing the surface topography of nanocoated archwires with the commercially available archwires in an environmental scanning electron microscope (ESEM). The ESEM images prove that the surface topography of the coated wires was found to be smoother with less surface deteriorations as compared to the commercially available wires.

Mane et al. (2012) studied rectangular NiTi and CuNiTi wires immersed in fluoride solution and artificial saliva (control) for 90 minutes at 37°C. The acidulated fluoride agents appeared to cause greater corrosive effects as compared to the neutral fluoride agents. The results suggest that using topical fluoride agents leads to corrosion of surface topography indirectly affecting the mechanical properties of the wire that will lead to prolonged orthodontic treatment.

Considering the dynamic aesthetic advances in the world and increased demand of patients for aesthetic, Orthodontics, as the oldest dental specialty, introduced ceramic brackets and aesthetic coated archwires(IIJIMA et al., 2012). Despite the contribution to the aesthetic gain of these arcs, the literature reports relatively little scientific evidence on these aesthetic materials due to its relatively recent introduction, which leads to the insecurity of many orthodontists using this material. In addition, the possible fragmentation of the aesthetic coating and greater resistance to the friction of these materials can generate uncertainty as to their use in clinical practice .

Currently, orthodontic aesthetic arches can be divided into two categories: [1] Metallic coated and [2] translucent (non-metallic) (LEE, 2008; YU LEE, 2011). Regarding coact aesthetics archwires, polymer-coated alloy wires were developed as cosmetic orthodontic wires in the early 2000 (ELAYYAN et al., 2008; ZEGAN et al., 2012; Da SILVA et al., 2013).

For example, resin materials reinforced with fiberglass have been widely used in dentistry for decades, but its application as an alternative to orthodontic metallic wires (CACCIAFESTA et al., 2008) has been recent. Currently, metallic arches can be coated metallic as rhodium for better aesthetic effect, or as polymers such as poly-tetra-Fluoroetileno [PTFE] (Teflon ®) or epoxy resin (diethylene glycol monobuty ether 1-5%, titanium Dioxide 1-5%, ethylene- 2ethylhexyl Glycolm ether, methyl acetate 0, 1% ketone. The PTFE (polytetrafluoroethylene), popularly known by the commercial brand "Teflon ®", is a synthetic polymer that basically consists of carbon and fluoride. Due to the strength of the carbon-fluoride bonds, the PTFE has the third lowest coefficient of friction of any known solid, heat-resistant, and hydrophobic (KRAVITZ, 2013).

Some PTFE-coated Cu-NiTi metallic wires (e.g. FLI ®, Rocky Mountain Orthodontics) are available on the market. The epoxy resin is basically a synthetic resin composed by the combination of epoxy and other polymers, among its properties, we can emphasize its adherence, resistance to solubility, electrical insulation, and dimensional stability. The threads coated by epoxy resin (e.g. Ultraesthetic™, G&H ® Orthodontics) are commonly coated on all surfaces of the wire or only the vestibular face of the wire to minimize friction problems and generally, extends to the second molar. The adhesion between the arc and coating continues to be a problem because the coating can separate from the metallic arc due to the mastication forces and activation of digestive enzymes. For example, about 25% of epoxy coating is lost during the first month of in vivo treatment (KRAVITZ, 2013) due to this low adhesion.

For translucent (non-metallic) arcs we can also find resin wires reinforced with glass fibers (FRP). These arcs (e.g. SimpliClear™, Biomers products LLC) present little plastic/elastic deformation, and can be considered crispy materials when compared to conventional metallic wires (KRAVITZ, 2013). Its use is limited to class I patients, with mild to moderate dental crowding, being contraindicated for patients requiring previous-posterior fixes. Fiberglass reinforced arches (translucent arcs) present mechanical properties, such as flexural resistance, something similar to the metallic arches of NiTi (BALLARD et al., 2012). These arcs show high translucency

compared to metallic arches (KRAVITZ, 2013) and are indicated for use with ceramic brackets.

As mentioned aesthetics coated archwires still pretty new in the market. They were first used in the early 2000; however, its demand has increaced over the years. Therefore, the scientific evidentce on its biological and mechanical properties still been established in the literature. Thus, this study purpose is to evaluate the aesthetic coated archwires mechanical properties, related to the surface roughness and its topographic characteristics.

3.2 Objectives

- A) Evaluate the topographic surface of orthodontic aesthetic archwires.
- B) Investigate the superficial roughness of orthodontic aesthetic archwires before and after brushing cycles.

3.3 Hypotheses

Orthodontic aesthetic archwires will present higher superficial roughness when compared to conventional metallic orthodontic archwires due to distinguished topographic surface.

3.4 Methodology

Seven commercially aesthetic orthodontic archwires brands and two conventional metallic orthodontic archwire used on the experiments are described on Table 1. They were rectangular nickel-titanium alloy arches measuring 0.016 "x 0.022" forming 9 sample type groups (n = 10) for the evaluation of the average roughness parameter (Ra) in μm . Total of 10 specimens of each type of wire were tested.

All the archwires used in the study were pre-contoured, and the anterior curve region was discarded, and only the straight segments were used to compose the

sample units. They were used as received from the manufacturer and clamped to a glass plate using plastic clamps coated with nylon to attach to the table of measurements.

Table 8 - Archwires specification and manufacturer information

GROUP CODE	PRODUCT	MANUFACTURER	SPECIFICATION	LOT#	QTY
A1	Tooth Tone® Coated Archwire	Ortho Technology	16x22 NiTi	PO19425	5
A2	NT3™ SE	American Orthodontics	16x22 NiTi	G74366	50
	NT3™ SE	American Orthodontics	16x22 NiTi	149464	20
	NT3™ SE	American Orthodontics	16x22 NiTi	152960	20
A3	FLI® Wire	RMO, Inc.	16x22 NiTi	WO-786152	5
	FLI® Wire	RMO, Inc.	16x22 NiTi	WO-727181	10
A4	Nitanium® Super Elastic	Ortho Organizers, Inc.	16x22 NiTi	183877	5
A5	Dany Coated Archwire	Dany BMT Co., Ltd.	16x22 NiTi	362412	4
A6	Bio-active RC	GC Orthodontics	16x22 NiTi RC	A346	5
A7	Copper Ni-Ti®	Ormco Corporation	16x22 NiTi Cu	51768825	20
A8	Niti Micro Dental White	Acme Monaco Co	16x22 NiTi		
A9	NiTi Dental White S	Acme Monaco Co	16x22 NiTi		

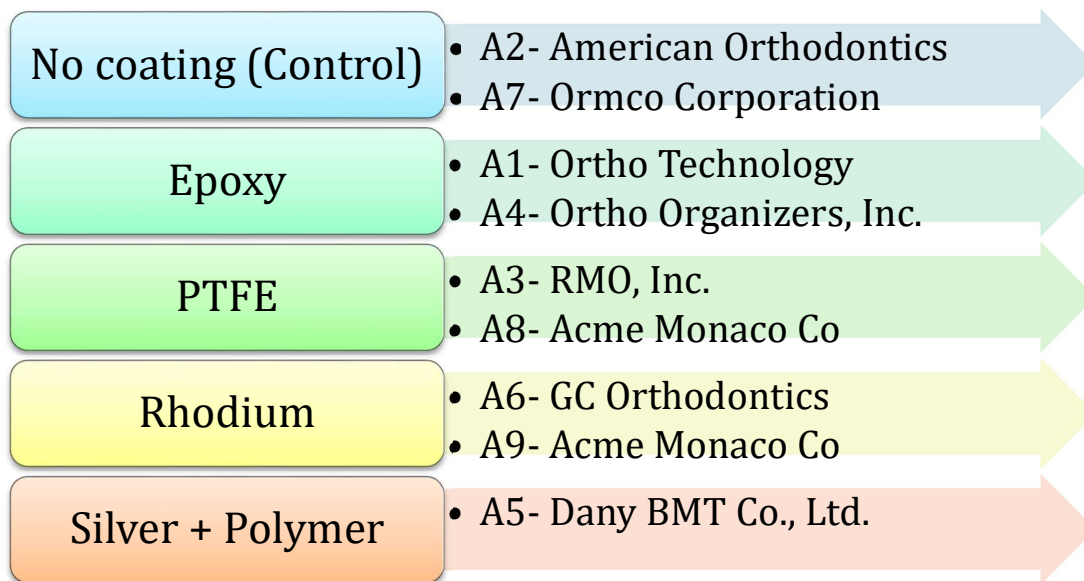


Figure 4 – Diagram illustrating material groups categories

3.4.1 Topographic Evaluation in Scanning Electron Microscopy (SEM)

Scanning electron microscopy (SEM) was performed as additional evaluation technique of the samples. The brands choice was made for convenience and available as received. A SEM machine (JSM 5600LV, Jeol, Japan) with a secondary electron detector (SE) allowed the preview of topography with high depth of focus.

Evaluated orthodontic materials were prepared on aluminum stubs ($n = 10$) and covered with a thin layer of gold-palladium (MED 010, Balzers Union, Liechtenstein), then observed in SEM (JSM 5600LV, Jeol) operating at 15 kV, in increments of 50 to 5,000X. Total of 10 specimens of each type of wire were tested.



Figure 5 – Scanning electron microscopy (SEM). SEM machine (JSM 5600LV, Jeol, Japan) with a secondary electron detector (SE) SEM Images (50x, 200x, 400x, 1000X)

Data were obtained through comparison of the images from archwires to an endodontic index modified according the orthodontic needs (Van Eldik et al., 2004; Perakaki et al., 2007). Debris scores: A - total absence of debris (index 0); B - presence of debris in less than 1/4 of the image (index 1); C - presence of debris between 1/4 and 3/4 of the image (index 2); and D - presence of debris in more than 3/4 of the image (index 3).

A single operator previously trained about the index protocol performed the categorization of the SEM photos.

3.4.2 Superficial Roughness and Simulated Toothbrush Abrasion

Samples of 10 mm of the different evaluated orthodontic archwires were made (n = 10). The samples were immersed in distilled water and subjected to ultrasonic cleaning for 5 minutes prior to analysis and prior to cycling procedures. A single operator previously trained about the protocol performed the measurements. The instrument was calibrated before the measurement in each group.

The surface roughness of each group was evaluated with a surface profilometer (Surf-Corder SE 1700, Kosaka Laboratory Ltd., Japan). Three measurements were made for each specimen at different locations and the mean was designated as the roughness value of each specimen. The parameters used were screening length of 1.25 mm, cut-off point of 0.25 mm, and measurement speed of 0.5 mm/s. Measurements were performed before and after a simulated toothbrush abrasion procedure. For the brushing simulation, soft-bristled toothbrushes (Sorriso; Colgate) were mounted on a brushing machine (MEV 3Y-XT; Odeme, Luzerna, SC, Brazil). Specimens per evaluated after 1200, 2400 and 3600 cycles of brushing applied in a linear movement of 2 cm, using a 1:3 wt% ratio dentifrice/water slurry (Oral-B® Pro-Saúde, Procter & Gamble. São Paulo, SP. Brazil).

Table 9 - Archwire group and Coating Type

GROUP CODE	PRODUCT	MANUFACTURER	Coating type
A1	Tooth Tone® Coated Archwire	Ortho Technology, BR	Epoxy Resin
A2	NT3™ SE	American Orthodontics, USA	No coating (Control)
A3	FLI® Wire	Rock Mountain Ortho, Inc. USA	PTFE
A4	Nitanium® Super Elastic	Ortho Organizers, Inc. USA	Epoxy Resin
A5	Dany Coated Archwire	Dany BMT Co., Ltd. Korea	Silver + Polymer
A6	Bio-active RC	GC Orthodontics, Japan	Rhodium
A7	Copper Ni-Ti®	Ormco Corporation, USA	No coating (Control)
A8	Niti Micro Dental White	Acme Monaco Co, USA	PTFE
A9	NiTi Dental White S	Acme Monaco Co, USA	Rhodium

[A1, A4]- Epoxi; [A3, A8]- PTFE; [A6, A9]- Rhodium; [A5]- Silver+Polymer; [A2, A7]- Control

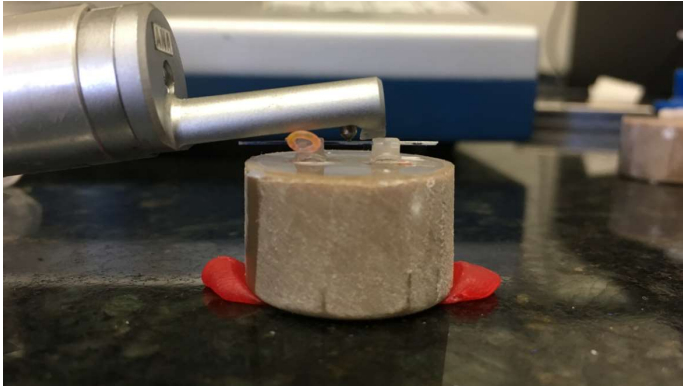


Figure 6 – Superficial Roughness measured BEFORE and AFTER Simulated Toothbrush Abrasion



Figure 7 – Brushing machine (MEV 3Y-XT; Odeme, Luzerna, SC, Brazil)

3.4.3 Statistical Analysis

Analysis of variance was performed considering the “brand factor” (different manufacturers) and the “area factor” (top or side of each archwire) with a complementary Tukey test ($\alpha=5\%$). Data were converted using a logarithmic function.

3.5 Results

3.5.1 Scanning Electron Microscopy (SEM) Results

Data were obtained through comparison of the images from archwires to an endodontic index modified according the orthodontic needs (Van Eldik et al., 2004; Perakaki et al., 2007). Debris scores: A - total absence of debris (index 0); B - presence of debris in less than 1/4 of the image (index 1); C - presence of debris between 1/4 and 3/4 of the image (index 2); and D - presence of debris in more than 3/4 of the image (index 3) were described, according to the classification of images on Figures 8 to 10.

Based on the classification of each SEM images, PTFE was the only archwire which showed the presence of debris in more than 3/4 of the images, which was determined to be index 3.

Figure 8 shows that the control group (A2) had index 0 (zero), and the epoxy groups, A1 and A4 had index 1.

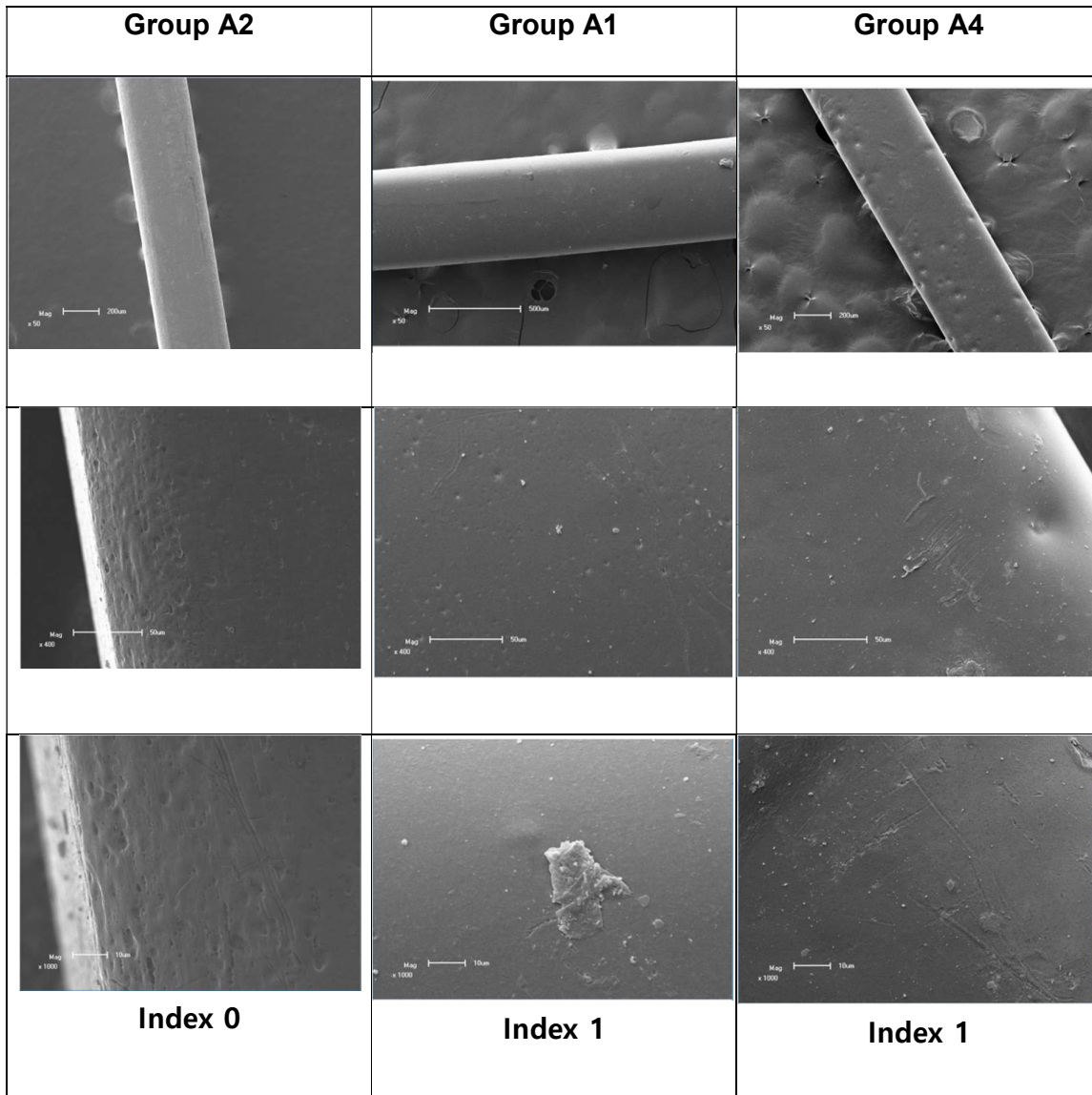


Figure 8 – Archiwires Group A2, A1, A4. SEM Images (50x, 400x, 1000X)

[A1, A4]- Epoxi; [A3, A8]- PTFE; [A6, A9]- Rhodium; [A5]- Silver+Polymer; [A2, A7]- Control

Figure 9 shows that the control group A7 had index 2 , and the Teflon groups presented index 3 (A3) and index 1 (A8), respectively.

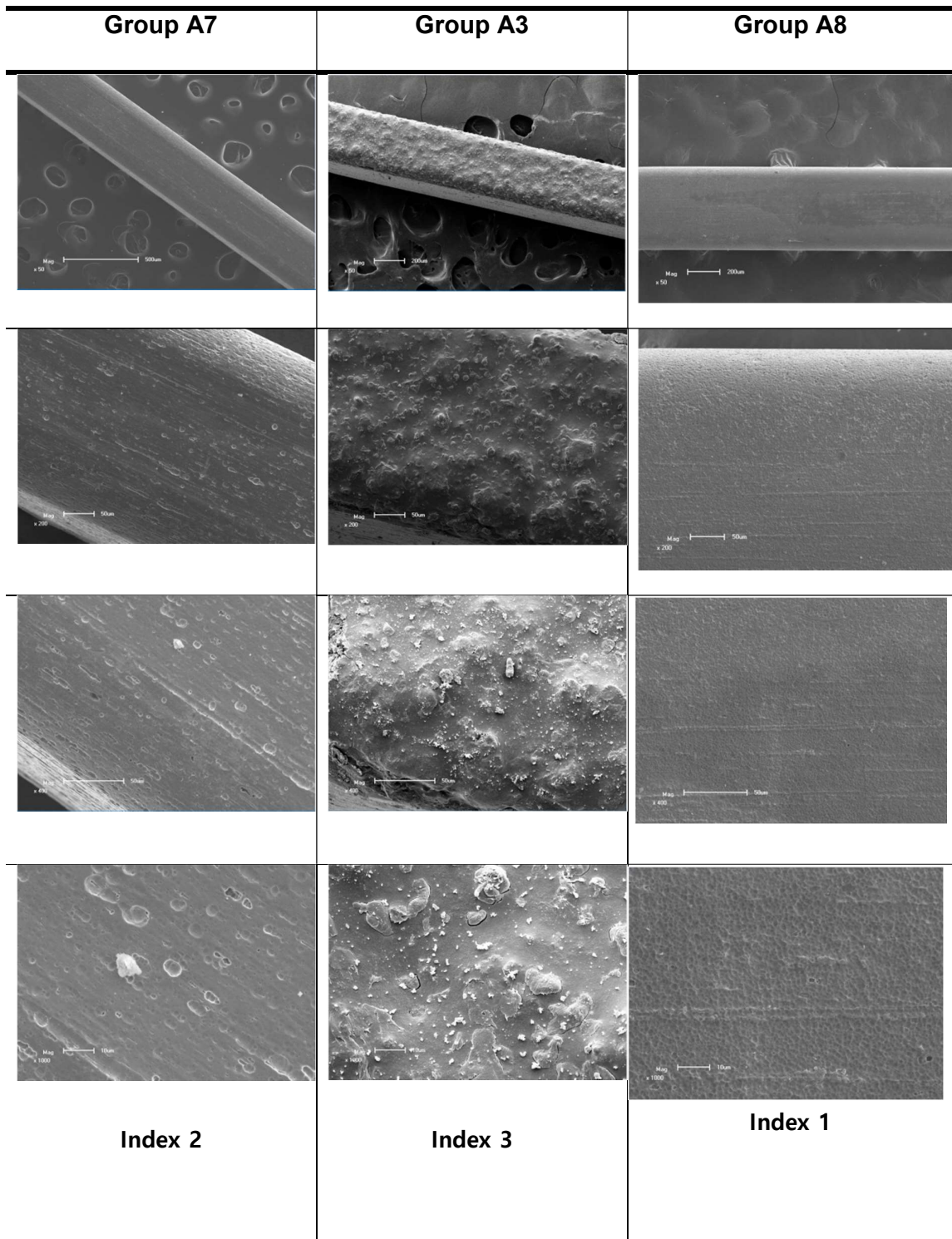


Figure 9 – Archiwires Group A7, A3, A8. SEM Images (50x, 200x, 400x, 1000X)

[A1, A4]- Epoxi; [A3, A8]- PTFE; [A6, A9]- Rhodium; [A5]- Silver+Polymer; [A2, A7]- Control

Figure 10 demonstrates that group A5, Silver+polymer had index 2, and the rhodium groups, A6 and A9 had index 1.

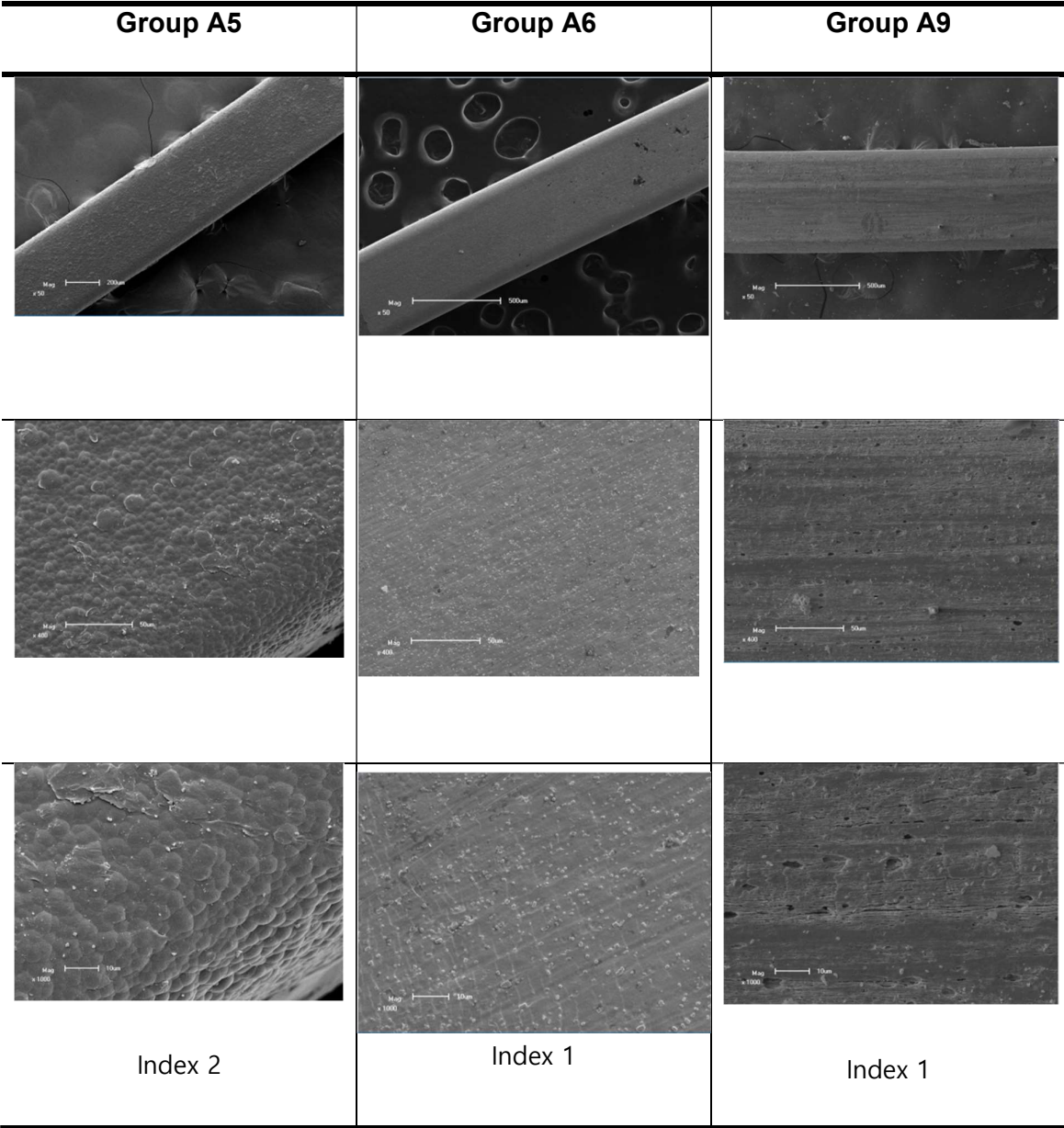


Figure 10 – Archiwires Group A5, A6, A9. SEM Images (50x, 400x, 1000X)

[A1, A4]- Epoxi; [A3, A8]- PTFE; [A6, A9]- Rhodium; [A5]- Silver+Polymer; [A2, A7]- Control

3.5.2 Superficial Roughness Results

The results of the calculation of roughness were obtained by average of the average for each sample and the standard deviation.

Table 10 shows the results of roughness for each group of arch wire according to brushing cycles. In other words, according to time and not among the archwire groups. Based on Table 10 results, group A8, Teflon, was the only group with roughness reduction that was significant statistically different after 3600 cycles, which corresponds to 3 months of brushing. However, the control groups and the Rhodium (A6) were the most stable groups according to initial roughness and after 3 months of brushing.

Table 10 - Surface roughness results μm Average and (Standard Deviation)

Group Code	Coating Type		Initial		1200 cycles		2400 cycles		3600 cycles
A2	Control	A	0.63 (0.29)	A	0.64 (0.26)	A	0.81 (0.18)	A	0.71 (0.24)
A7	Control	A	0.49 (0.16)	A	0.75 (0.28)	A	0.66 (0.28)	A	0.69 (0.08)
A1	Epoxy Resin	B	0.82 (0.46)	A	1.43 (0.58)	A	1.52 (0.61)	AB	1.16 (0.55)
A4	Epoxy Resin	AB	1.45 (0.67)	A	1.64 (0.60)	B	0.80 (0.37)	B	0.77 (0.26)
A3	PTFE	A	4.24 (0.80)	A	3.63 (0.89)	AB	2.60 (0.30)	B	2.16 (0.20)
A8	PTFE	A	1.56 (0.34)	AB	1.48 (0.44)	B	1.12 (0.35)	C	0.70 (0.17)
A6	Rhodium	A	0.71 (0.41)	A	0.76 (0.39)	A	0.88 (0.34)	A	0.66 (0.05)
A9	Rhodium	A	3.83 (0.83)	AB	2.76 (0.55)	B	2.05 (0.34)	B	1.90 (0.14)
A5	Silver + Polymer	A	1.71 (0.37)	A	1.39 (0.42)	B	0.79 (0.23)	B	0.79 (0.22)

Figures 6 through 9 provide a graphical representation on the evolution of roughness for each arch wire group according to brushing cycles.

Figure 6 confirm our finds on Table 10 were the Teflon group had the highest initial roughness among all wires.

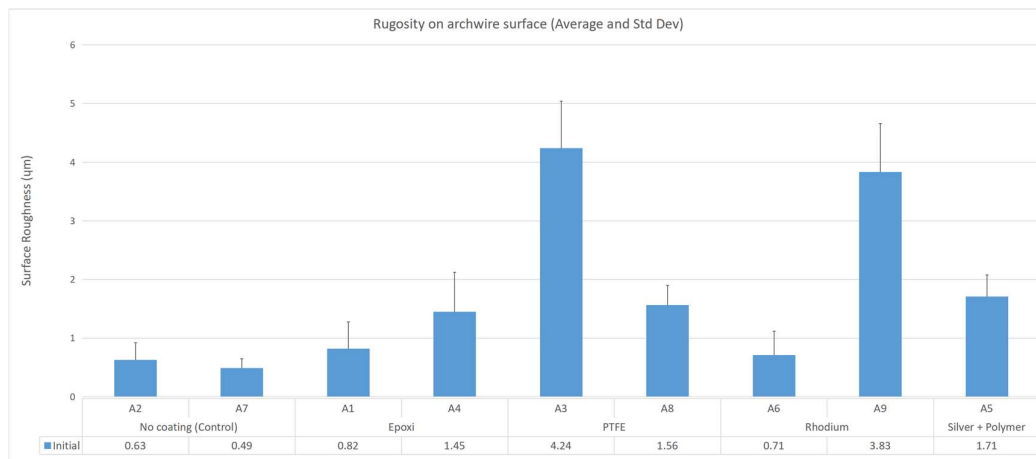


Figure 6 - Graphical representation of surface roughness (Average and Standard Deviation).

Figure 7 shows the superficial roughness performance of all groups initially and after 3600 brushing cycles. The control groups and the A1, Epoxy group were the only wires to present an increase in the surface roughness after 3 months of brushing cycles.

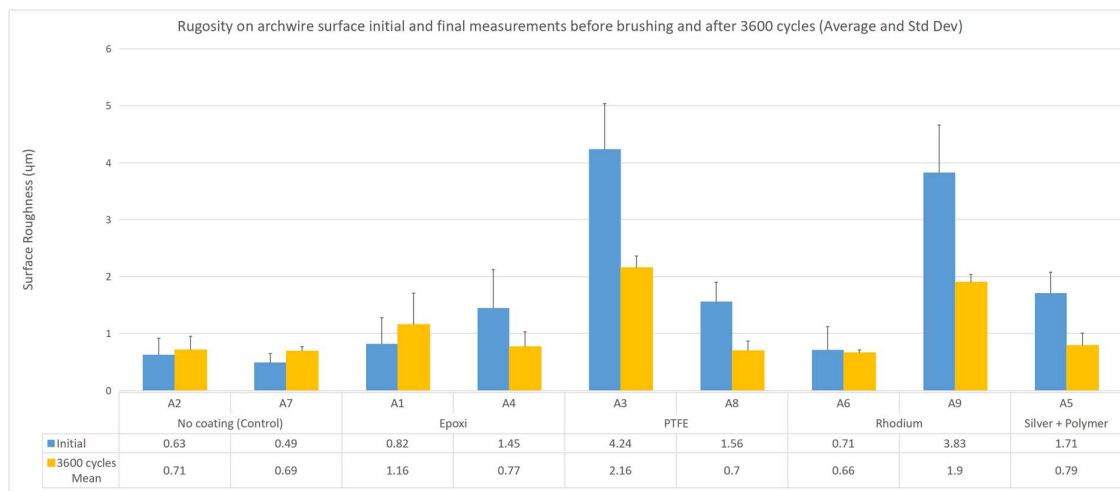


Figure 7 - Graphical representation of surface roughness in µm before brushing and after 3600 brushing cycles (Average and Standard Deviation) by arch wire group

Figure 8 demonstrates that the archwire with most significant superficial roughness reduction from initial to after 3 months of brushing was A3, the Teflon group. And the most stable groups were both control groups and A6, Rhodium, which clinically is a more desirable outcome to predict wire behavior during treatment.

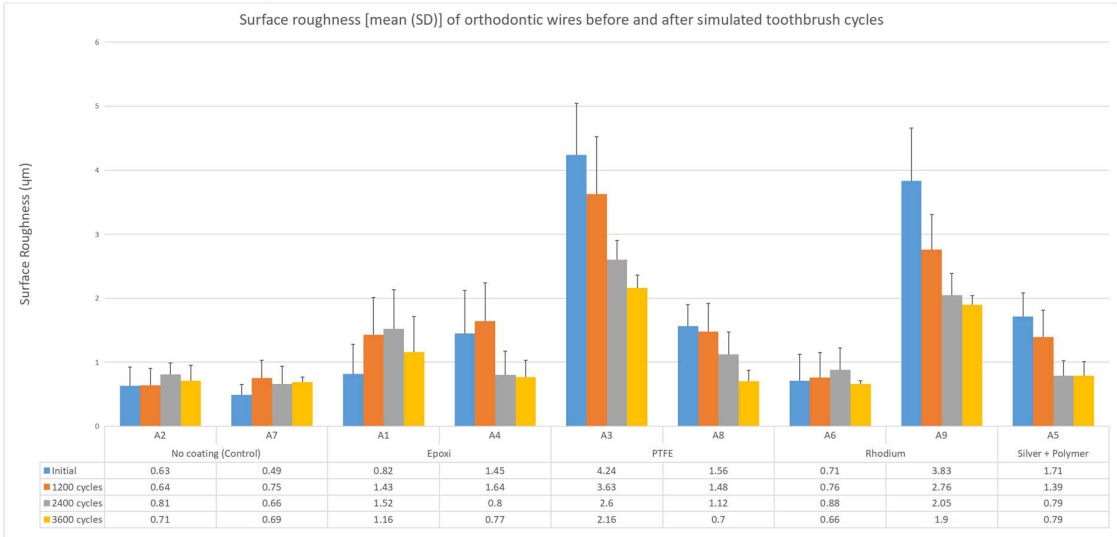


Figure 8 - Graphical representation of surface roughness in μm progression according to brushing cycles (initial, 1200, 2400, 3600 cycles) (Average and Standard Deviation) by arch wire group

Figure 9 demonstrates that control archwires increase their roughness 13% and 41%, respectively for A2 and A7. Also, the Epoxy group was the only group to present both behaviors increase in roughness of 41% (A1) and decrease of 47% (A4). And the Teflon, Rhodium and Silver+polymer groups all decreased their roughness after 360 brushing cycles. The greatest reduction was group A8-Teflon 55% and A5-Silver+polymer 54%.

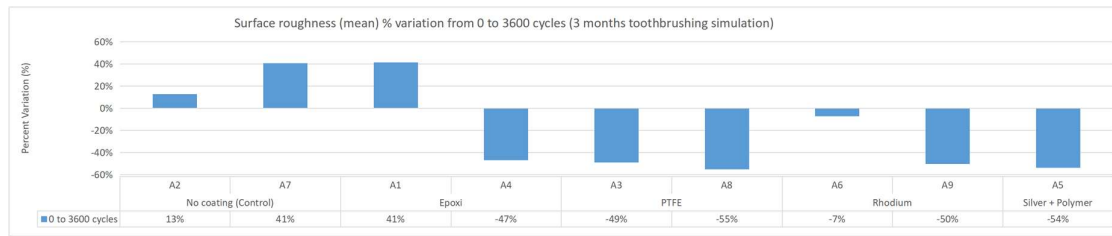


Figure 9 - Graphical representation of surface roughness % variation from initial to 3600 brushing cycles in μm (Average and Standard Deviation) by arch wire group

In summary, regarding superficial roughness results variation (before and after 3600 brushing cycles): The 2 control wires without coating were the ones presenting an increased in roughness of +13% (A2) and +41% (A7) after cycles.

PTFE presented the greatest decrease -49% (A3) and -55% (A8), followed by Silver + Polymer (A5) wire decreased -54%.

Rhodium decreased -7% (A6) and -50% (A9), and Epoxy Resin had both behaviors one wire increased 41% (A1) and the other decrease -47% (A4).

3.6 Discussion

3.6.1 Scanning Electron Microscopy Discussion

The surface topography of an orthodontic wire is an essential property known to influence its mechanical characteristics, esthetic appearance, corrosion behavior, and/or its biocompatibility (DAEMS et al., 2009). Clinically, a rough surface encourages greater plaque accumulation, influences its friction properties, and increases corrosion and color instability (BOURAUUEL et al., 1998; DOSHI et al., 2011; SAUNDERS et al., 1994; ELIADES & ATHANASIOU 2002).

Our SEM results reported that PTFE coated archwires presented the highest levels of topographic irregularity.

Choi et al., 2015 also used scanning electron microscopy to analyze the surface roughness of archwires. They have an uncoated control group, epoxy, Teflon

and Silver+Polymer groups. Both Epoxy and Teflon wire groups exhibited less surface roughness and the Silver+Polymer had the highest irregularity, which are results contrary to our finds where, Teflon had the highest topographic irregularities.

On the other hand, Da Silva et al., 2013 found great roughness to Epoxy resin coated archwires compared to those of conventional stainless steel (SS) and nickel titanium (NiTi) ones after 21 days of oral exposure. They used (ortho organizer, ortho technology, orthometric, Esthetic flexy super elastic) evaluated with a stereoscope and in a scanning electron microscope. Also, Epoxy coating wires showed a severe deterioration and a greater surface roughness than control counterparts (conventional SS and NiTi wires).

3.6.2 Superficial Roughness Discussion

Regarding the superficial roughness, our analyses found an increase in roughness in the control and Epoxy coated groups of 40% after brushing.

These results are confirmed in the literature by Rudge et al., 2015. The authors compared surface roughness of four fully coated tooth colored [Forestadent: Biocosmetic (FB) and Titanol Cosmetic (FT); TOC Tooth Tone (TT); and Hawley Russell Coated Superelastic NiTi (HRC)]; two partially coated tooth colored [DB Euroline Microcoated (DB) and TP Aesthetic NiTi (TP)]; two rhodium coated [TOC Sentalloy (TS) and Hawley Russell Rhodium Coated Superelastic NiTi (HRR)]; and two controls: stainless steel [Forestadent Steel (FS)] and NiTi archwire [Forestadent Titanol Superelastic (FN)]. Roughness coefficients were from low to high: FB; FN; TT; FS; TS; HRR; FT; DB; TP; HRC. Friction coefficients were from low to high: TP; FS; FN; HRR; FT; DB; FB; HRC; TS; TT.

In conclusion, coated archwires generally exhibited higher friction than uncoated controls. Also, aesthetic archwires investigated had either low surface roughness or low frictional resistance but not both properties simultaneously. Authors reported that causes for friction are likely to be multifactorial and do not appear to be solely determined by surface roughness.

Another study that agreed with our finds was Elayyan et al., 2008. Such examined the mechanical and physical properties of retrieved coated nickel-titanium (NiTi) archwires compared with uncoated samples. Ultraesthetic coated archwires (G&H Company) were investigated, which are Epoxy resin coated. All measured roughness parameters had greater surface roughness for the retrieved coated archwires. Surface roughness of coated archwires increased after use. Coated archwires have a low aesthetic value, with 25 per cent of the coating lost within 33 days in vivo.

However, Mousavi et al., 2017 compare the surface roughness of four coated esthetic wires with that of a conventional orthodontic wire. NiTi Memory wire (American Orthodontics, USA) as a control group; Orthocosmetic Elastinol (Ortho Organizers, USA); Perfect (Hubit, Korea); Imagination (Gestenco, Sweden); EverWhite (American Orthodontics, USA). They found that the average range of the 4 parameters was the highest for the uncoated Ni-Ti Memory wire (control group) while the coated wires showed the lowest values. Taking into account the study limitations, the surface roughness values of NiTi uncoated archwires were significantly higher than those of the coated wires.

Another found of our study was a roughness reduction in Teflon, Silver+Polymer and Rhodium coated groups about 50% after 3600 brushing cycles.

Our finds are supported by D' Anto et al., 2012, who agreed that Teflon presented less roughness, but reported high rugosity to Rhodium. These authors found that Titanium Memory Esthetic, Teflon coated wire, was the least rough among the NiTi archwires, being slightly rougher than Stainless Steel. On the other hand, the Sentalloy High Aesthetic, which is produced by ion implantation of rhodium, showed the highest values of roughness. The Rhodium and the Teflon coating are the most common archwire surface treatments according to Husmann et al., 2002 and Elayyan et al., 2010.

Finally, another study by Ryu and colleagues, 2015 found that Silver+Polymer was less rough and Epoxy coated wires presented higher rugosity. Ryu et al., 2015, evaluated three aesthetic NiTi archwires: silver-platinum- and polymer-coated NiTi

Natural Dany (Dany group), epoxy resin-coated Orthoforce Ultraesthetic™ (Ultra group), and Teflon®-coated Perfect (Perfect group).

The Dany group (2037.5 ± 527.3 nm) had the highest peak-to-peak surface roughness in the coated areas, followed by the Ultra group (811.1 ± 407.5 nm) and the Perfect group (362.7 ± 195.8 nm). The authors concluded that aesthetic archwire coated by silver or gold have a higher surface roughness when compared to arches coated only by Epoxy resin or Teflon or uncoated archwires.

Considering the absolute number of this study, among all material and coating types we could not identify great variations of behavior between the different groups. The observable difference was found on PTFE coating with the highest roughness in the topographic surface, which disagrees with Teflon properties presented in the market. Teflon is the commercial material for PTFE (polytetrafluoroethylene), which is a synthetic polymer that basically consists of carbon and fluoride. Due to the strength of the carbon-fluoride bonds, the PTFE has the third lowest coefficient of friction of any known solid, heat-resistant, and hydrophobic.

However, after 3600 brushing cycles, which corresponds to 3 months of brushing, Teflon presented one of lowest roughness, so this find confirms the PTFE properties described in the literature as its lowest coefficient of friction and unadherent.

3.7 Conclusion

In conclusion, based on this *in vitro* study limitation we can conclude that Teflon coated archwires, even though presented a very irregular surface in the beginning, it decreased significantly its roughness along the orthodontic treatment to a significant reduction in roughness found in the coated archwires tested in this experiment. We didn't identify a similar trend in other materials or esthetic coatings.

The most stable groups regarding surface roughness before and after brushing were both control groups and Rhodium, which clinically is a more desirable outcome to predict wire behavior during treatment.

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4 Considerações Finais da Tese

Essa tese utilizou os protocolos de estudos *in vitro* pelas normas ISO, as quais representam o padrão mais alto de qualidade e rigorosidade de tais experimentos, os quais proporcionam resultados altamente válidos e reproduzíveis com baixo custo e eficiência quando comparados com estudos *in vivo*. Estudos *in vitro* são altamente recomendados, em especial, para avaliação de novos materiais dentários os quais por serem produtos desenvolvidos em laboratório usados em humanos, podem ter suas propriedades mecânicas e óticas rigorosamente avaliadas em tais estudos sem ter a necessidade de espera pelo aumento de seu uso em pacientes nas práticas odontológicas. Tais estudos, são muito bem vindos na literatura, pois auxiliam no desenvolvimento e aprimoramento de tais materiais.

É importante salientar que todos os arcos utilizados nesse estudo foram doados pelos fabricantes. Não existiu nenhum conflito de interesse relacionado aos resultados dessa tese.

Considerando as limitações de estudos *in vitro*, mesmo que os sistemas usados sigam as normas ISO que representam o mais alto padrão de validade e confiabilidade em experimentos laboratoriais, tais estudos apresentam certas limitações em reproduzir na íntegra a realidade dos sistemas humanos nas bancadas de laboratórios.

Sendo assim, levando em consideração as vantagens e desvantagens desse estudo, os valores de rugosidade e topografia superficial dos arcos revestidos por Teflon apresentaram significativamente maior acúmulo de biofilme e irregularidade quando comparado com arcos ortodônticos metálicos convencionais. No entanto, arcos revestidos por PTFE apresentaram uma redução considerável nos índices de rugosidade durante tratamento ortodôntico e, principalmente, após escovação dentária, tal mecanismo que reproduz a realidade da clínica odontológica de pacientes em tratamento ortodôntico.

Em conclusão, nenhum dos arcos estéticos avaliados nesse estudo apresentou características estatisticamente diferentes dos arcos ortodônticos convencionais. No entanto, os arcos de Teflon apresentaram uma maior aderência de biofilme e rugosidade as quais foram reduzidas após a simulação de ciclos de escovação. Sendo assim, baseado nos resultados dessa tese concluímos que os resultados de ambos os experimentos indicam que não houve diferença significativa entre os diferentes arcos estéticos e arcos metálicos. Essas descobertas nos mostram que os fabricantes estão investindo constantemente em tecnologia e estão comprometidos no desempenho desses materiais para entregar um material de maior qualidade e mais eficiente ao mercado.

Finalmente, conclui-se que os arcos estéticos têm comportamento semelhante aos arcos de metal convencionais. Os ortodontistas podem usar arcos estéticos, levando em consideração um maior cuidado com higiene oral em pacientes com arcos revestidos por Teflon devido a uma maior rugosidade superficial e maior acúmulo de biofilme. .

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ZEGAN, G.; SODOR, A.; MUNTEANU, C. Surface characteristics of retrieved coated and nickel-titanium orthodontic archwires. **Romanian Journal of Morphology and Embryology**, v. 53, p. 935–939, 2012.

ZHANG, N., MA, Y., WEIR, M.D., XU, H.H., BAI, Y., MELO, M.A. Current insights into the modulation of oral bacterial degradation of dental polymeric restorative materials. **Materials**, v.10, n. 5, p.507.

6 Anexos

6. 1 Websites dos materiais e produtos

1. Nitanium® Super Elastic by Ortho Organizers, Inc.
<http://www.henryscheinortho.com/orthodontic-archwires-nitanium>
2. Tooth Tone® Coated Archwire by Ortho Technology
<https://www.orthotechnology.com/shop/wire-products/tooth-tone-coated-archwire-and-accessories/tooth-tone-coated-nickel-titanium-full-form-archwire-rectangle/>
3. Bio-active RC by Gc Orthodontics Europe GmbH
<https://www.gcorthodontics.eu/GC/fr/content/bio-active>
4. NT3™ SE by American Orthodontics <http://www.americanortho.com/wire-NiTi.html>
5. Niti Micro Dental White by Acme Monaco Co
<http://acmemonaco3.com/archwires/superelasticnickel.html>
6. NiTi Dental White S by Acme Monaco Co
<http://acmemonaco3.com/archwires/microdentalarch.html>
7. Copper Ni-Ti® byOrmco Corporation <https://ormco.com/products/copper-niti/>
8. FLI® Wire by RMO, Inc. <https://www.rmortho.com/products/fli-wire> ~~trashed/~~
9. Dany Coated Archwire by Dany BMT Co., Ltd
https://danybmt.en.ec21.com/DANY-Coated-Wire--9505883_9505899.html
10. GAC PAK by DENTSPLY INTERNATIONAL, Inc.
<https://www.dentsplysirona.com/en-us/products/orthodontics/archwires.html/Orthodontics/Archwires/Stainless-Steel/c/1000324.html>

6. 2 Quadro de materiais

GROUP CODE	PRODUCT	MANUFACTURER	SPECIFICATION	Coating type	QTY	Made in	CITY, COUNTRY
A1	Tooth Tone® Coated Archwire	Ortho Technology	16x22 NiTi	Epoxi	5	Brazil	Lutz, FL, USA
A2	NT3™ SE	American Orthodontics	16x22 NiTi	No coating (Control)	50	USA	Sheboygan, WI, USA
	NT3™ SE	American Orthodontics	16x22 NiTi		20	USA	Sheboygan, WI, USA
	NT3™ SE	American Orthodontics	16x22 NiTi		20	USA	Sheboygan, WI, USA
A3	FLJ® Wire	RMO, Inc.	16x22 NiTi	PTFE	5	USA	Denver, CO, USA
	FLJ® Wire	RMO, Inc.	16x22 NiTi		10	USA	Denver, CO, USA
A4	Nitanium® Super Elastic	Ortho Organizers, Inc.	16x22 NiTi	Epoxi	5	USA	Carlsbad, CA, USA
A5	Dany Coated Archwire	Dany BMT Co., Ltd.	16x22 NiTi	Silver + Polymer	4	Korea	Gyeonggi-do, Korea
A6	Bio-active RC	Gc Orthodontics Europe GmbH	16x22 NiTi RC	Rhodium	5	Japan	Breckerfield, Germany
A7	Copper Ni-Ti®	Ormco Corporation	16x22 NiTi Cu	No coating (Control)	20	USA	Glendora, CA, USA
A8	Niti Micro Dental White	Acme Monaco Co	16x22 NiTi	PTFE	10	USA	New Britain, CT, USA
A9	NiTi Dental White S	Acme Monaco Co	16x22 NiTi	Rhodium	10	USA	New Britain, CT, USA