UNIVERSIDADE FEDERAL DE PELOTAS

Programa de Pós-Graduação em Odontologia

TESE

Uso de benzodioxolas em sistemas de fotoiniciação de adesivos odonbtológicos

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USO DE BENZODIOXOLAS EM SISTEMAS DE FOTOINICIAÇÃO DE ADESIVOS ODONTOLÓGICOS

Tese apresentada ao Programa de Pós-Graduação em Odontologia, Área de concentração em Dentística da Faculdade de Odontologia da UNIVERSIDADE FEDERAL DE PELOTAS, como requisito parcial à obtenção do título de Doutor em Odontologia.

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DEDICATÓRIA

Dedico este trabalho:

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“Quando sonhamos sozinhos, é só um sonho;
quando sonhamos juntos, é o início de uma nova realidade"
Dom. Helder Câmara.

NOTAS PRELIMINARES

A presente dissertacao foi redigida Segundo o Manual de Normas para Dissertacoes, Teses e Trabalhos Cientificos da Universidade Federal de Pelotas de 2006, adotando o Nivel de Descricao 4 – estruturas em Artigos, que consta no Apêndice D do referido manual. Disponivel no endereco eletronico:
http://www.ufpel.tche.br/prg/sisbi/documentos/Manual_normas_UFPel_2006.pdf

RESUMO

LIMA, Giana da Silveira. **Uso de benzodioxolas em sistemas de fotoiniciação de adesivos odontológicos** 2009. **123 f.** Tese (Doutorado). Programa de Pós-graduação em Odontologia, Universidade Federal de Pelotas, Pelotas, RS/Brasil.

O objetivo deste estudo foi avaliar a efetividade de componentes derivados de benzodioxolas, como co-iniciadores da polimerização radicalar de um adesivo autocondicionante experimental. Para compor sistemas adesivos autocondicionantes experimentais foi desenvolvido um *primer*, composto por monômeros metacrilatos e solventes. O adesivo foi formulado utilizando uma resina adesiva modelo, composta por 50% de bisfenol A glicidil dimetacrilato (Bis-GMA), 25% de 2-hidroxietil metacrilato (HEMA) e 25% de trietilenoglicol dimetacrilato (TEGDMA), em massa. Canforoquinona (CQ) na concentração 1% molar foi utilizada como fotoiniciador da polimerização da resina modelo. Os grupos experimentais foram formulados com diferentes co-iniciadores na resina adesiva: 1,3-benzodioxola (BDO) e álcool piperonílico (AP), em diferentes concentrações molares (0.25, 0.50, 1, 2, 4, 8, 16 %). Adicionalmente um grupo com amina terciária, etil,4-dimetilamino benzoato (EDAB) como co-iniciador, foi formulado como controle. Características e propriedades físicas, químicas e mecânicas do polímero obtido pelos adesivos experimentais foram avaliadas utilizando as metodologias de cinética de polimerização, sorção e solubilidade, resistência à flexão e módulo de elasticidade. A resistência de união à microtração (MPa) ao esmalte e à dentina, com a caracterização do tipo de fratura foi investigada. Adicionalmente, análise morfológica da interface adesiva em dentina foi avaliada. Os resultados indicaram que o BDO e PA foram co-iniciadores efetivos para sistemas fotoiniciadores à base de canforoquinona (CQ). Comparações entre os adesivos experimentais com co-iniciadores derivados de benzodioxolas (A_{BDO} e A_{AP}) e amina (A_{EDAB}), mostraram performance similar na avaliação da cinética de polimerização, resistência à flexão, sorção e solubilidade da resina adesiva modelo avaliada. Na avaliação da resistência de união ao esmalte e à dentina, foi detectada diferença estatística e houve predominância de falhas mistas. A_{PA} apresentou maior resistência de união que A_{EDAB}, enquanto A_{BDO} mostrou resultados intermediários. A camada híbrida para todos os grupos apresentou uma espessura entre 1 e 2 µm. Não foi observada diferença na homogeneidade da interface adesiva em dentina. BDO e PA se revelaram alternativas viáveis à amina como co-iniciadores para a polimerização radicalar. Ademais, estas benzodioxolas são mais promissoras e vantajosas que as aminas, por sua biocompatibilidade e presença na dieta humana.

Palavras chave: Sistemas adesivos. Fotoiniciação. Fotoiniciadores. Co-iniciadores, Benzodioxolas. Aminas.

ABSTRACT

LIMA, Giana da Silveira. Use of benzodioxoles in photoinitiation systems of dental adhesives **2009. 123 p. Thesis (Doctorate) – Post Graduate Program, School of Dentistry, Federal University of Pelotas / RS, Brazil.**

The objective of this study was to evaluate the effectiveness of benzodioxole derivatives as co-initiators of radical polymerization of experimental self-etching adhesive systems. To compose the experimental self-etching adhesive systems a primer, containing methacrylate monomers and solvents, was developed. A monomer mixture, based on 50 wt % of Bisphenol A glicidyl dimethacrylate (Bis-GMA), 25 wt% of triethyleneglycol dimethacrylate (TEGDMA) and 25 wt% of 2-hydroxyethyl methacrylate (HEMA), was used as a model dental adhesive resin. Camphorquinone (CQ) 1 mol % was used as a photoinitiator to initiate polymerization. Different co-initiators (1,3-benzodioxole and piperonyl alcohol) and concentrations (0.25, 0.50, 1, 2, 4, 8, 16 mol %) were used in a model dental adhesive resin, to compose the experimental groups. Additionally, tertiary amine (EDAB) was used as co-initiator in the control group. The physical, chemical and mechanical properties and characteristics of the polymer obtained for the experimental adhesives (A_{BDO}, A_{PA}, A_{EDAB}) were evaluated using polymerization kinetics, sorption and solubility, flexural strength and elastic modulus. The microtensile bond strength (μTBS) to enamel and dentin, and fracture mode were investigated. Adicionalmente morphological analysis of the dentin bonding Interface were evaluated. The results indicated that the BDO and PA were effective co-initiators for the photoinitiator system based on CQ. Comparisons between the benzodioxole derivative co-initiators and traditionally used amine EDAB, showed similar performance in the kinetics of polymerization, flexural strength, water sorption and solubility of the model dental adhesive resin evaluated. In the microtensile bond strength dentin means were higher than enamel and mixed failures were predominant. A_{PA} showed higher bond strengths than A_{EDAB}, while A_{BDO} showed intermediate data. The hybrid layer for all groups was shown to be shallow (1-2 μm thick). No appreciable differences in homogeneity were detected along the bonded interface. BDO and PA were feasible alternatives to conventional amine as co-initiator of radical polymerization, moreover, as these benzodioxoles are found in the human diet, this characteristic made them more promising and advantageous to use in dental adhesive resin formulations than amine.

Keywords: Adhesive systems. Photoinitiation System. Photoinitiators. Co-initiators. Benzodioxoles. Amines.

Lista de Abreviaturas

A	resina adesiva ou adesivo
A_{BDO}	resina adesiva + co-iniciador 1,3-benzodioxola
A_{EDAB}	resina adesiva + co-iniciador etil, 4-dimetilamino benzoato
AF	Falha adesiva
ANOVA	Análise de Variância
AP ou PA	álcool piperonílico
A_{PA}	resina adesiva + co-iniciador álcool piperonílico
ATR	refletância total atenuada
B	largura em milímetros
BDO	Benzodioxola
Bis-GMA	bisfenol A glicidil dimetacrilato
Bis-MEP	bis-metacriloiloxietil hidrogênio fosfato
CA	Falha coesiva em adesivo
CDC-Bio	Centro de Desenvolvimento e Controle de Biomateriais
CEP	Comitê de Ética e Pesquisa
CQ	Canforoquinona
CS	Falha coesiva no substrato
D	deflexão em milímetros
E	módulo de elasticidade
EDAB	etil 4-dimetilamino benzoato
F	força máxima em Newtons
FO	Faculdade de Odontologia
GPa	Gigapascal
H	altura em mm
HEMA	2-hidroxietil metacrilato
ISO	Organização Internacional de Padronização
L	distância em milímetros
M	Falha mista

m1	massa um ou primeira massa
m2	massa dois ou segunda massa
m3	massa três ou terceira massa
MEP	metacriloiloxietil dihidrogênio fosfato
mol %	percentual molar
MPa	Megapascal
N	Newton
NDK	número de dureza Knoop
ºC	graus Celsius
pH	potencial hidrogeniônico
R	Radical
Rp	taxa de polimerização
$Rp^{\text{máx}}$	máxima taxa de polimerização
RT-FTIR	Infravermelho Transformada de Fourier – Tempo Real
SD	Desvio padrão
SEM	Microscópio eletrônico de varredura
SL	Solubilidade
TEGDMA	trietilenoglicol dimetacrilato
UFPel	Universidade Federal de Pelotas
UFRGS	Universidade Federal do Rio Grande do Sul
V	Volume
WS ou SA	sorção em água
wt %	percentual em massa
X	Vezes
μTBS	resistência de união à microtração
σ	resistência à flexão

SUMÁRIO

1 PROJETO DE TESE[§]

[§] Projeto redigido segundo o Manual de Normas da Universidade de Federal de Pelotas disponível em: <http://www.ufpel.tche.br/prg/sisbi>. Acesso em: 20 junho. 2009.

1.1 INTRODUÇÃO

Os sistemas de fotoiniciação utilizados para a polimerização radicalar de materiais odontológicos fotopolimerizáveis usualmente são constituídos por dois componentes: o fotoiniciador, que pode absorver luz diretamente, e o co-iniciador, que não absorve luz, mas atua como agente redutor. O co-iniciador doa prótons para a estabilização do radical gerado na molécula excitada do fotoiniciador, gerando espécies reativas com tempo de vida suficiente para iniciar a polimerização. (ANDRZEJEWSKA, 2001)

Canforoquinona (CQ) e amina constituem o sistema de fotoiniciação mais amplamente utilizado na polimerização radicalar de materiais dentários à base de metacrilatos. (JAKUBIAK et al., 2003, MUSANJE; FERRACANE; SAKAGUCHI, 2009, SCHROEDER; COOK; VALLO, 2008, SCHROEDER; VALLO, 2007) A polimerização fotoiniciada ocorre pela reação em cadeia entre os radicais livres, formados pelo sistema fotoiniciador, e os monômeros. (JAKUBIAK, et al., 2003) A CQ é um típico fotoiniciador ativado pela luz visível com absorbância entre 450-500 nm (λ_{max}=468 nm) (OGUNYINKA et al., 2007) e requer um agente redutor para que ocorra uma polimerização eficiente. (ANDRZEJEWSKA, 2001, CHEN; FERRACANE; PRAHL, 2007) Apesar deste sistema ser comumente utilizado, a CQ apresenta uma coloração fortemente amarelada e a amina por sua vez, pode apresentar um amarelamento com o passar do tempo, o que pode representar um problema em restaurações estéticas. (NEUMANN et al., 2006, OGUNYINKA, et al., 2007, SHIN; RAWLS, 2009)

As aminas, co-iniciadoras da polimerização ativada por luz, são doadoras de hidrogênio na polimerização radicalar. (ANDRZEJEWSKA, 2001) Sua influência positiva na reação de polimerização, no grau de conversão e nas propriedades do polímero formado está bem descrita. (MUSANJE; FERRACANE; SAKAGUCHI, 2009, OGLIARI et al., 2007, SHIN; RAWLS, 2009, YOSHIDA; GREENER, 1994, YOSHIDA; GREENER, 1993) No entanto, apesar do bom desempenho como co-iniciadoras da polimerização, as aminas apresentam potencial citotóxico e mutagênico. (ALBRECHT; STEPHENSON, 1988, JOSEPHY et al., 1998, LISO et al., 1997, PELKA et al., 2000, WEISBURGER et al., 1978)

Alternativas para reduzir a citotoxicidade de resinas odontológicas vêm sendo testadas em resinas modelo, como a substituição de amina por outro co-iniciador menos tóxico. (LIU et al., 2007, SCHNEIDER et al., 2008, SHI; NIE, 2007, SHI; NIE, 2007) Neste aspecto, vários anti-oxidantes presentes nos alimentos são de grande interesse, pois se ligam facilmente a radicais livres e não são tóxicos aos tecidos do organismo humano. (AMES, 1983, JOSHI et al., 2005, KUMAGAI et al., 1991, MICALE; ZAPPALA; GRASSO, 2003) Além disso, estudos têm demonstrado que derivados das benzodioxolas tem potencial anticarcinogênico, (MICALE; ZAPPALA; GRASSO, 2002, TSENG; TSHENG; LEE, 2001) antifúngico, antibacteriano (TAGASHIRA et al., 1997) e radioprotetor (MATOS et al., 2004), entre outros.

Benzodioxolas são substâncias que podem ser extraídas de produtos naturais como melissa (Figura 1), (TAGASHIRA & OHTAKE, 1998) manjericão (Figura 2), canela (Figura 3), noz moscada (Figura 4) entre outros, pela obtenção de óleos essenciais. (HICKEY, 1948) Ademais, derivados das benzodioxolas são amplamente encontradas na flora brasileira. (MOREIRA et al., 2007) Estas substâncias são antioxidantes que apresentam alto sinergismo com moléculas de canforoquinona. (LIU, et al., 2007) Alguns estudos demonstraram que a substituição de amina terciária por benzodioxolas (SHI; NIE, 2007, LIU, et al., 2007), caracterizados na tabela 1, em um sistema de fotoiniciação binário (canforoquinona + benzodioxola) de uma blenda modelo não afeta as propriedades mecânicas do polímero formado, quando comparado ao sistema convencional CQ / amina.

Figura 1. Melissa (*Melissa officinalis*)

Figura 2. Manjericão (*ocimum basilicum*)

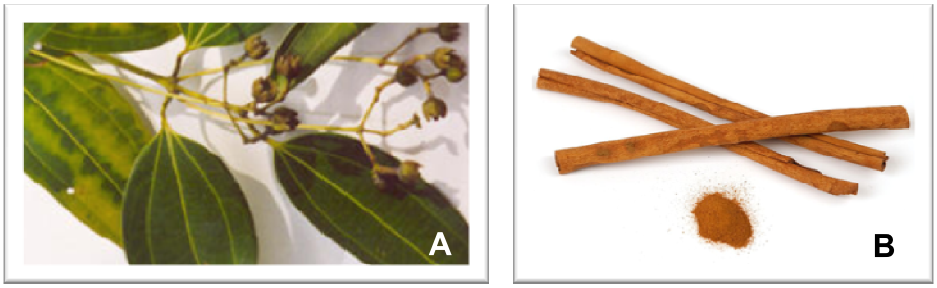
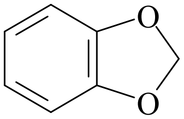
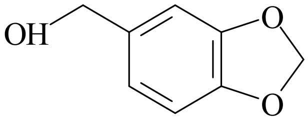


Figura 3. Canela (Cinnamomum verum), (A) folhas e flores e (B) cascas do tronco.[**]

Figura 4. Noz moscada (Myristica fragrans), (A) folhas e flores e (B) noz.[**]

[**] Fonte: Site do Instituto Brasileiro de Plantas Medicinais (IBPM) < http://www.ibpm.org.br > acesso em 26 abril, 2009.

Tabela 1. Benzodioxolas avaliadas neste estudo.

Substância	**1,3-benzodioxola**	**Álcool piperonílico**
Fórmula molecular	$C_7H_6O_2$	$C_8H_8O_3$
Peso molecular	122,121	152,147
Fórmula estrutural		
Sinônimos	1,2- (dioximetileno) benzeno	5-metanol - 1,3-benzodioxola Álcool benzil - 3,4-(dioximetileno) Fenilmetanol - 3,4-(dioximetileno)
Coloração	Incolor	Levemente amarelado

Dentre os materiais à base de metacrilatos utilizados em odontologia, os sistemas adesivos são resinas de composição específica e de baixa viscosidade, que constituem íntimo contato com o substrato dental. (DE MUNCK et al., 2005) Assim, estes materiais têm a capacidade de penetrar a estrutura do substrato desmineralizado bem como os túbulos dentinários, enfrentando desafios para a reação de polimerização, como a presença umidade, (GUO et al., 2007, HIGASHI et al., 2009) e desafios de biocompatibilidade. (COX et al., 1998, GOLDBERG, 2008, GUO et al., 2008) O emprego destes co-iniciadores em sistemas adesivos mostra-se um alternativa interessante, considerando a fundamental importância de sua biocompatibilidade.

Desta forma, substâncias derivadas das benzodioxolas apresentam-se como potenciais substitutas às aminas, na busca de sistemas de fotoiniciação biocompatíveis para a composição de sistemas adesivos odontológicos, apresentando vantagens na aplicação, manuseio e manufatura destes adesivos.

1.2 JUSTIFICATIVA

Os sistemas adesivos são materiais amplamente utilizados na odontologia restauradora atual, entre eles os sistemas adesivos autocondicionantes vêm ganhando destaque no mercado mundial, tornando-se uma realidade na clínica odontologia atual. Entretanto estes materiais ainda possuem limitações, entre elas o potencial citotóxico e genotóxico de determinados componentes de sua fórmula. A utilização de derivados das benzodioxolas, como doadoras de prótons, para sistemas de fotoiniciação de monômeros metacrilatos, surge como uma alternativa à amina terciária, que é uma substância tradicionalmente utilizada com esta função, apresentando a vantagem de serem substâncias derivadas de produtos naturais e constituintes de alimentos da dieta humana. Além disso, o apelo atual pelo emprego de substâncias naturais, e desenvolvimento e processos de manufatura com mínima geração de resíduos e riscos à saúde, é contemplado com o emprego destes produtos.

1.3 OBJETIVOS

Desenvolver um sistema adesivo autocondicionante experimental com sistema de fotoiniciação composto por co-iniciadores derivados das benzodioxolas, como substitutos às aminas terciárias.

Avaliar o desempenho destes adesivos compostos por co-iniciadores *alternativos* por meio da investigação da cinética de polimerização, resistência à flexão, módulo de elasticidade, microdureza e resistência de união em esmalte e dentina e compará-los ao adesivo que utiliza amina terciária como co-iniciador.

A hipótese a ser testada é que o tipo de co-iniciador na composição do sistema adesivo experimental desenvolvido, não influencia suas características e propriedades físico-químicas, mecânicas e adesivas testadas.

1.4 MATERIAIS E MÉTODOS

1.4.1 Considerações iniciais

Este projeto foi encaminhado ao Comitê de Ética e Pesquisa da Faculdade de Odontologia da Universidade Federal de Pelotas (CEP– FO/UFPel) e aprovado sob parecer 090/2009 (Anexo 1). O projeto caracteriza-se por um estudo interdisciplinar (Odontologia - Química) e interinstitucional (UFPel - UFRGS) sendo os experimentos executados em ambas instituições.

1.4.2 Desenvolvimento do sistema adesivo autocondicionante experimental

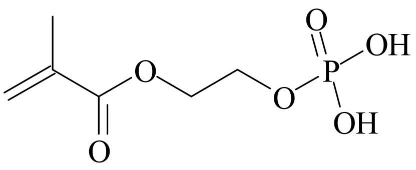
1.4.2.1 *Primer*

O *primer* autocondicionante experimental será formulado utilizando os reagentes: 2-hidroxiletil metacrilato (HEMA), Aldrich, Steinheim, Alemanha; metacriloiloxietil dihidrogênio fosfato (MEP) e bis-metacriloiloxietil hidrogênio fosfato (Bis-MEP), sintetizados no Centro de Desenvolvimento e Controle de Biomateriais (CDC-Bio/UFPel), representados na figura 5. Como solventes serão utilizados: etanol absoluto (Synth, Diadema, SP/Brasil) e água destilada. Estes reagentes (Tabela 2) serão misturados mecanicamente por 1 minuto e em seguida levados à cuba ultrassônica (Plana CT CBU 100 / 1LDG, Tatuapé, SP, Brasil), por 10 minutos para homogeneização da mistura. O pH do *primer* será mensurado em um pHMetro digital (An 2000 microprocessado, Analion, Riberão Preto, SP, Brasil).

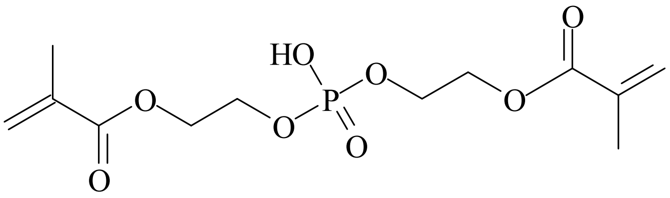
Tabela 2. Composição do *primer* utilizado nos sistemas adesivos autocondicionantes experimentais avaliados.

Composição	% massa
HEMA	30
MEP / Bis-MEP	30
Etanol	20
Água	20
Total (%)	100

Abreviaturas: HEMA: 2-hidroxiletil metacrilato; MEP: metacriloiloxietil dihidrogênio fosfato; Bis-MEP: bis-metacriloiloxietil hidrogênio fosfato.



Metacriloiloxietil dihidrogênio fosfato (MEP)



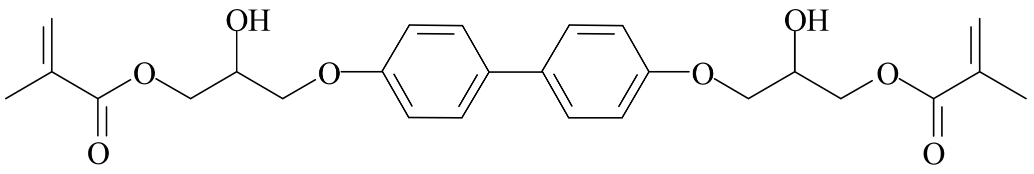
Bismetacriloiloxietil hidrogênio fosfato (Bis-MEP)

Figura 5. Estrutura molecular do monômero fosforado MEP (metacriloiloxietil dihidrogênio fosfato) e Bis-MEP (bis-metacriloiloxietil hidrogênio fosfato) sintetizados no CDC-Bio / UFPel.

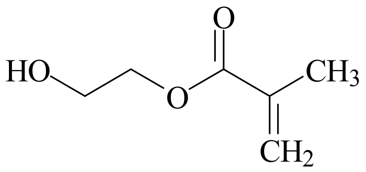
1.4.2.2 Adesivo

Resina adesiva modelo

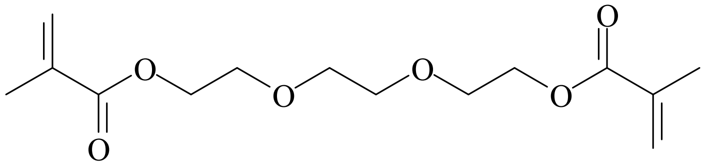
Os adesivos experimentais serão formulados a partir de uma resina adesiva modelo composta por (concentração em massa): 50% de Bisfenol A glicidil dimetacrilato (Bis-GMA), 25% de 2-hidroxiletil metacrilato (HEMA) e 25% de Trietilenoglicol dimetacrilato (TEGDMA), cujas estruturas moleculares estão mostradas na figura 6. Os monômeros componentes da resina adesiva modelo serão misturados mecanicamente por 5 minutos e homogeneizados em cuba ultrassônica (Plana CT), por 10 minutos.



Bisfenol A glicidil dimetacrilato (BisGMA)



2-hidroxietil metacrilato (HEMA)



Trietilenoglicol dimetacrilato (TEGDMA)

Figura 6. Estrutura molecular dos monômeros componentes da resina adesiva modelo

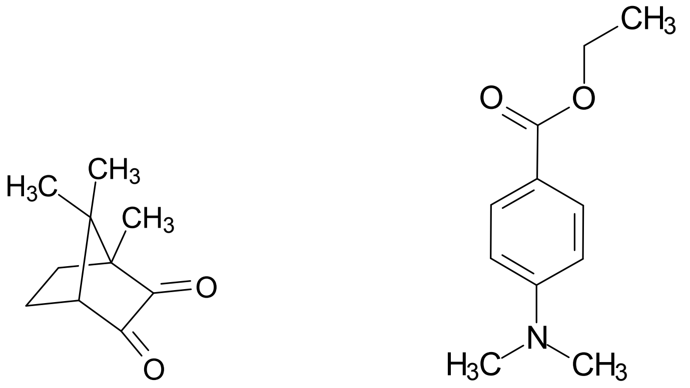
Avaliação de concentração das benzodioxolas (co-iniciadores)

Para formulação dos adesivos experimentais, será realizada uma avaliação da influência da concentração das benzodioxolas no grau de conversão, resistência à flexão e módulo de elasticidade. De acordo com o grupo experimental serão adicionados fotoiniciador (CQ) e co-iniciadores para compor o sistema de fotoiniciação. Para determinação da concentração de trabalho, serão adicionadas diferentes concentrações de co- iniciadores à resina adesiva modelo, adicionada de CQ a 1 % molar, considerando a molaridade da resina. As concentrações (% molar) avaliadas, para cada co-iniciador testado, estão dispostas na tabela 3. Um grupo composto por um sistema de fotoiniciação amplamente empregado nos produtos comercialmente disponíveis (CQ / amina), disposto na figura 7, constituirá o grupo controle. Este terá como co-iniciador a amina terciária EDAB [Etil 4-(dimetilamino) benzoato] que será empregada em uma única concentração (1 % molar), de acordo com dados da literatura. (OGLIARI et al., 2008, OGLIARI, et al., 2007)

Tabela 3. Diferentes concentrações de benzodioxolas avaliadas.

Concentração	**Grupos experimentais**						
[% molar]	**0,250**	**0,500**	**1,00**	**2,00**	**4,00**	**8,00**	**16,0**
1,3 Benzodioxola	BDO$_{0,25}$	BDO$_{0,50}$	BDO$_{1,0}$	BDO$_{2,0}$	BDO$_{4,0}$	BDO$_{8,0}$	BDO$_{16,0}$
Álcool Piperonílico	AP$_{0,25}$	AP$_{0,50}$	AP$_{1,0}$	AP$_{2,0}$	AP$_{4,0}$	AP$_{8,0}$	AP$_{16,0}$

Abreviaturas: BDO: 1,3 Benzodioxola ; AP: Álcool Piperonílico.



Canforoquinona (CQ) Etil 4-(dimetilamino) benzoato (EDAB)

Figura 7. Estrutura molecular do fotoiniciador, canforoquinona e do co-iniciador utilizado como controle, etil 4-(dimetilamino) benzoato.

Após determinação da influência das concentrações de cada co-iniciador alternativo na cinética de conversão, taxa de polimerização, resistência à flexão e módulo de elasticidade, será selecionada uma concentração de trabalho para cada benzodioxola. As concentrações eleitas constituirão os adesivos experimentais junto ao grupo controle com amina terciária, de acordo com a tabela 4, e estes terão suas características físico-químicas, mecânicas e adesivas avaliadas.

Tabela 4. Composição dos sistemas de fotoiniciação das resinas adesivas, componentes dos sistemas adesivos autocondicionantes experimentais avaliados.

	Resinas Adesivas (A)		
Composição (% molar / % massa)	**A_{BDO}**	**A_{AP}**	**A_{EDAB}**
Canforoquinona	1 / 0,6	1 / 0,6	1 / 0,6
1,3 Benzodioxola (BDO)	**?**	-	-
Álcool Piperonílico (AP)	-	**?**	-
Etil 4-(dimetilamino) benzoato (EDAB)	-	-	1 / 0,8

Abreviaturas: Bis-GMA: Bisfenol A glicidil dimetacrilato. TEGDMA: Trietilenoglicol dimetacrilato HEMA: 2-hidroxiletil metacrilato; MEP: metacriloiloxietil dihidrogênio fosfato; Bis-MEP: bis-metacriloiloxietil hidrogênio fosfato; EDAB Etil 4-(dimetilamino) benzoato. ? : Dados que serão obtidos após avaliação prévia de diferentes concentrações.

1. 4.3 Caracterização físico-química

1.4.3.1 Avaliação da cinética de polimerização dos adesivos experimentais

A cinética de conversão dos grupos experimentais será avaliada em tempo real por meio de um espectrofotômetro Infravermelho por Transformada de Fourier (RT-FTIR, Shimadzu Prestige21 Spectrometer, Shimadzu, Japão) equipado com dispositivo de reflectância total atenuada (ATR), (Figuras 8A e 8B), composto por um cristal horizontal de seleneto de zinco (ZnSe), com espelhos de angulação de 45º (PIKE Technologies, EUA). Um suporte será acoplado para a fixação da unidade fotoativadora ao espectrofotômetro, permitindo a padronização de uma distância de 5 mm entre a extremidade da ponteira de fibra ótica e a amostra (Figura 8C). As amostras serão dispensadas diretamente no cristal de ZnSe, com auxílio de uma micropipeta (~ 3 µl), e fotoativadas por 60 segundos. Cada adesivo experimental será analisado em triplicata. Para o monitoramento de varredura será utilizado o software IRSolution (Figura 8D), utilizando a apodização de Happ-Genzel, em uma faixa espectral entre 1750 e 1550 cm^{-1}, resolução de 8 cm^{-1} e velocidade de deslocamento de espelho de 2,8 mm/s. A análise será realizada em ambiente com temperatura controlada de 23º C e umidade relativa de < 60 %. O grau de conversão, por segundo, será calculado considerando a intensidade da vibração do tipo estiramento da dupla ligação carbono-carbono na freqüência de 1635 cm^{-1}. O estiramento simétrico do anel aromático em 1610 cm^{-1} das amostras polimerizadas e não polimerizadas será utilizado como padrão interno. Posteriormente, os dados obtidos serão plotados em uma curva ajustada pelo parâmetro regressivo não-linear de Hill 3 e será calculada a taxa de polimerização.

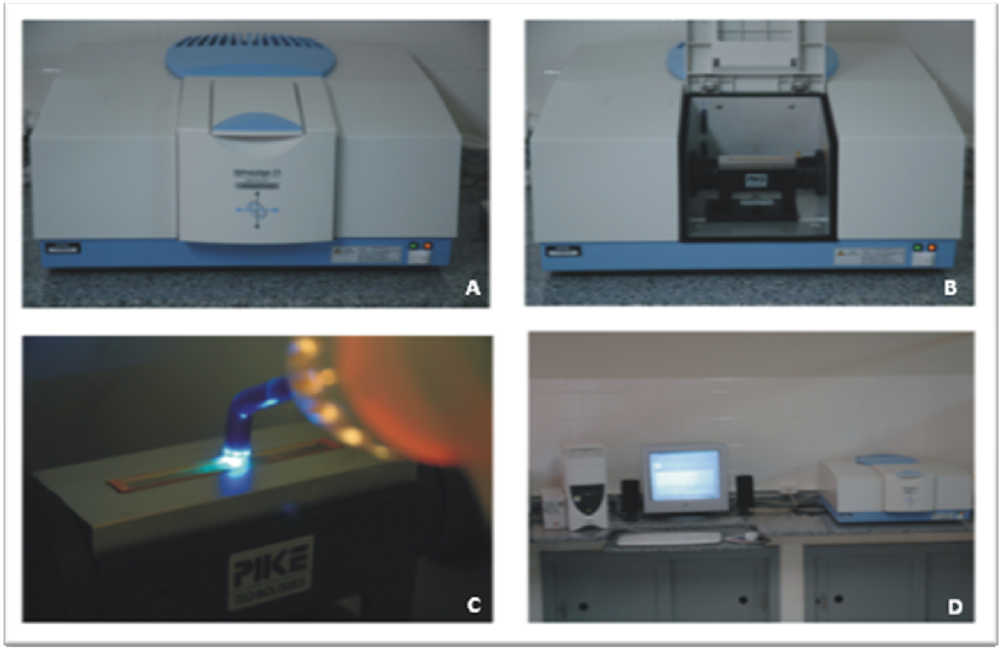


Figura 8. Espectrofotômetro por Transformada de Fourier Shimadzu, Prestige 2.1 (A) com dispositivo de reflectância atenuada ATR (B). Unidade fotopolimerizadora de lâmpada halógena acoplada ao cristal de seleneto de zinco (C), sistema conectado ao software IR Solution para obtenção dos espectros em tempo real (D).

1.4.3.2 Avaliação do potencial hidrogeniônico (pH) do primer autocondicionante experimental.

O potencial hidrogeniônico do *primer* será aferido com o auxílio de um pHMetro (An 2000 microprocessado, Analion, Ribeirão Preto, SP/Brasil) utilizando um eletrodo combinado de pH (Analion, V 631), mostrados na figura 9, após calibração do equipamento com as soluções padrão. O pH do *primer* será aferido num volume de 5 ml, com o eletrodo mergulhado na solução após 1 minuto de estabilização, então o valor de pH será anotado. O procedimento de aferição será realizado em triplicata e o valor médio adotado com valor do pH da solução.

Figura 9. pHMetro An 2000 microprocessado, Analion

1.4.3.3 Sorção e Solubilidade em água

Confecção dos espécimes

Dez espécimes por grupo serão confeccionados dispensando-se o material em um molde circular metálico com 1 mm de espessura e 15 mm de diâmetro, recoberto por tiras de poliéster nas duas extremidades. Em seguida, cada espécime será fotopolimerizado em ambos os lados em 9 posições diferentes, durante 20 s com uma fonte de luz halógena (XL 3000, 3M ESPE, St. Paul, MN, EUA). Em seguida, todos os espécimes terão seus bordos polidos com lixas de granulação 600 seguida de 1200. Todas as etapas do desenvolvimento desta metodologia seguirão as especificações ISO 4049/2000.

Ensaio de Sorção e Solubilidade em água

Os espécimes serão colocados em um dessecador contendo sílica e armazenados em estufa a 37° C. A massa de dois corpos de prova de cada grupo avaliado será mensurada repetidamente até sua estabilização. Após isso, a massa de todos os demais espécimes também será mensurada. Essa massa será denominada de m1, pois representa a massa seca do espécime antes dos fenômenos de Sorção e Solubilidade ocorrerem.

Após a definição da m1, todos os espécimes de um mesmo grupo serão colocados dentro de um recipiente contendo água destilada, na concentração de 10 ml para cada corpo de prova presente. Os recipientes serão armazenados dentro de uma estufa a 37° C, e após uma semana a massa de todos os espécimes será mensurada novamente e desta vez denominada de m2, representando a massa úmida do espécime após este ter sorvido água do meio em que foi armazenado.

Em seguida, os corpos de prova serão armazenados novamente, no dessecador com sílica, dentro de uma estufa a 37° C, para obter uma terceira massa (m3), que representa a massa final do espécime após o fenômeno de solubilidade, lixiviação de componentes do mesmo. Após a estabilização desta massa, a m3 de cada corpo de prova será mensurada.

A Sorção de água (SA) e a Solubilidade (SL) serão calculadas a partir das seguintes fórmulas:

$$\mathbf{SA} = \frac{\mathbf{m2 - m3}}{\mathbf{V}}, \qquad \mathbf{SL} = \frac{\mathbf{m1 - m3}}{\mathbf{V}},$$

onde: V representa o Volume dos espécimes, que será obtido da seguinte maneira:

$$\mathbf{V = \pi R^2 h,}$$

sendo que: *π* representa o valor constante de 3,14; *R* o raio do espécime; e *h* a altura do mesmo. O raio será obtido através da média entre dois diâmetros perpendiculares de cada espécime; e a altura será obtida pela média entre cinco alturas diferentes para cada um dos espécimes, mensurados com um paquímetro digital (Mitutoyo, Suzano, SP, Brasil).

1.4.4 Caracterização Mecânica

1.4.4.1 Resistência à mini-flexão e módulo de elasticidade

Confecção dos espécimes

Serão confeccionados corpos de prova (10 / grupo) com auxílio de uma matriz metálica bipartida (dimensão interna 10 x 2 x 2 mm) posicionada sobre uma tira de poliéster e encaixada em uma base metálica. A matriz metálica será lubrificada com solução de graxa de silicone 0,5% e os adesivos, de acordo com o grupo experimental, dispensados no interior da matriz e recobertos na superfície superior com outra tira de poliéster. Para fotoativação dos corpos de prova será utilizado o aparelho fotopolimerizador de luz halógena XL 3000, em duas janelas de exposição para cada lado do espécime. Os palitos obtidos terão os excessos removidos e as laterais serão polidas com auxílio de lixas de carbeto de silício, granulação 600 e 1200 e serão armazenados em água destilada à 37° C por 24 horas.

Ensaio de mini-flexão

Os palitos terão sua largura e espessura mensuradas, utilizando um paquímetro digital (Mitutoyo) com precisão de 0,01 mm, para o cálculo da área de cada espécime. O corpos de prova serão testados em uma máquina de ensaios mecânicos (Emic, DL 500, São José dos Pinhais, PR, Brasil) com velocidade de 1 mm / min. até sua falha. A resistência à flexão (σ) será calculada em megapascal (MPa) de acordo com a equação:

$$\boldsymbol{\sigma = 3LF / 2BH2}$$

onde F é a força máxima, em Newtons; L é a distância, em milímetros, entre os suportes; B é a largura, em milímetros e H é a altura (mm), da amostra, medida imediatamente antes do teste.

O módulo de elasticidade será calculado pela relação entre os valores da tensão e da deformação linear específica, na fase elástica, cujo valor é proporcional às forças de atração entre os átomos. A expressão matemática usada para o cálculo desta constante é:

$$\mathbf{E = FL^3 / 4BH^3d}$$

Sendo: F é a força máxima, em Newtons; L é a distância, em milímetros, entre os suportes; B é a largura, em milímetros e H é a altura (mm) da amostra e d é a deflexão (mm) correspondente a carga F.

1.4.4.2 Avaliação da microdureza

Confecção dos espécimes

Os corpos-de-prova (10 / grupo) serão confeccionados com o auxílio de uma matriz de silicona, circular de 1 mm de altura x 5 mm de diâmetro recoberta de ambos os lados com uma tira de poliéster. O adesivo será dispensado na matriz e fotoativado por 20 segundos de cada lado. Para leitura de microdureza, os corpos de prova serão incluídos em resina epóxi (Fiberglass, Porto Alegre, RS, Brasil), dispensada em tubo de PVC. Após a fixação dos discos, o conjunto será levado até uma politriz metalográfica (Aropol - E, Arotec S.A. Indústria e Comércio, Cotia, SP, Brasil) para acabamento e polimento. Lixas de granulação decrescente (180, 320, 400, 600 e 1200) serão utilizadas até a obtenção de polimento satisfatório para a leitura de microdureza.

Ensaio de microdureza

O ensaio de dureza Knoop será realizado em um aparelho universal para ensaios de microdureza, Microdurômetro (Futuretech FM 700, Tóquio, Japão), com procedimento automático de aplicação de 10gf de carga durante 10 segundos. As mensurações de microdureza serão realizadas mediante aumento de 500x. Após a calibração manual, o aparelho converterá automaticamente a distância entre as extremidades da diagonal maior do losango (formato resultante da edentação no adesivo, que será mensurado manualmente pelo operador), em número de dureza Knoop (NDK), que é a força em gf dividida pela área em mm^2. Para cada corpo-de-prova serão feitas cinco medições de microdureza e calculada uma média de microdureza Knoop.

1.4.5. Caracterização Adesiva

1.4.5.1 Resistência de união à microtração (µTBS) em dentina e esmalte

Confecção dos espécimes

a. Preparo dos dentes

Trinta e seis molares humanos, com período de extração inferior a três meses serão utilizados para esta avaliação. Após a extração, os dentes terão a porção radicular, o periodonto e a polpa, removidos, em seguida serão armazenados em solução de cloramina T 0,5%, durante sete dias. Posteriormente os dentes serão transferidos para água destilada, armazenados em um freezer e mantidos congelados até seu uso. Os dentes serão distribuídos aleatoriamente entre os seis grupos, correspondentes aos: três sistemas adesivos (A_{BDO}, A_{AP}, A_{EDAB}) aplicados em dois substratos (esmalte e dentina). Para os 18 dentes em que será utilizado o esmalte como substrato para adesão, as coroas serão seccionadas ao meio, no sentido mésio-distal e os fragmentos correspondentes ao esmalte vestibular, utilizados. A superfície vestibular será polida com lixas de carbeto de silício, granulação 600, durante 1 min., sob velocidade de 100 rpm em politriz metalográfica (Aropol- E), para obter uma superfície plana e padronizar a superfície do esmalte. A superfície do esmalte será lavada com água destilada e inspecionada em estereomicroscópio ótico com aumento 150 x para assegurar a ausência de dentina na superfície.

Para os dentes em que será restaurada a dentina, será removida a porção oclusal (0,5 mm abaixo do sulco central) e em seguida a dentina exposta será polida com lixas de carbeto de silício, granulação 600, durante 1 min., sob velocidade de 100 rpm em politriz metalográfica (Aropol – E), para padronização da lama dentinária. Depois de cortados e polidos, os dentes serão lavados abundantemente com água destilada e a superfície de dentina será inspecionada em estereomicroscópio ótico com aumento 150 x para assegurar a ausência de esmalte remanescente na superfície.

b. Confecção das restaurações

O *primer* autocondicionante experimental será aplicado vigorosamente sobre o substrato seco durante 30 segundos, em seguida será aplicado jato de ar a uma distância padronizada de 10 cm durante 10 segundos. De acordo com o grupo, o adesivo será aplicado e fotoativado por 20 segundos com aparelho fotopolimerizador de luz halógena XL 3000, com irradiância de 600 mW/cm^2, aferida em radiômetro (Demetron Research Corporation, P/N 10503, Danbury, EUA). A restauração será confeccionada utilizando quatro incrementos, de 1 mm de compósito restaurador (Charisma, Heraeus Kulzer, Alemanha, cor B1), fotoativados por 20 segundos, de acordo com as recomendações do fabricante.

c. Corte das restaurações

Após armazenagem por 24 horas em água destilada à 37°C, os dentes serão seccionados em cortadeira de precisão (ISOMET 1000, Buheler, IL, EUA). Serão realizados cortes perpendiculares à interface de união de maneira a produzir palitos com área de secção transversal de aproximadamente 0,5 mm^2, obtendo-se cerca de 4 palitos por dente e 20 a 24 palitos por grupo.

Ensaio de microtração

Cada palito terá as dimensões mensuradas, para o cálculo da área de união, utilizando um paquímetro digital (Mitutoyo). Os palitos serão fixados em um dispositivo desenvolvido para ensaios de microtração com o auxílio de um adesivo à base de cianoacrilato (Super Bonder Gel, Loctite). As amostras serão tracionadas a uma velocidade de 0,5 mm / min. até sua falha em uma máquina de ensaios mecânicos (Emic DL 500, São José dos Pinhais, PR, Brasil) com uma célula de carga de 100N e a resistência de união mensurada (MPa).

1.4.5.2 Análise de fratura em microscopia de luz

Todas as amostras fraturadas correspondentes à dentina e ao esmalte serão observadas através de microscópio de luz disponível em microdurômetro (Futuretech) com um aumento de 100 x e 500 x para a determinação do modo de falha de cada amostra. As falhas serão classificadas em coesivas em adesivo, resina ou dentina, adesivas ou do tipo mistas.

1.4.6 Análise estatística e cálculo de amostra

O número de repetições especificadas nas metodologias acima partiu dos valores mais comumente utilizados na literatura. No entanto este trabalho compreende um projeto de desenvolvimento de um novo material, utilizando composição inédita para posterior avaliação de desempenho. Desta forma não existem dados na literatura que permitam embasamento científico para cálculo de amostra. Será utilizado ensaio piloto e de acordo com o poder do teste (igual ou superior a 0.8) será estabelecido o número de repetições necessárias para a execução do estudo, sendo a metodologia realizada em situações controladas. De posse dos resultados dos experimentos, o método estatístico mais apropriado será escolhido com base na aderência ao modelo de distribuição normal e igualdade de variância. Para todos os testes será considerado o valor $p<0,05$ como estatisticamente significante. Para a realização da análise estatística, será utilizado o programa estatístico SigmaStat 3.01 Systat INC.

1.5 REFERÊNCIAS

ALBRECHT, W. N.; STEPHENSON, R. L. Health hazards of tertiary amine catalysts **Scand J Work Environ Health**, v.14, n.4, p.209-219, 1988.

AMES, B. N. Dietary carcinogens and anticarcinogens. Oxygen radicals and degenerative diseases **Science**, v.221, n.4617, p.1256-1264, 1983.

ANDRZEJEWSKA, E. Photopolymerization kinetics of multifunctional monomers **Progress in Polymer Science**, v.26, n.4, p.605-665, 2001.

CHEN, Y. C.; FERRACANE, J. L.; PRAHL, S. A. Quantum yield of conversion of the photoinitiator camphorquinone **Dent Mater**, v.23, n.6, p.655-664, 2007.

COX, C. F.; HAFEZ, A. A.; AKIMOTO, N.; OTSUKI, M.; SUZUKI, S.; TARIM, B. Biocompatibility of primer, adhesive and resin composite systems on non-exposed and exposed pulps of non-human primate teeth **Am J Dent**, v.11 Spec No, p.S55-63, 1998.

DE MUNCK, J.; VAN LANDUYT, K.; PEUMANS, M.; POITEVIN, A.; LAMBRECHTS, P.; BRAEM, M.; VAN MEERBEEK, B. A critical review of the durability of adhesion to tooth tissue: methods and results **J Dent Res**, v.84, n.2, p.118-132, 2005.

GOLDBERG, M. In vitro and in vivo studies on the toxicity of dental resin components: a review **Clin Oral Investig**, v.12, n.1, p.1-8, 2008.

GUO, X.; SPENCER, P.; WANG, Y.; YE, Q.; YAO, X.; WILLIAMS, K. Effects of a solubility enhancer on penetration of hydrophobic component in model adhesives into wet demineralized dentin **Dent Mater**, v.23, n.12, p.1473-1481, 2007.

GUO, X.; WANG, Y.; SPENCER, P.; YE, Q.; YAO, X. Effects of water content and initiator composition on photopolymerization of a model Bis-GMA/HEMA resin **Dent Mater**, v.24, n.6, p.824-831, 2008.

HICKEY, M. J. Investigation of the chemical constituents of Brazilian sassafras oil **J Org Chem**, v.13, n.3, p.443-446, 1948.

HIGASHI, C.; MICHEL, M. D.; REIS, A.; LOGUERCIO, A. D.; GOMES, O. M.; GOMES, J. C. Impact of adhesive application and moisture on the mechanical properties of the adhesive interface determined by the nano-indentation technique **Oper Dent**, v.34, n.1, p.51-57, 2009.

JAKUBIAK, J.; ALLONAS, X.; FOUASSIER, J. P.; SIONKOWSKA, A.; ANDRZEJEWSKA, E.; LINDEN, L. A.; RABEK, J. F. Camphorquinone-amines photoinitating systems for the initiation of free radical polymerization **Polymer**, v.44, n.18, p.5219-5226, 2003.

JOSEPHY, P. D.; EVANS, D. H.; PARIKH, A.; GUENGERICH, F. P. Metabolic activation of aromatic amine mutagens by simultaneous expression of human cytochrome P450 1A2, NADPH-cytochrome P450 reductase, and N-acetyltransferase in Escherichia coli **Chem Res Toxicol**, v.11, n.1, p.70-74, 1998.

JOSHI, R.; KUMAR, M. S.; SATYAMOORTHY, K.; UNNIKRISNAN, M. K.; MUKHERJEE, T. Free radical reactions and antioxidant activities of sesamol: pulse radiolytic and biochemical studies **J Agric Food Chem**, v.53, n.7, p.2696-2703, 2005.

KUMAGAI, Y.; LIN, L. Y.; SCHMITZ, D. A.; CHO, A. K. Hydroxyl radical mediated demethylenation of (methylenedioxy)phenyl compounds **Chem Res Toxicol**, v.4, n.3, p.330-334, 1991.

LISO, P. A.; VAZQUEZ, B.; REBUELTA, M.; HERNAEZ, M. L.; ROTGER, R.; SAN ROMAN, J. Analysis of the leaching and toxicity of new amine activators for the curing of acrylic bone cements and composites **Biomaterials**, v.18, n.1, p.15-20, 1997.

LIU, S.; SHI, S.; HOU, G.; NIE, J. Benzodioxole derivative as coinitiator for dental resin **Acta Odontol Scand**, v.65, n.6, p.313-318, 2007.

MATOS, M. A.; MONTE, M. J.; SOUSA, C. C.; ALMEIDA, A. R.; MORAIS, V. M. Thermodynamic study of sesamol, piperonyl alcohol, piperonylic acid and homopiperonylic acid: a combined experimental and theoretical investigation **Org Biomol Chem**, v.2, n.6, p.908-914, 2004.

MICALE, N.; ZAPPALA, M.; GRASSO, S. Synthesis and antitumor activity of 1,3-benzodioxole derivatives **Farmaco**, v.57, n.10, p.853-859, 2002.

MICALE, N.; ZAPPALA, M.; GRASSO, S. Synthesis and cytotoxic activity of 1,3-benzodioxole derivatives. Note II **Farmaco**, v.58, n.5, p.351-355, 2003.

MOREIRA, D. R. M.; LEITE, A. C. L.; FERREIRA, P. M. P.; COSTA, P. M.; LOTUFO, L. V. C.; MORAES, M. O.; BRONDANI, D. J.; PESSOA, C. O. Synthesis and antitumor evaluating of peptidyl-like derivatives containing the 1,3-benzodioxole system. **Eur J Med Chem**, v. 42, p.351-357, 2007.

MUSANJE, L.; FERRACANE, J. L.; SAKAGUCHI, R. L. Determination of the optimal photoinitiator concentration in dental composites based on essential material properties **Dent Mater**, 2009.

NEUMANN, M. G.; SCHMITT, C. C.; FERREIRA, G. C.; CORREA, I. C. The initiating radical yields and the efficiency of polymerization for various dental photoinitiators excited by different light curing units **Dent Mater**, v.22, n.6, p.576-584, 2006.

OGLIARI, F. A.; ELY, C.; LIMA, G. S.; CONDE, M. C.; PETZHOLD, C. L.; DEMARCO, F. F.; PIVA, E. Onium salt reduces the inhibitory polymerization effect from an organic solvent in a model dental adhesive resin **J Biomed Mater Res B Appl Biomater**, v.86, n.1, p.113-118, 2008.

OGLIARI, F. A.; ELY, C.; PETZHOLD, C. L.; DEMARCO, F. F.; PIVA, E. Onium salt improves the polymerization kinetics in an experimental dental adhesive resin **J Dent**, v.35, n.7, p.583-587, 2007.

OGUNYINKA, A.; PALIN, W. M.; SHORTALL, A. C.; MARQUIS, P. M. Photoinitiation chemistry affects light transmission and degree of conversion of curing experimental dental resin composites **Dent Mater**, v.23, n.7, p.807-813, 2007.

PELKA, M.; DANZL, C.; DISTLER, W.; PETSCHELT, A. A new screening test for toxicity testing of dental materials **J Dent**, v.28, n.5, p.341-345, 2000.

SCHNEIDER, L. F.; CONSANI, S.; SAKAGUCHI, R. L.; FERRACANE, J. L. Alternative photoinitiator system reduces the rate of stress development without compromising the final properties of the dental composite **Dent Mater**, 2008.

SCHROEDER, W. F.; COOK, W. D.; VALLO, C. I. Photopolymerization of N,N-dimethylaminobenzyl alcohol as amine co-initiator for light-cured dental resins **Dent Mater**, v.24, n.5, p.686-693, 2008.

SCHROEDER, W. F.; VALLO, C. I. Effect of different photoinitiator systems on conversion profiles of a model unfilled light-cured resin **Dent Mater**, v.23, n.10, p.1313-1321, 2007.

SHI, S.; NIE, J. Investigation of 3,4-methylenedioxybenzene methoxyl methacrylate as coinitiator and comonomer for dental application **J Biomed Mater Res B Appl Biomater**, v.82, n.2, p.487-493, 2007.

SHI, S.; NIE, J. A natural component as coinitiator for unfilled dental resin composites **J Biomed Mater Res B Appl Biomater**, v.82, n.1, p.44-50, 2007.

SHIN, D. H.; RAWLS, H. R. Degree of conversion and color stability of the light curing resin with new photoinitiator systems **Dent Mater**, 2009.

Site do Instituto Brasileiro de Plantas Medicinais (IBPM) < http://www.ibpm.org.br > acesso em 26 abril, 2009.

TAGASHIRA, M.; OHTAKE,Y. A new antioxidant 1.3-benzodioxole from Melissa officinalis. **Planta Med,** v. 64, p.555-558, 1998.

TAGASHIRA, M.; UCHIYAMA, K.; YOSHIMURA, T.; SHIROTA, M.; UEMITSU, N. Inhibition by hop bract polyphenols of cellular adherence and water-insoluble glucan synthesis of mutans streptococci **Biosci Biotechnol Biochem**, v.61, n.2, p.332-335, 1997.

TSENG, T. H.; TSHENG, Y. M.; LEE, Y. J. Cytotoxicity effects of di- and tri-hydroxybenzaldehydes as a chemopreventive potential agent on tumor cells **Toxicology**, v.161, n.3, p.179-187, 2001.

WEISBURGER, E. K.; RUSSFIELD, A. B.; HOMBURGER, F.; WEISBURGER, J. H.; BOGER, E.; VAN DONGEN, C. G.; CHU, K. C. Testing of twenty-one environmental aromatic amines or derivatives for long-term toxicity or carcinogenicity **J Environ Pathol Toxicol**, v.2, n.2, p.325-356, 1978.

YOSHIDA, K.; GREENER, E. H. Effect of photoinitiator on degree of conversion of unfilled light-cured resin **J Dent**, v.22, n.5, p.296-299, 1994.

YOSHIDA, K.; GREENER, E. H. Effects of two amine reducing agents on the degree of conversion and physical properties of an unfilled light-cured resin **Dent Mater**, v.9, n.4, p.246-251, 1993.

1.6 ORÇAMENTO

Quadro 1 - Orçamento previsto para a viabilização do projeto.

Descrição	**Qtde.**	**Custo Unit. (R$)**	**Total R$**
1,3 Benzodioxola [1,2-(Metilendioxi)-benzol] (Aldrich)	50 g	160,00	160,00
Álcool Piperonílico (Aldrich)	25 g	99,00	99,00
Etil-dimetilamino benzoato (Fluka)	100 g	300,00	300,00
Canforoquinona (Esstech)	25 g	410,00	410,00
Bisfenol A glicidil dimetacrilato	500g	188,00	188,00
Trietilenoglicol dimetacrilato	250ml	197,00	197,00
2-hidroxietil metacrilato (Aldrich)	500 g	525,80	525,80
Pentóxido de fósforo (Synth)	100 g	265,00	265,00
Etanol absoluto (Synth)	500 ml	9,74	19,48
Cloreto de Metileno	500 ml	60,00	180,00
Cera pegajosa	4 cx.	25,00	100,00
Compósito restaurador commercial	6	60,00	360,00
Nitrogênio liquid	10 l	2,00	20,00
Cromatografia de camada delgada	1cx	900,00	900,00
Ressonância magnética	2 análises	100,00	200,00
Infravermelho FTIR	2 análises	80,00	160,00
Gel de cianoacrilato	8 un.	10,00	80,00
Disco diamantado de corte	1 un.	1600,00	1600,00
Lixas metalográficas	30 un.	1,50	45,00
Resina epóxica	1 kit	150,00	150,00
Cartucho de tinta – impressora	1	150,00	150,00
Folhas A4	2 pct.	5,00	10,00
Impressão da Tese	5 un.	50,00	250,00
Transporte e estadias - GBMD 2009	-	-	2.000,00
Serviço de revisão do Inglês	2	400,00	800,00
		Total R$	**6.371,28**

1.7 CRONOGRAMA

Quadro 2. Cronograma previsto de atividades.

Tempo	MÊS	REVISÃO DE LITERATURA	SUBMISSÃO AO COMITÊ DE ÉTICA	ENSAIO LABORATORIAL	REDAÇÃO DO ARTIGO CIENTÍFICO	SUBMISSÃO DO ARTIGO CIENTÍFICO	ECERRAMENTO
2007	Julho	X					
	Agosto	X		X			
	Setembro	X		X			
	Outubro	X		X			
	Novembro	X		X			
	Dezembro	X					
2008	Janeiro	X					
	Fevereiro	X		X	X		
	Março	X		X	X		
	Abril	X		X			
	Maio	X		X			
	Junho	X		X			
	Julho	X		X			
	Agosto	X		X			
	Setembro	X		X	X		
	Outubro	X		X	X		
	Novembro	X					
	Dezembro	X					
2009	Janeiro	X					
	Fevereiro	X		X			
	Março	X		X			
	Abril	X	X	X			
	Maio	X		X	X		Qualificação
	Junho	X			X		
	Julho					X	Defesa

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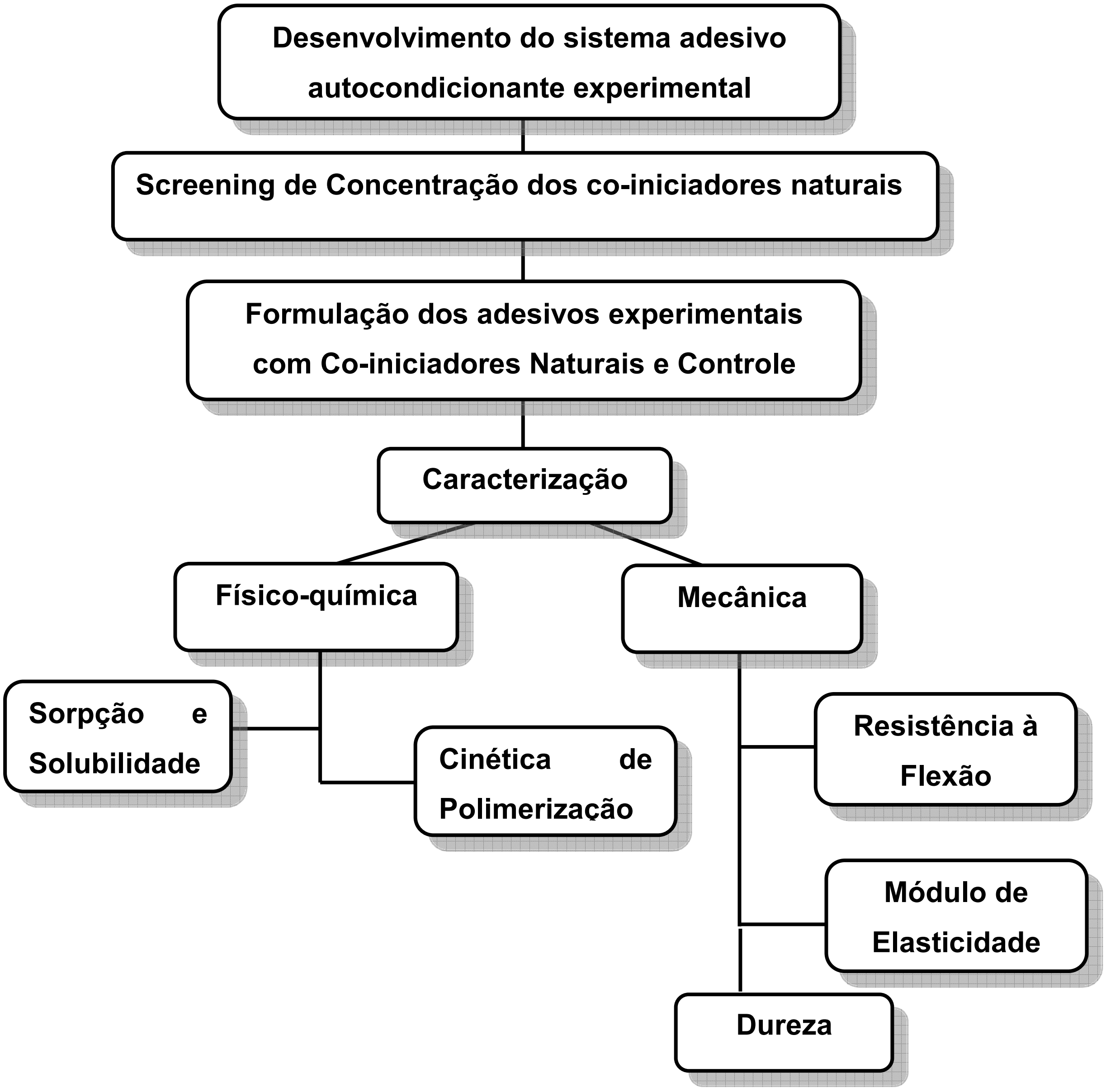
1.8 FINANCIAMENTO

O projeto conta com financiamento:
EDITAL UNIVERSAL CNPq (478731/2007-8). Novos componentes fosfatados polimerizáveis aplicados no desenvolvimento de sistemas adesivos autocondicionantes, Edital Universal CNPQ 2007 (2007-2009). Valor R$ 34.465,00;

Esstech, com doação de reagentes.

1.9 ORGANOGRAMAS DO PROJETO

Correspondente ao Artigo 1.



Nota: Este artigo será submetido ao periódico Acta Biomaterialia

Correspondente ao Artigo 2.

Nota: Este artigo será submetido ao periódico Dental Materials

2 ARTIGO 1*

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Title: Benzodioxoles as alternative co-initiators for radical polymerization in a model dental adhesive resin.

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Abstract

This study evaluated the effectiveness of benzodioxole derivatives as co-initiators of radical polymerization in a model dental adhesive resin. To compose the model dental adhesive resin, a monomer mixture based on 50 wt % of bisphenol A glicidyl dimethacrylate (Bis-GMA), 25 wt% of triethyleneglycol dimethacrylate (TEGDMA) and 25 wt% of 2-hydroxyethyl methacrylate (HEMA), was used. Camphorquinone (CQ) 1 mol % was used as a photoinitiator to initiate polymerization. 1,3-benzodioxole (BDO) and piperonyl alcohol (PA) were used as co-initiators at 0.25, 0.5, 1, 2, 4, 8 and 16 mol % level. Additionally, tertiary amine (EDAB) was used as co-initiator in the control group. The physical, chemical and mechanical properties of the polymer formed in the experimental adhesives were evaluated using the kinetics of polymerization, sorption and solubility, flexural strength and elastic modulus. The results indicated that BDO and PA were effective co-initiators in the photoinitiator system based on CQ. Comparisons between the benzodioxole derivatives co-initiators and traditionally used amine EDAB, showed similar performance in the kinetics of polymerization, flexural strength, water sorption and solubility of the model dental adhesive resin evaluated. The findings suggested that BDO and PA were feasible alternatives to conventional amine as co-initiator, moreover, the characteristics of BDO in the human diet made its use in dental adhesive resin formulations more promising and advantageous than using amine.

Keywords: adhesive systems, photoinitiation, co-initiators, benzodioxoles, amines.

Introduction

Several photoinitiators have been developed for dental materials, and continuing efforts are being made to obtain more efficient photoinitiator systems, exhibiting faster polymerization rates, better photosensitivity, and improved polymer properties.[1-4] The polymerization of visible light-activated dental polymers is generally photoinduced by camphorquinone(CQ)–amine combinations, which have been applied in most of commercial resins.[5, 6] CQ (2,3-bornanedione) has 166.22 g/mol molecular weight and an absorption peak around 468 nm.[6] As a Type II photoinitiator, CQ requires a co-initiator.[7] The initiation of polymerization occurs through the H-donor radical (R·), whereas the inactive ketyl radical disappears by a radical coupling process.[1] CQ presents low polymerization quantum yield and polymerization efficiency due to the formation of radicals following light absorption and the chain initiation performance of these radicals, which varies with the light source.[8]

The co-initiators most commonly used associated with CQ are tertiary aliphatic or aromatic amines.[6, 7] These photoinitiation systems are effective, generating polymers with adequate mechanical properties.[5, 7, 9] However, amines are known to be potentially toxic and mutagenic, and aromatics amines have been noteworthy carcinogens.[10-14] Amines are also known to form by-products during the photoreaction, causing yellow to brown discoloration under the influence of light and heat.[15]

Contemporary restorations have failed because of pigmentation and deficient marginal sealing.[16-18] Investigation into the longevity, [19-21] biocompatibility and cytotoxicity of the chemical constituents of adhesive materials have been gaining prominence. [22-24] In vitro and in vivo studies have demonstrated that adhesive components have cytotoxic effects, [22-24] moreover, these effects could be due to deficient polymerization. From this aspect it is strategic to investigate components that could promote improvements in biological characteristics, and maintain good mechanical properties.

Co-initiators alternative to amine have been studied.[4, 25, 26] Some antioxidants present in the human diet are interesting because of their protective action against free radicals produced under oxidative stress associated with their biocompatibility.[27] Benzodioxole derivatives (Table 1) are widely distributed in nature and have been shown to have antitumor, radioprotective, cosmetic,[28] antifungal, antibacterial and many other biological activities.[29]

Considering the promising use of benzodioxole derivatives for methacrylate radical polymerization, the aim of this study was to advance the knowledge about polymer network development according to the type and concentration of benzodioxole derivatives used as co-initiators in a model dental adhesive resin. The null hypothesis tested was that there would be no differences in the physicochemical and mechanical characteristics of these adhesive resins when compared with the amine-based co-initiator.

Materials and Methods

Reagents

Bisphenol A glycidyl dimethacrylate (Bis-GMA), triethylene glycol dimethacrylate (TEGDMA), 2-hydroxyethyl methacrylate (HEMA) and camphorquinone (CQ) were supplied by Esstech Inc. (Essington, PA, USA) and used without further purification. The 1,3 benzodioxole (BDO), piperonyl alcohol (PA) and ethyl 4-dimethylamino benzoate (EDAB) were purchased from Aldrich Chemical Co (Milwaukee, WI, USA) and used without further processing. To perform the monomer light-activation, a quartz-tungsten-halogen light-activation unit (XL 3000, 3M ESPE, St. Paul, MN, USA) was used and the irradiation value ($\geq$ 650 mW/cm^2) was confirmed with a digital power meter (Ophir Optronics, Danvers, MA, USA).

Formulations

A model dental adhesive resin was formulated by the intensive mixing of 50 wt % Bis-GMA, 25 wt % TEGDMA and 25 wt % HEMA. CQ was added at 1 mol % for all groups, according to the monomer moles. Three initiator systems were investigated: CQ+BDO, CQ+PA and CQ+EDAB (Table 2). The alternative co-initiators were tested at various molar concentrations: 0.25, 0.5, 1, 2, 4, 8 or 16 mol % of either BDO or PA. The control resin was composed of CQ and 1 mol % of EDAB (a sufficient concentration observed in previous studies) [3, 7]. No radical scavenger was added to avoid interference with the polymerization kinetics.

Kinetics of polymerization by RT-FTIR spectroscopy

The degree of conversion of the experimental adhesive resins was evaluated using real time Fourier Transform infrared spectroscopy (RT-FTIR) with a Shimadzu Prestige 21 spectrometer (Columbia, MD, USA) equipped with an attenuated total reflectance device composed of a horizontal ZnSe crystal, with a 45º mirror angle (PIKE Technologies, Madison, WI, USA). A support was coupled to the spectrometer, holding the light-curing unit and standardizing the distance between the fiber tip and the sample at 5 mm.

The IR Solution software (Shimadzu) was used in the monitoring scan mode, using Happ-Genzel apodization, at a range of 1750 and 1550 cm^{-1}, resolution of 8 cm^{-1} and mirror speed of 2.8 mm/s. With this setup, 1 scan every 1 second was acquired during light-activation.[30] Analysis was performed at a controlled room temperature of 23 ± 1 ºC and relative humidity < 60 %. The sample (3 µL) was dispensed directly onto the ZnSe crystal and immediately photo-activated for 60 s. The degree of conversion for each scan was calculated as described in a previous study, [3] considering the intensity of carbon-carbon double bond stretching vibration (peak height) at 1635 cm^{-1}, and as an internal standard, the symmetric ring stretching was used at 1610 cm^{-1} from the polymerized and unpolymerized samples.

Kinetics of polymerization data were plotted and Hill's 3 parameter nonlinear regressions were used for curve fitting. As the coefficient of determination was close to 0.99 for all curves, the rate of polymerization (R_p) was calculated considering the data fitting. For characterization of different concentrations of alternative co-initiators, the data were grouped as shown in Figures 1 and 2. The Kinetics and rate of polymerization behavior of groups A_{EDAB}, A_{BDO} e A_{PA} are illustrated in Figure 3.

Flexural strength and Elastic modulus

Flexural strength (σ) and elastic modulus (E) were obtained using a mini-flexural three-point bending test. Ten bar-shaped specimens (10 mm length × 2 mm width × 2 mm height) were made for each resin, using customized stainless steel molds. The model dental adhesive resin was placed into the mold, which was placed on top of an acetate strip. The top and bottom surfaces of the specimens were then light-polymerized with two irradiations of 20 s on each side. After polymerization, the specimens were removed from the mold and stored in distilled water at 37 ± 1 °C for 24 h. The flexural test was performed in a universal testing machine at a crosshead speed of 0.5 mm/min. The maximum loads were obtained and the σ was calculated in MPa using the following formula:

$$\sigma = 3FL/(2BH^2)$$

where F is the maximum load (N); L is the distance between the supports rods (mm); B is the width of the specimen (mm) and H, the height (mm).

The elastic modulus, in MPa, was determined as:

$$E=FL^3/4BH^3d$$

where F is the maximum load; L is the distance between the supports; B is the width of the specimen, H is the height of the specimen, and d is the deflection (mm) corresponding to the load F.

To evaluate the influence of benzodioxole derivative co-initiator type and concentration , the flexural strength and elastic modulus data were submitted to two-way ANOVA (factor concentration and factor co-initiator) and Tukey's *post hoc* test for multiple comparisons in different concentrations and co-initiators used. For co-initiator comparisons the formulations shown in Table 2 were used. Data for σ and E, were submitted to one-way ANOVA and Tukey's *post hoc* test for comparisons with the control.

Water sorption and solubility

Water sorption (WS) and solubility (SL) were determined according to ISO specifications 4049/2000. Ten disc-shaped specimens were fabricated in an aluminum mold between two glass slides covered with polyethylene film. After removing the glass slides, the specimens were light-activated for 20 s on both top and bottom surfaces. Immediately after polymerization, the specimens were placed in a desiccator containing freshly dried silica gel and calcium chloride. After 24 h, the samples were removed, stored in a desiccator at 23 °C for 1 h and weighed on an analytical balance with a precision of 0.001mg (AUW 220D, Shimadzu Corp., Kyoto, Japan). This cycle was repeated until a constant mass ($m1$) was obtained. Thickness and diameter of the specimens were randomly measured in five locations using a digital caliper (Digimatic Caliper 500-144B, Mitutoyo Sul Americana, Suzano, SP, Brazil) rounded off to the nearest 0.01 mm, and these measurements were used to calculate the volume (V) of each specimen (in mm^3). The discs were immersed in distilled water at 37°C for 7 days, then removed, blotted dry and weighed ($m2$). After weighing, the specimens returned to the desiccator and were weighed daily to record a third constant mass ($m3$). For each disc, WS and SL were calculated using the following formulas:

$$WS = (m_2 - m_3) / V \qquad SL = (m_1 - m_3) / V$$

Data for WS and SL, were submitted to one-way ANOVA and Tukey's *post hoc* test for multiple comparisons. The statistical significance for all analysis was established at the 0.05 level of significance.

Results

Kinetics of polymerization

Figures 1 and 2 show the kinetics of polymerization of experimental adhesive resins at the same CQ concentration (1 mol %) and different BDO and PA molar concentrations (0.25, 0.50, 1, 2, 4, 8 and 16 %). The effects of the concentration and type of the benzodioxole derivative were investigated by the R_p performance, illustrated in Figure 4. BDO showed smaller effectiveness than PA, at the lower concentrations (0.25 % and 0.50 %), as indicated by the kinetics and rate of polymerization analysis.

The kinetics of polymerization of the selected concentration (4 mol %) of benzodioxole derivatives co-initiators were compared with EDAB as co-initiator (control group), and are shown in Figure 3. The rate of polymerization showed that the control group presented higher Rp^{max} than the other groups.

Flexural strength and elastic modulus

For flexural strength only the factor *concentration* was statistically significant ($p<0.001$) according to Two-way ANOVA. (Figure 5). Two-Way ANOVA used for comparisons of Elastic modulus showed that the factors *concentration* ($P<0.001$), *co-initiators* ($P=0.031$) and *interaction* ($P<0.001$) were significant. The co-initiator factor

(BDO or PA), was significant for the concentrations of 1, 2 and 16 mol %. Comparisons for concentration are demonstrated in Figure 6. No significant differences were detected between concentrations of 1, 2 and 4 molar % regarding flexural strength and elastic modulus. The groups A_{PA} and A_{EDAB} showed similar flexural strength means. (Table 3) However, the elastic modulus mean of Group A_{BDO} was statistically higher than that of group A_{EDAB} (p=0.007) but similar to that of group $A_{PA,}$ which showed intermediate and statistically similar means, according one way ANOVA.

Sorption and solubility

Groups A_{BDO}, A_{PA}, A_{EDAB} showed similar means of sorption and solubility in water according to One-Way ANOVA (Table 4).

Discussion

The R_p is a critical issue for methacrylates in dental applications, particularly in the case of dental adhesives. This class of materials is polymerized *in situ*, in an adverse environment, prone to contamination by water and other biological fluids. Several factors may influence the R_P in radical photoinitiated polymerizations, such as temperature, [31] degree of functionality of the monomer, [1] concentration of the photoinitiator [6] and co-initiator, as well their nature, [32, 33] and the effectiveness of the curing light. [8]

In the present study, the kinetics of polymerization was remarkably influenced by the concentration of the benzodioxole derivative, irrespective of the type of co-initiator used. As observed in Figure 1 (a) and (b), at 20 s, 0.25 mol % of BDO was unable to promote the initiation of polymerization and 0.5 mol % of BDO obtained an insignificant degree of conversion (10.4 %). Observing the calculated R_P data, 0.5 % of BDO showed autoacceleration beginning at 15 s and an R_P^{max} of 1.8 % s^{-1} at 34 s. Corroborating these findings, it was not possible to obtain solid specimens for the σ and tests. When 1 mol% of BDO was added, a sudden increase in the reactivity of the system was observed with degree of conversion reaching an average of 25 %. Autoacceleration started in the first few seconds, reaching an R_P^{max} of 2.4 % s^{-1} at 20 s. With 4 mol% of BDO the effect of concentration on the reactivity was less pronounced, with degree of conversion around 45 % and an R_P^{max} occurring between 13 and 16 s. Unlike the low content groups (0.25 mol % and 0.5 mol % concentrations), the formulations of 1 or more mol% were able to produce solid samples. Considering that the group with 4 mol% of BDO at 20 s reached a degree of conversion similar to those of the higher BDO concentrations and a similar performance to that of the EDAB at 1 mol %, this benzodioxole group was chosen for water sorption and solubility analysis. Table 2 shows that there were no significant differences in water sorption and solubility between the BDO, PA and EDAB groups, even with the molar excess of BDO when compared with EDAB. This can be explained by the significant effect of the cross-link density and the network parameter on the hygroscopic behavior of the polymer.[34]

PA appears to be more effective in promoting initiation than BDO at low concentrations of 0.25 and 0.5 mol %, where is possible to observe a degree of conversion of 13.3 % and 22 % respectively at 20 s of light-activation time. This can be explained by the hypothetical mechanism showed in Figure 7, where three possible pathways of radical formation in the PA co-initiator are depicted (Diagrams 3, 4 and 5). Similar results were obtained elsewhere using PA as co-initiator. [27] However, for the other higher concentrations shown in Figure 4, the efficiency of BDO and PA was similar, indicating that a minimum concentration of BDO to initiate the polymerization was reached. Furthermore, in the action mechanism of the PA there are three donation possibilities (Figure 7), but it seems that there is a competition in the proton donation, explaining why PA kinetics and rate of polymerization are similar to the BDO.

Studies determining the minimum concentration of CQ and EDAB, [7, 35] which causes maximum polymerization, minimum shrinkage stress and sufficient depth of cure have been reported.[7] It is known that when low amine concentrations are present (< 0.1 wt %), the rate of radical formation is independent of the amine concentration, while for intermediate concentrations (between 0.1 and 0.5 wt %) the rate of radical formation was independent of the amine concentration and dependent only on the amine reactivity.[36] Therefore the concentration of amine and CQ used in this study could be considered high enough. In addition, the concentration of CQ and amine should be as low as possible in clinical situations, because of the yellowness and the tendency to internal discoloration of the resin.[33] As a matter of fact the system CQ and amine, extensively used in commercial adhesives, have shown strong evidences with regard to their effectiveness and durability.[16, 20, 37]

BDO and PA were shown to be effective co-initiators for dental adhesive resins light-activated by halogen light, being comparable with the conventional amine EDAB, presenting similar physical, chemical and mechanical characteristics. Moreover, the characteristic of BDO and PA being natural components makes them promising alternatives as photoinitiating systems for dental biomaterials, indicating that some biological improvements in dental adhesive resins could be obtained by using benzodioxole derivatives as co-initiators. However more in-depth evaluations and long-term analyses are necessary.

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References

1. Andrzejewska E. Photopolymerization kinetics of multifunctional monomers. Prog Polym Sci 2001;26:605-665.
2. Ogliari FA, Ely C, Lima GS, Conde MC, Petzhold CL, Demarco FF, Piva E. Onium salt reduces the inhibitory polymerization effect from an organic solvent in a model dental adhesive resin. J Biomed Mater Res B Appl Biomater 2008;86:113-118.
3. Ogliari FA, Ely C, Petzhold CL, Demarco FF, Piva E. Onium salt improves the polymerization kinetics in an experimental dental adhesive resin. J Dent 2007;35:583-587.
4. Schneider LF, Consani S, Sakaguchi RL, Ferracane JL. Alternative photoinitiator system reduces the rate of stress development without compromising the final properties of the dental composite. Dent Mater 2008.
5. Jakubiak J, Allonas X, Fouassier JP, Sionkowska A, Andrzejewska E, Linden LA, Rabek JF. Camphorquinone-amines photoinitiating systems for the initiation of free radical polymerization. Polymer 2003;44:5219-5226.
6. Chen YC, Ferracane JL, Prahl SA. Quantum yield of conversion of the photoinitiator camphorquinone. Dent Mater 2007;23:655-664.

7. Musanje L, Ferracane JL, Sakaguchi RL. Determination of the optimal photoinitiator concentration in dental composites based on essential material properties. Dent Mater 2009.

8. Neumann MG, Schmitt CC, Ferreira GC, Correa IC. The initiating radical yields and the efficiency of polymerization for various dental photoinitiators excited by different light curing units. Dent Mater 2006;22:576-584.

9. Conde MC, Zanchi CH, Rodrigues-Junior SA, Carreno NL, Ogliari FA, Piva E. Nanofiller loading level: Influence on selected properties of an adhesive resin. J Dent 2009;37:331-335.

10. Brennan RJ, Schiestl RH. The aromatic amine carcinogens o-toluidine and o-anisidine induce free radicals and intrachromosomal recombination in Saccharomyces cerevisiae. Mutat Res 1999;430:37-45.

11. Albrecht WN, Stephenson RL. Health hazards of tertiary amine catalysts. Scand J Work Environ Health 1988;14:209-219.

12. Josephy PD, Evans DH, Parikh A, Guengerich FP. Metabolic activation of aromatic amine mutagens by simultaneous expression of human cytochrome P450 1A2, NADPH-cytochrome P450 reductase, and N-acetyltransferase in Escherichia coli. Chem Res Toxicol 1998;11:70-74.

13. Liso PA, Vazquez B, Rebuelta M, Hernaez ML, Rotger R, San Roman J. Analysis of the leaching and toxicity of new amine activators for the curing of acrylic bone cements and composites. Biomaterials 1997;18:15-20.

14. Weisburger EK, Russfield AB, Homburger F, Weisburger JH, Boger E, Van Dongen CG, Chu KC. Testing of twenty-one environmental aromatic amines or derivatives for long-term toxicity or carcinogenicity. J Environ Pathol Toxicol 1978;2:325-356.

15. Schneider LF, Pfeifer CS, Consani S, Prahl SA, Ferracane JL. Influence of photoinitiator type on the rate of polymerization, degree of conversion, hardness and yellowing of dental resin composites. Dent Mater 2008;24:1169-1177.

16. da Rosa Rodolpho PA, Cenci MS, Donassollo TA, Loguercio AD, Demarco FF. A clinical evaluation of posterior composite restorations: 17-year findings. J Dent 2006;34:427-435.

17. Opdam NJ, Loomans BA, Roeters FJ, Bronkhorst EM. Five-year clinical performance of posterior resin composite restorations placed by dental students. J Dent 2004;32:379-383.

18. Gaengler P, Hoyer I, Montag R, Gaebler P. Micromorphological evaluation of posterior composite restorations - a 10-year report. J Oral Rehabil 2004;31:991-1000.

19. Toledano M, Osorio R, Osorio E, Aguilera FS, Yamauti M, Pashley DH, Tay F. Durability of resin-dentin bonds: effects of direct/indirect exposure and storage media. Dent Mater 2007;23:885-892.

20. De Munck J, Van Landuyt K, Peumans M, Poitevin A, Lambrechts P, Braem M, Van Meerbeek B. A critical review of the durability of adhesion to tooth tissue: methods and results. J Dent Res 2005;84:118-132.

21. van Dijken JW. Durability of three simplified adhesive systems in Class V non-carious cervical dentin lesions. Am J Dent 2004;17:27-32.

22. Goldberg M. In vitro and in vivo studies on the toxicity of dental resin components: a review. Clin Oral Investig 2008;12:1-8.

23. Schweikl H, Hiller KA, Eckhardt A, Bolay C, Spagnuolo G, Stempfl T, Schmalz G. Differential gene expression involved in oxidative stress response caused by triethylene glycol dimethacrylate. Biomaterials 2008;29:1377-1387.

24. Teixeira HM, Do Nascimento AB, Hebling J, De Souza Costa CA. In vivo evaluation of the biocompatibility of three current bonding agents. J Oral Rehabil 2006;33:542-550.

25. Shi S, Nie J. Investigation of 3,4-methylenedioxybenzene methoxyl methacrylate as coinitiator and comonomer for dental application. J Biomed Mater Res B Appl Biomater 2007;82:487-493.

26. Wang K, Yang D, Xiao M, Chen X, Lu F, Nie J. Sesamin as a co-initiator for unfilled dental restorations. Acta Biomater 2009;in press:in press.

27. Liu S, Shi S, Hou G, Nie J. Benzodioxole derivative as coinitiator for dental resin. Acta Odontol Scand 2007;65:313-318.

28. Matos MA, Monte MJ, Sousa CC, Almeida AR, Morais VM. Thermodynamic study of sesamol, piperonyl alcohol, piperonylic acid and homopiperonylic acid: a combined experimental and theoretical investigation. Org Biomol Chem 2004;2:908-914.

29. Regnier GH, Canevari RV, Laubie MV, Poignant JB. Benzodioxoles Compounds. United States Patent, vol. 3917597. France: Science-Union Et Cie, France, 1975.

30. Faria-e-Silva AL, Casselli DS, Lima GS, Ogliari FA, Piva E, Martins LR. Kinetics of conversion of two dual-cured adhesive systems. J Endod 2008;34:1115-1118.

31. Cook WD. THERMAL ASPECTS OF THE KINETICS OF DIMETHACRYLATE PHOTOPOLYMERIZATION. Polymer 1992;33:2152-2161.

32. Ogunyinka A, Palin WM, Shortall AC, Marquis PM. Photoinitiation chemistry affects light transmission and degree of conversion of curing experimental dental resin composites. Dent Mater 2007;23:807-813.

33. Schroeder WF, Vallo CI. Effect of different photoinitiator systems on conversion profiles of a model unfilled light-cured resin. Dent Mater 2007;23:1313-1321.

34. Ogliari FA, Ely C, Zanchi CH, Fortes CB, Samuel SM, Demarco FF, Petzhold CL, Piva E. Influence of chain extender length of aromatic dimethacrylates on polymer network development. Dent Mater 2008;24:165-171.

35. Schroeder WF, Cook WD, Vallo CI. Photopolymerization of N,N-dimethylaminobenzyl alcohol as amine co-initiator for light-cured dental resins. Dent Mater 2008;24:686-693.

36. Cook WD. Photopolymerization kinetics of dimethacrylates using the camphorquinone amine initiator system Polymer 1992:600-609.

37. Peumans M, De Munck J, Van Landuyt K, Lambrechts P, Van Meerbeek B. Five-year clinical effectiveness of a two-step self-etching adhesive. J Adhes Dent 2007;9:7-10.

Tables

Table 1. Benzodioxole derivatives used

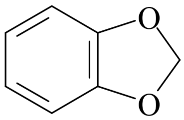
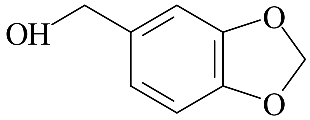
Derivative	**1,3-Benzodioxole**	**Piperonyl alcohol**
Molecular formula	$C_7H_6O_2$	$C_8H_8O_3$
Molecular weight	122.121	152.147
Molecular structure		
Synonyms	1,2- (Methylenedioxy)benzene	1,3-Benzodioxole-5-methanol 3,4-(Methylenedioxy)phenylmethanol 3,4-(Methylenedioxy)benzyl alcohol

Table 2. Composition of the different binary photoinitiating system used in comparisons of types of co-initiator

	Adhesive Resins (A)		
Composition (mol %)	A_{BDO}	A_{PA}	A_{EDAB}
Camphorquinone (CQ)	1	1	1
1,3 Benzodioxole (BDO)	4	-	-
Piperonyl alcohol (PA)	-	4	-
Ethyl 4-dimethylamino benzoate (EDAB)	-	-	1

Table 3. Flexural strength and elastic modulus of the adhesive resins with different photoinitiating systems evaluated (n=10).

Adhesive Resin (A)	Flexural strength* (MPa)	Elastic Modulus (GPa)
A_{BDO}	84.84 ± 14.02	1.19 ± 0.19 ^{a}
A_{PA}	84.57 ± 6.20	1.06 ± 0.18 ^{ab}
A_{EDAB}	74.91 ± 15.06	0.92 ± 0.14 ^{b}

Abbreviations: EDAB: Ethyl 4-dimethylaminobenzoate; BDO: 1,3 benzodioxole; PA: piperonyl alcohol. Values followed by different lowercase superscript letters in columns show statistical differences for photo-initiation system ($p<0.05$). * There were no statistical differences for adhesive resins ($p<0.05$).

Table 4. Water sorption and solubility of the experimental adhesive resins with different photoinitiator systems evaluated (n=10)

Adhesive Resin (A)	Water sorption (µg/mm³)*	Solubility (µg/mm³)*
A_{BDO}	0.221 ± 0.011	0.206 ± 0.010
A_{PA}	0.227 ± 0.014	0.211 ± 0.013
A_{EDAB}	0.226 ± 0.016	0.211 ± 0.016

Abbreviations: EDAB: Ethyl 4-dimethylaminobenzoate; BDO: 1,3 benzodioxole; PA: piperonyl alcohol.

* There were no statistical differences for adhesive resins for water sorption and solubility.

Figures Legends

Figure 1. **(a)** Polymerization kinetics of CQ/BDO (camphorquinone / 1,3-benzodioxole), binary photoinitiator system with different amounts of co-initiator; **(b)** rate of polymerization of the different BDO molar fractions.

Figure 2. **(a)** Polymerization kinetics of CQ/PA (camphorquinone / piperonyl alcohol), binary photoinitiator system with different amounts of co-initiator; **(b)** rate of polymerization of the different piperonyl alcohol molar fractions.

Figure 3. Comparison between the degree of conversion and rate of polymerization of the different binary photoinitiation systems tested: A_{BDO} (camphorquinone / 1,3-benzodioxole), A_{PA} (camphorquinone / piperonyl alcohol), A_{EDAB} (camphorquinone / ethyl 4-dimethylamine benzoate).

Figure 4. Comparison between the rate of polymerization for screening the concentration of co-initiator proposed for photoinitiator systems based on CQ and benzodioxole derivatives for adhesive resins.

Figure 5. Flexural strength evaluation of the benzodioxoles derivatives co-initiators tested in different concentrations. Different lowercase letters indicate statistical differences according to factor concentration.

Figure 6. Elastic modulus evaluation of the different concentrations of benzodioxole derivatives co-initiators tested. Different lowercase letters indicate statistical differences according to factor concentration.

Figure 7. Photochemical rearrangement mechanism of benzodioxoles derivatives evaluated in this study (1,3-benzodioxole and piperonyl alcohol) with camphorquinone in the experimental adhesive resin.

Figures

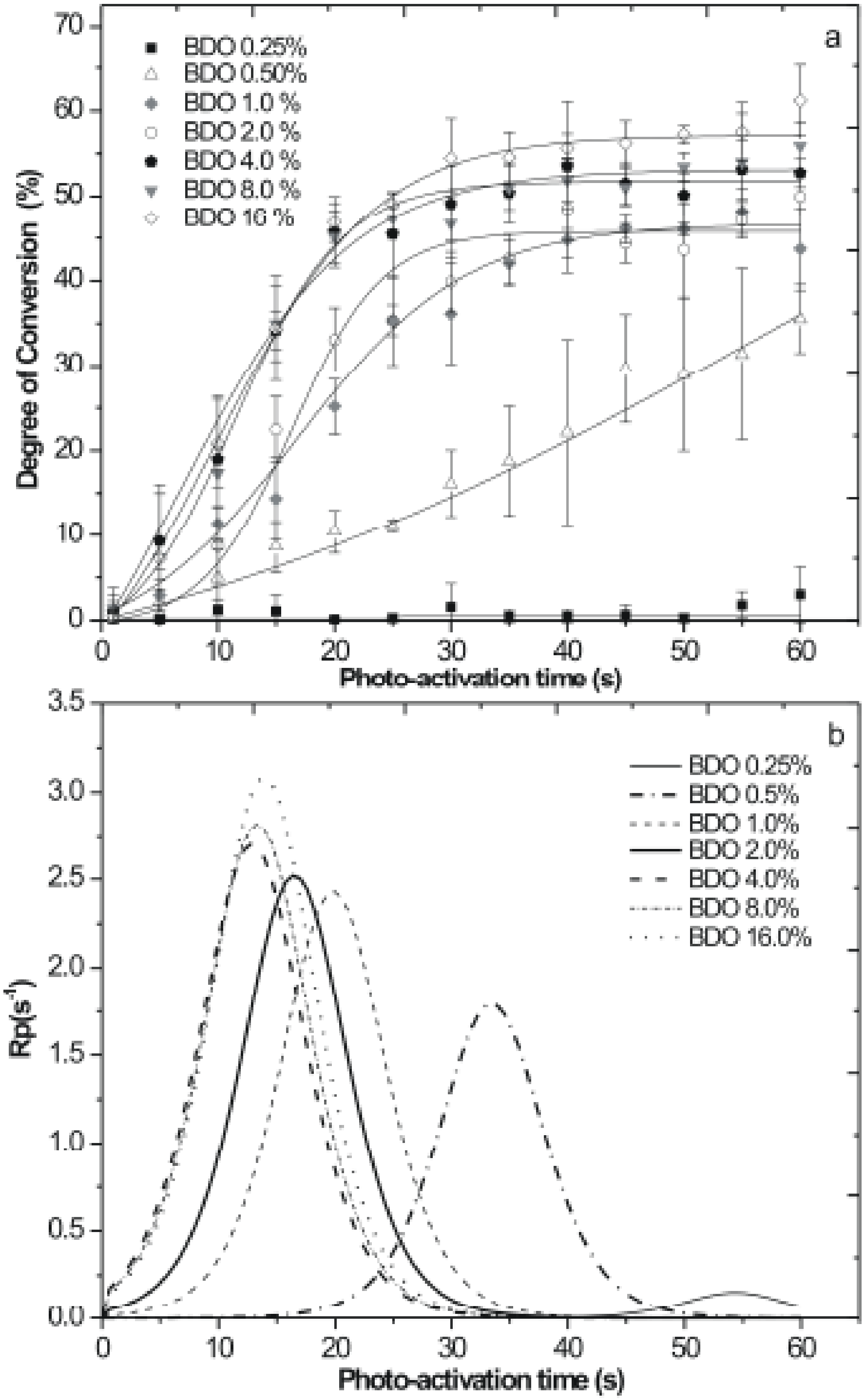


Figure 1

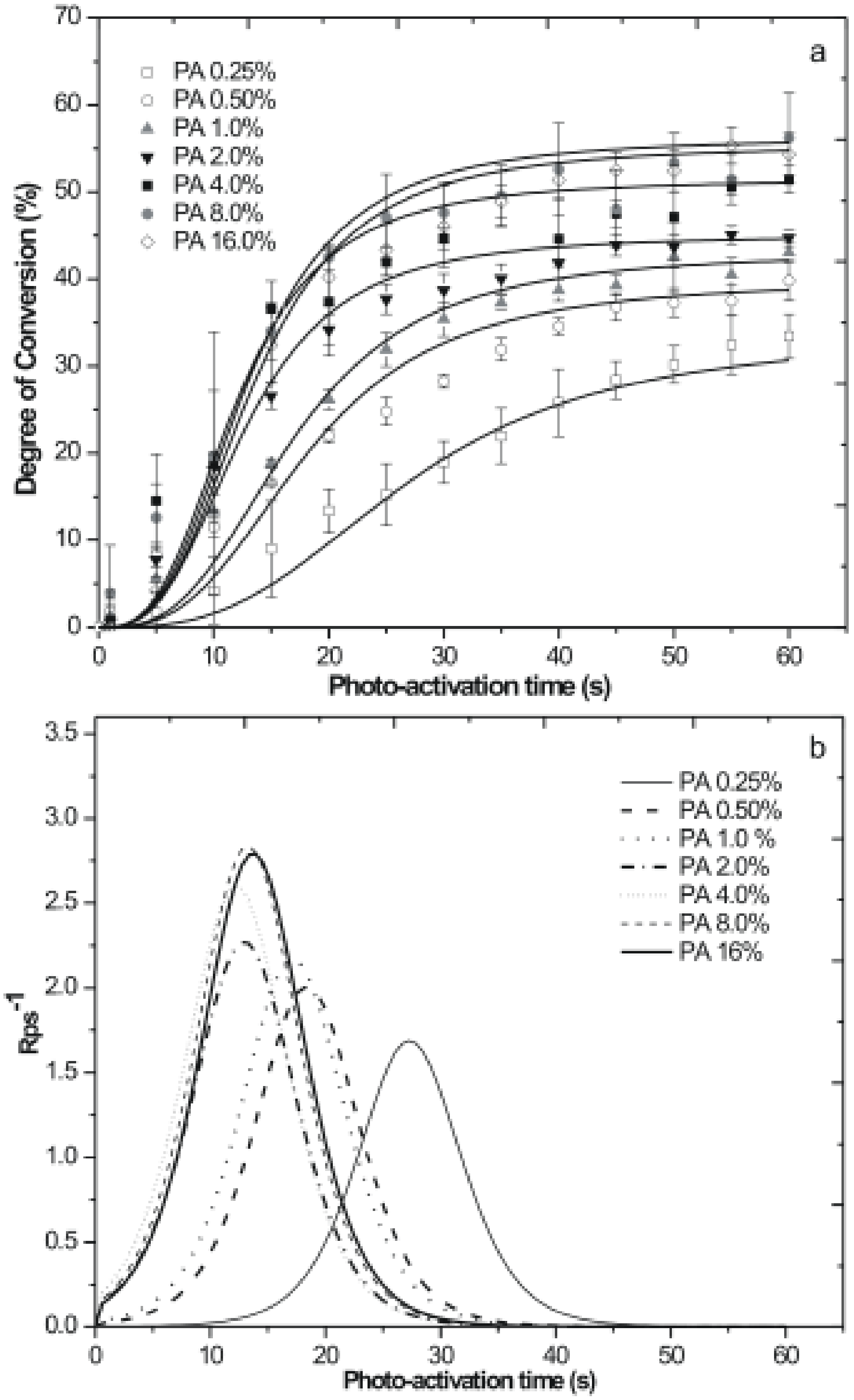

a
Degree of Conversion (%)
Photo-activation time (s)
0
10
20
30
40
50
60
70
PA 0.25%
PA 0.50%
PA 1.0%
PA 2.0%
PA 4.0%
PA 8.0%
PA 16.0%
b
$R_p s^{-1}$
0.0
0.5
1.0
1.5
2.0
2.5
3.0
3.5
PA 0.25%
PA 0.50%
PA 1.0 %
PA 2.0%
PA 4.0%
PA 8.0%
PA 16%

Figure 2

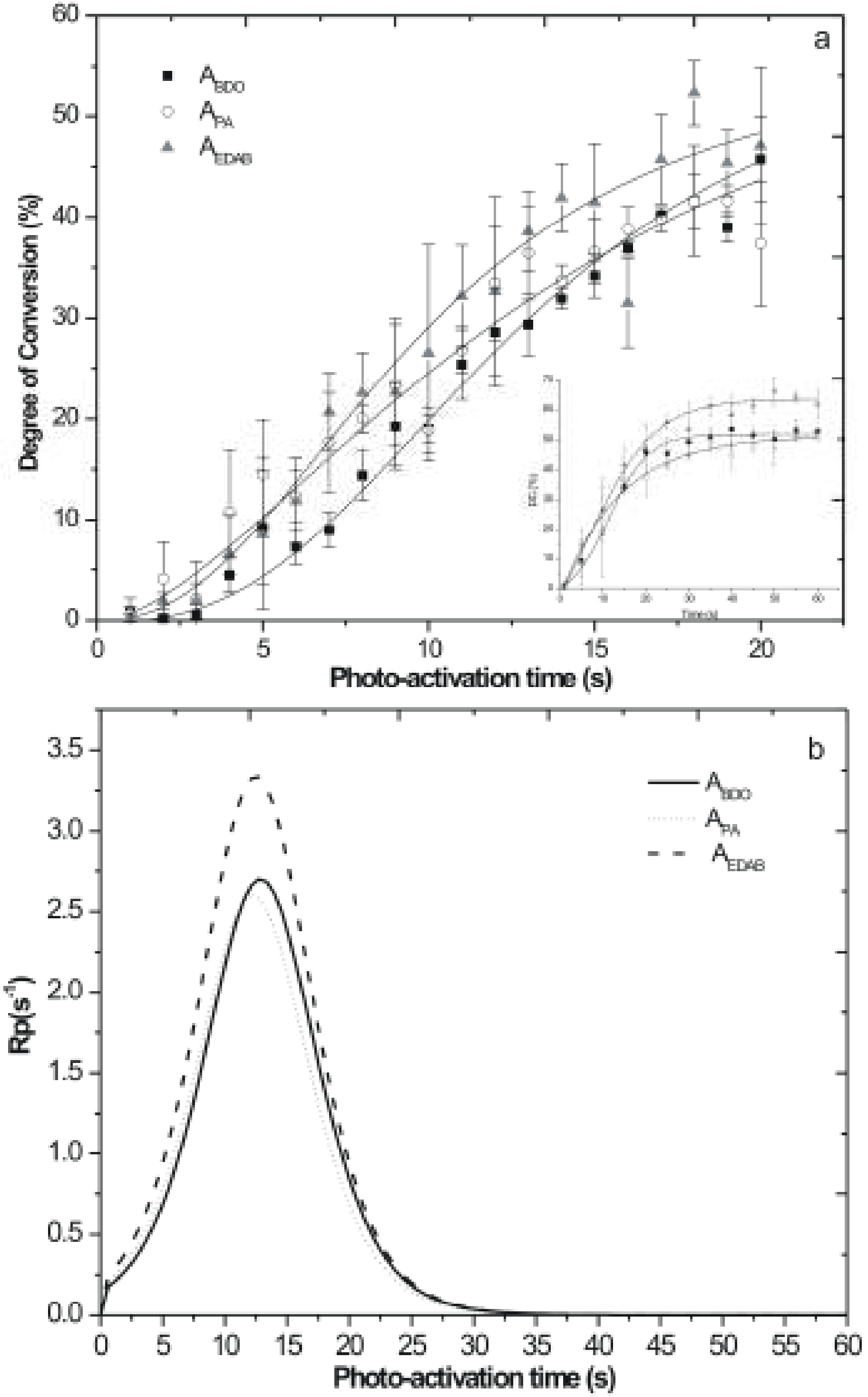

a
A_{BDO}
A_{PA}
A_{EDAB}
Degree of Conversion (%)
0
10
20
30
40
50
60
Photo-activation time (s)
0
5
10
15
20
b
A_{BDO}
A_{PA}
A_{EDAB}
Rp(s^{-1})
0.0
0.5
1.0
1.5
2.0
2.5
3.0
3.5
Photo-activation time (s)
0
5
10
15
20
25
30
35
40
45
50
55
60

Figure 3

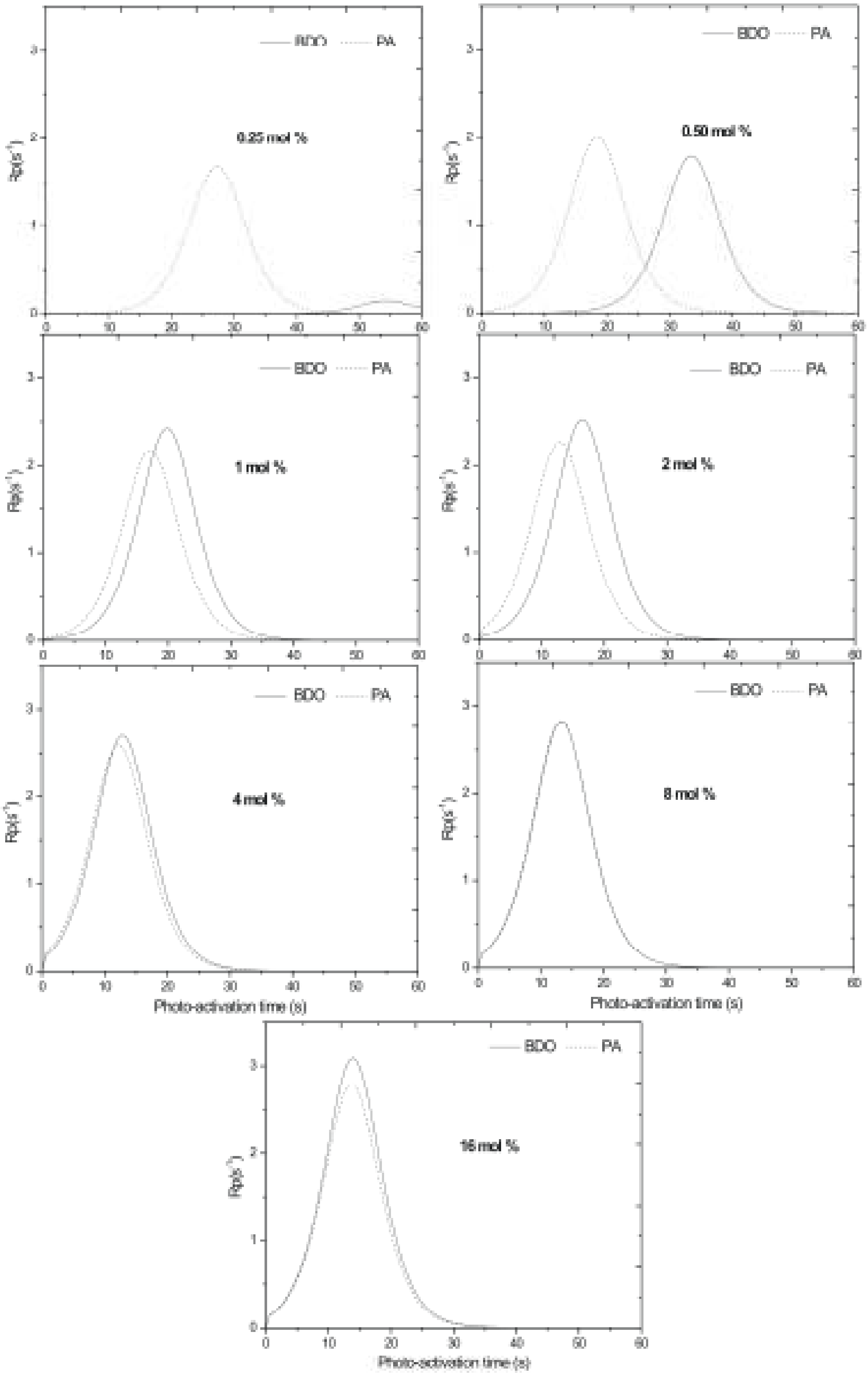
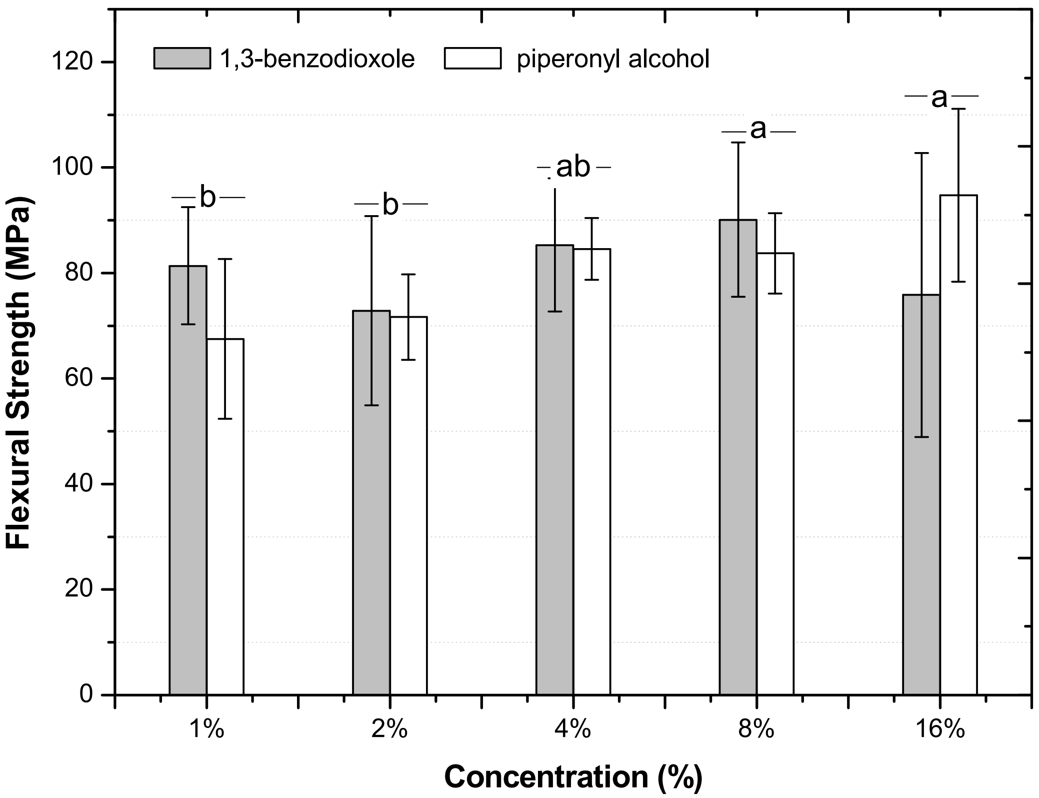
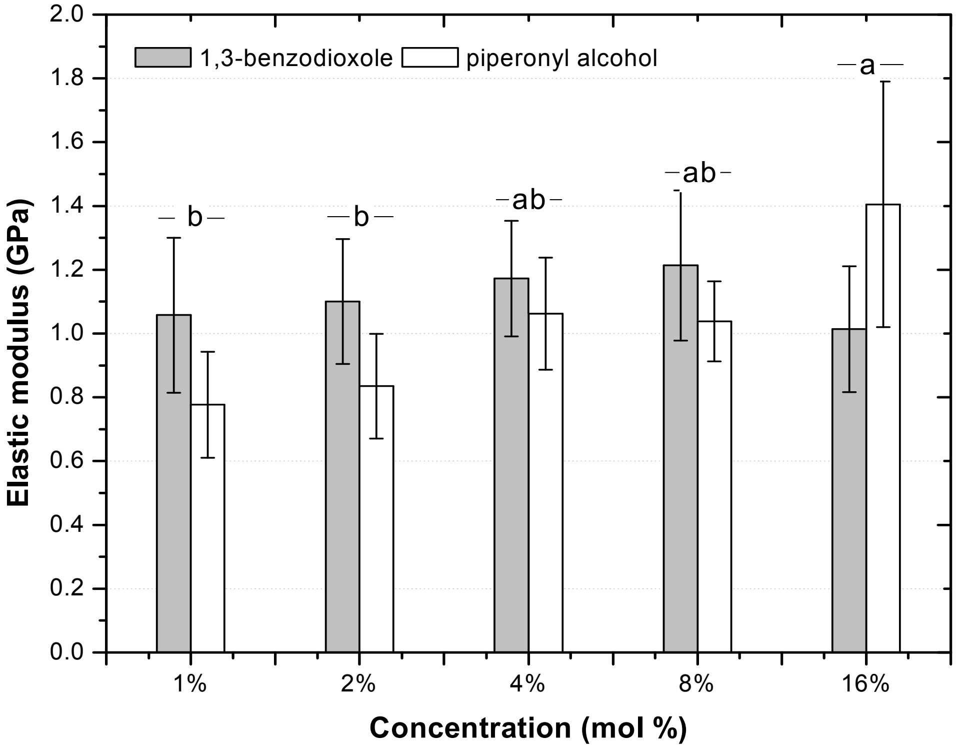

BDO
PA
0.25 mol %
0.50 mol %
1 mol %
2 mol %
4 mol %
8 mol %
16 mol %
Rp(s⁻¹)
0
1
2
3
0
10
20
30
40
50
60
Photo-activation time (s)

Figure 4



1,3-benzodioxole
piperonyl alcohol
Flexural Strength (MPa)
Concentration (%)
0
20
40
60
80
100
120
1%
2%
4%
8%
16%
b
b
ab
a
a

Figure 5



Elastic modulus (GPa)
2.0
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
1,3-benzodioxole
piperonyl alcohol
b
b
ab
ab
a
1%
2%
4%
8%
16%
Concentration (mol %)

Figure 6

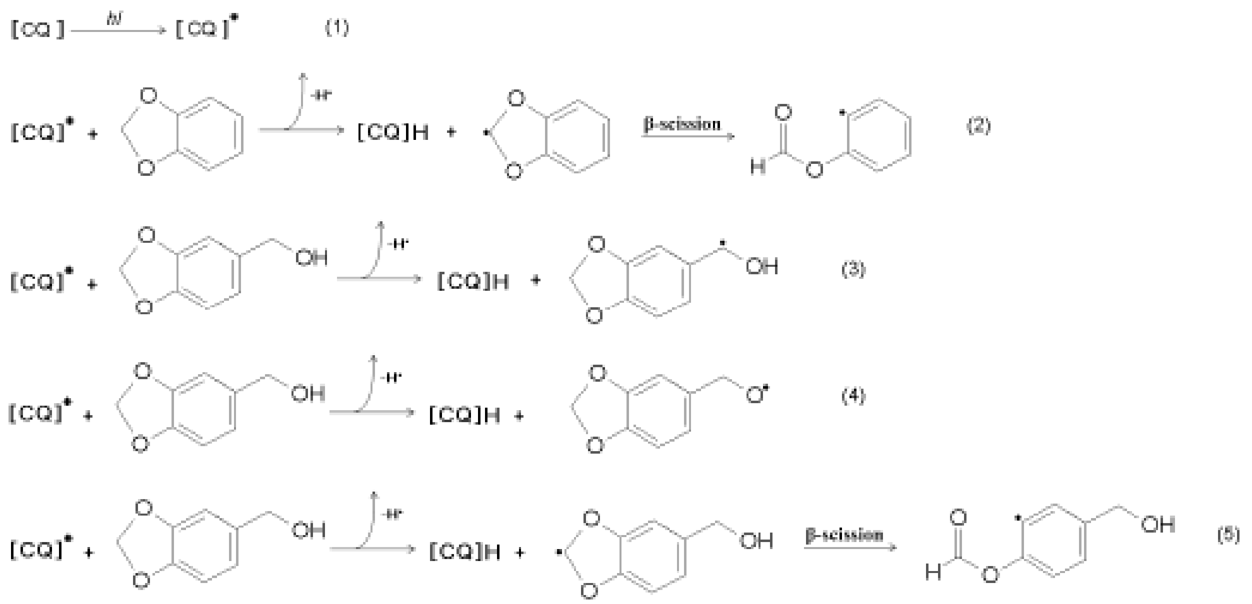


Figure 7

3 ARTIGO 2[*]

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Use of benzodioxole derivatives as natural coinitiators for dental adhesive systems

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Short title: Benzodioxoles as coinitiators for adhesives

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Abstract

Objectives: To evaluate the use of the benzodioxole derivatives, 1,3-benzodioxole (BDO) and piperonyl alcohol (PA) as natural coinitiators alternative to amine ethyl 4-dimethylamino benzoate (EDAB) in experimental self-etching adhesive systems.

Methods: A model comonomer adhesive resin of Bis-GMA, TEGDMA and HEMA was tested. Camphorquinone (1 mol%) was used as photo-sensitizer together with BDO (A_{BDO}, 4 mol%), PA (A_{PA}, 4 mol%) or EDAB (A_{EDAB}, 1 mol%) as co-initiator. The blend was used associated with an experimental self-etching primer. Bond strength to human enamel/dentin was evaluated by microtensile testing and failure analysis in light microscope. Data were submitted to Two-way ANOVA (factors coinitiator and substrate) and Tukey's *post hoc* test ($P < 0.05$). Morphology of the dentin bonding interfaces was assessed by SEM.

Results: Both factors individually were statistically significant ($P < 0.001$) whereas interaction was not significant. Means for microtensile bond strength were statitstically higher in dentin substrate ($P < 0.001$). Overall data grouped for coinitiators A_{PA} showed mean with statistically higher bond strengths than A_{EDAB} ($P < 0.001$) whereas A_{BDO} showed intermediate values. Adhesive and mixed failures were predominant. The hybrid layer for all groups was shown to be shallow (1-2 µm thick). No appreciable differences in homogeneity were detected along the bonding interface. The resin tags for A_{EDAB} were cylindrical with a smooth, clean surface. For A_{BDO} and A_{PA}, the tags were trunk-conical with rougher surfaces than for A_{EDAB}. For A_{PA}, the surface of the tags was slightly rougher compared with A_{BDO}, with accessory hybridized tubules.

Significance: The benzodioxole derivatives are promising alternatives to amines as coinitiators for the radical polymerization of dental adhesive resins.

Keywords

Amines, coinitiators, dental bonding, dental materials, dioxoles, microtensile strength, photopolymerization, self-etching adhesives

1 Introduction

In order to be suitable for clinical applications, light-cured dental resins and composites must meet several criteria. The polymerization reaction needs to be efficient, sufficiently rapid, with low heat generation. Some aspects related to the photoinitiating system of dental materials raise concern with regard to biocompatibility. The constituents of materials should not cause any toxic, inflammatory, allergic, carcinogenic or mutagenic reactions in the patient.[1]

Satisfactory mechanical properties and degree of conversion of polymers have been obtained through combination of camphorquinone (CQ) as photo-sensitizer with different types of amines as coinitiators.[1-5] However, amines are known for their toxicity and mutagenicity potential,[6,7] and also tend to degrade and cause discoloration of the polymerized material over the course of time. This has encouraged research efforts to find alternative coinitiators as a substitute for amine and improve the biocompatibility of photoinitiating systems.[8-10] In this context, benzodioxole derivatives found in natural components appear to be alternative coinitiators, providing initiating reactivity comparable with that of amines [11,12] as well as being nontoxic.[15] Benzodioxoles are present in plant products of the human diet and are widely available in nature.[13] These components have also been shown to have many biological actions, such as antifungal, antibacterial, antioxidant and antitumor activities.[14-16]

Previous studies have evaluated the use of benzodioxoles as potential coinitiators in restorative resins.[11,12,17] The use of alternative coinitiators, however, would be of great importance for adhesive systems, as they are applied in close proximity to the vital pulp tissue, and it's possible to avoid the lixiviation of toxic substances through the dentin tubules. Improvements in the biocompatibility of these systems should be obtained by using natural coinitiators. Thus, the aim of this study was to evaluate the effectiveness of benzodioxole derivatives as coinitiators of an experimental self-etching adhesive system on the bond strength to dentin/enamel and micromorphology of the dentin bonding interfaces. The null-hypotheses tested were (i) the bond strength between enamel and dentin is similar (ii) the different coinitiator type will show similar microtensile bond strength.

2 Material and Methods

2.1 Reagents

Bisphenol A glycidyl dimethacrylate (Bis-GMA), triethylene glycol dimethacrylate (TEGDMA), 2-hydroxyethyl methacrylate (HEMA) and camphorquinone (CQ) were supplied by Esstech Inc. (Essington, PA, USA) and used without further purification. The coinitiators ethyl 4-dimethylaminobenzoate (EDAB), 1,3-benzodioxole (BDO) and piperonyl alcohol (PA) were purchased from Aldrich Chemical Co. (Milwaukee, WI, USA) and used as received. The molecular structures of the coinitiators are shown in Figure 1. Ethanol was obtained from Synth (Diadema, SP, Brazil). Light-activation was performed for 20 seconds with a quartz-tungsten-halogen unit (XL 3000, 3M ESPE, St. Paul, MN, USA) with 650 mW/cm^2 irradiation confirmed with a digital power meter (Ophir Optronics, Danvers, MA, USA).

2.2 Self-etching Adhesive Development

2.2.1 Primer Formulation

One experimental self-etching primer was evaluated in this study. The primer was formulated by mixing methacrylate and acidic monomers, using water and ethanol as solvents as previously described by Lima et al, 2008.[18] The pH of 1.5, was measured using a digital pHmeter (Analion PM, Ribeirão Preto, SP, Brazil), stabilized in the solution for 1 min before measuring.

2.2.2 Adhesive Resin Formulation

A model dental adhesive resin was formulated by intensive mixing of 50 wt % of Bis-GMA, 25 wt % of TEGDMA and 25 wt % of HEMA. A fraction of 1 mol % of CQ relative to the monomer moles was added. Three adhesive resins with different binary photoinitiating systems were tested: CQ + BDO (A_{BDO}), CQ + PA (A_{PA}) and CQ + EDAB (A_{EDAB}) that was used as control. The molar concentrations of all reagents of the photoinitiator system are shown in Table1. Concentrations were determined based on pilot studies on the reactivity of the coinitiators. No additional radical scavengers were added to avoid interference in polymerization.

2.3 Microtensile Bond Strength (µTBS) Test

2.3.1 Specimen Preparation

Thirty six human molars were obtained under approval of the Local Research Ethics Committee, School of Dentistry (process 090/09) and in accordance with Human Teeth Bank requirements. The teeth were randomly assigned into six groups (six teeth per group) according to the adhesive resin or dental substrate (enamel or dentin) tested.

In groups in which dentin was tested, the occlusal enamel was removed to expose middle dentin. The occlusal enamel and dentin surfaces were wet polished with 600-grit SiC papers for 1 min to standardize the smear layer. After washing and drying with absorbent paper, the prepared surfaces were etched with the experimental primer for 30 s and gently air-dried for 10 s, then a single coat of each adhesive resin was applied and light-activated for 20 s. A restoration was built up using small increments of a restorative composite (Charisma, Heraeus Kulzer, Hanau, Germany). After storage in distilled water at 37 °C for 24 h, the specimens were sectioned perpendicularly to the bonding interfaces using a cooled diamond saw at low speed (Isomet 1000, Buehler; Lake Bluff, IL, USA). Beams with a cross-sectional surface area of approximately 0.7 mm^2 were obtained and at least three beams per tooth were allocated to μTBS testing (n = 20 per group).

2.3.2 μTBS Evaluation and Failure Analysis

The beams were precisely measured with a digital caliper (Mitutoyo, Tokyo, Japan) and fixed to the tensile testing device. The microtensile test was performed in a mechanical testing machine (DL500; EMIC, São José dos Pinhais, PR, Brazil) at a crosshead speed of 0.5 mm/min until failure.[19] Bond strengths were calculated in MPa. Data were gathered considering the factors substrate (dentin and enamel) and co-initiators (A_{PA}, A_{BDO} and A_{EDAB}) and submitted to Two-way ANOVA and Tukey's *post hoc* test at 95% confidence level. After debonding, the fractured specimens were evaluated under a light microscope at 100X and 500X magnifications. Failure modes were classified as adhesive, cohesive within substrate (enamel or dentin), cohesive within adhesive resin or mixed failure.

2.4 Morphological Analysis of the Bonding Interfaces

The protocol used for preparing the specimens was based on a previous study [20]. Two 2 mm-thick dentin discs were obtained from the mid-coronal portion of molars (n=4) using a diamond saw under running water. The dentin surfaces were polished with 600-grit SiC papers for 30 s to standardize the smear layer, and the self-etching system/adhesive resins applied as described before. A thin layer of light-activated resin cement was applied, and the two dentin discs were bonded to each other to form a dentin-cement-dentin sandwich specimen. The bonded specimens were stored in distilled water at 37 °C, for 24 h. The specimens were then vertically sectioned at the dentin-adhesive-cement interfaces and embedded in cold-cure polyester resin (Fiberglass; Porto Alegre, RS, Brazil).

After 24 h, the cut surfaces were wet-polished with 600, 1200, 1500, 2000, and 2500-grit SiC papers, and 3, 1, 0.25 and 0.1 µm polycrystalline diamond suspensions (Metadi Supreme; Buehler). The specimens were ultrasonically cleansed for 10 min between each polishing step. The surfaces were etched with 50 % phosphoric acid solution for 5 s, rinsed with distilled water for 30 s and deproteinized by immersion in 2.5 % NaOCl solution for 10 min. After ultrasonic cleansing for 1 h, the specimens were dry-stored in a container with silica gel for 2 h, at room temperature. The specimens were coated with gold and the bonding interfaces examined by scanning electron microscopy (SEM – SSX-550; Shimadzu, Tokyo, Japan).

3 Results

3.1 µTBS and Failure Analysis

Data were transformed using square root transform to fit normality and equality of variances as requirement to run parametric statistics. Comparisons of µTBS means are shown in Figure 2**.** The statistical analysis revealed that the coinitiator type was a significant factor (P < 0.001). The overall data for A_{PA} (44.1 ± 21.8 MPa) showed significantly higher bond strength than the A_{EDAB} (33.3 ± 16.7 MPa, P < 0.001) whereas A_{BDO} (39.4 ± 20.2 MPa) presented intermediate means being similar to both groups. It was found that significantly smaller bond strength was found in enamel when compared with dentin, (23.8 ± 9.3 < 54.1 ± 16.0 respectively) considering overall data grouped according to substrate (P < 0.001). The interaction between coinitiator and substrate was not significant.

The failure analysis (Table 2) showed a predominance of mixed failures for all groups in dentin (≥ 60 %). In enamel, adhesive and mixed failures were detected in similar proportions. None cohesive failures within the adhesive resin were detected irrespective of the coinitiator type.

3.2 Morphological Analysis of the Bonding Interfaces

Figure 3 shows SEM pictures of the bonding interfaces for all groups. The hybrid layer for all groups was shown to be shallow, less than approximately 2 µm thick. No appreciable differences in homogeneity and continuity of the hybrid layer were detected along the bonding interface between the materials. The resin tags observed for the A_{EDAB} group were cylindrical with a smooth, clean surface. For the A_{BDO} and A_{PA} groups, the resin tags detected were trunk-conical-shaped, with the larger base of the cone facing up to the hybrid layer. In addition, the surface of the resin tags for the A_{BDO} and A_{PA} groups was rougher than that for the A_{EDAB} group. For the A_{PA} group, the surface of the tags was even slightly rougher compared with that of the BDO system, with the presence of accessory hybridized tubules.

4 Discussion

The two null hypothesis tested were rejected. Microtensile bond strength means were lower in enamel than dentin. The present study showed that group A_{PA} showed a statistically higher microtensile bond strength than the control group whereas A_{BDO} group was similar to both groups. Some studies have described the use of benzodioxole derivatives as co-initiators for resin-based materials. [11,12,17] Whereas, up to this time, the present study is first to report the use of these co-initiators in adhesive systems. The replacement of amine, ethyl 4-dimethylamino benzoate (EDAB) by natural alternatives such as benzodioxole derivates in dental adhesives is a feasible rationale, since they decrease the risk of toxicity during manufacture, generation of toxic residues and environmental impact. Furthermore, the gradual substitution of dental adhesive components by natural derivates certainly will decrease cytotoxicity and promote advances in the biocompatibility of biomaterials in dentistry.

A coinitiator is necessary for effective photopolymerization using CQ-based systems,[21,22] because when exposed to light, CQ has a limited lifetime to react and start the radical polymerization.[4] The photoinitiation reaction of this binary system involves hydrogen abstraction from the excited sensitizer to the coinitiator. [21] The benzodioxole derivatives were shown to be efficient alternative co-initiators.

Unlike amines, the reaction mechanism of PA and BDO involves hydrogen abstraction through the cyclic acetal radicals generated by the photoirradiation onto cyclic acetal compounds, which might be rearranged rapidly by ß-scission to the corresponding ester radicals. For PA, the hydrogen abstraction from the methylene-bridge carbon could also occur to form a methylenedioxybenzene radical and initiate the polymerization with the methacrylate monomers.[11]

According to the statistical analysis no significant differences were detected between the benzodioxoles derivatives components. This might be explained by the fact that the molecular structure of BDO and PA are very similar, as shown in Figure 1 and may lead to similar effects. The bond strength of A_{PA} was significantly higher when compared with that of A_{EDAB}. The hybridization profile verified for A_{PA} also suggests good interaction of this material with the dentin substrate. These features are probably related to the higher hydrophilicity of PA due to the presence of the methanol group. The hydrophilicity increases the solubility of the adhesive resin with coinitiator and facilitates the hybridization to dentin, substrate which has natural-moisture content.

The hybridization features showed in SEM micrographs suggest that in the more hybridized tubules, benzodioxoles derivatives co-initiators were found, when compared with the amine. To achieve optimal dentin bond strength, the adhesive must penetrate the demineralized dentin tubules entrance and branches before polymerization. Although the actual contribution of hybridized resins tags to bond strengths is not sufficiently explained, [23] A_{EDAB} showed more adhesive failures in comparison with the natural co-initiators. This outcome provides additional evidence that the use of benzodioxoles results in better interaction with the dentin substrate.

It is known that immediate bonding to enamel is not affected by the hydrophilicity of the bonding system. [24] The lower bond strength values to enamel than dentin are explained by the demineralization capacity of the phosphoric acid monomers present in the self-etching primer, and consequently the superficial interaction with enamel and thus less potential for micro-mechanical interlocking.[25] Nowadays, low bond strength has been a concern when the self-etching primers of adhesives systems available on market were applied to enamel.[26,27] The microtensile bond strength means and hybridization mode features found in the present study indicate that benzodioxole derivatives are effective and promising alternatives to amines as co-initiators in dental adhesive systems. However, the long-term performance of bonding systems with benzodioxoles as co-initiators warrants investigation, since the more hydrophilic character of these systems may influence the longevity of the bonds.

5 Conclusions

The morphology of the bonding interface associated with the adhesive performance revealed that 1,3-benzodioxole and piperonyl alcohol are effective and promising alternatives to amine as co-initiators for the radical photopolymerization of camphorquinone-based initiating system in dental adhesives formulations.

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Referências

1. Jakubiak J, Allonas X, Fouassier JP, Sionkowska A, Andrzejewska E, Linden LA, et al. Camphorquinone-amines photoinitating systems for the initiation of free radical polymerization. *Polymer* 2003;44**:**5219-5226.

2. Ogunyinka A, Palin WM, Shortall AC, Marquis PM. Photoinitiation chemistry affects light transmission and degree of conversion of curing experimental dental resin composites. *Dent Mater* 2007;23**:**807-13.

3. Schroeder WF, Cook WD, Vallo CI. Photopolymerization of N,N-dimethylaminobenzyl alcohol as amine co-initiator for light-cured dental resins. *Dent Mater* 2008;24**:**686-93.

4. Chen YC, Ferracane JL, Prahl SA. Quantum yield of conversion of the photoinitiator camphorquinone. *Dent Mater* 2007;23**:**655-64.

5. Musanje L, Ferracane JL, Sakaguchi RL. Determination of the optimal photoinitiator concentration in dental composites based on essential material properties. *Dent Mater* 2009.

6. Brennan RJ, Schiestl RH. The aromatic amine carcinogens o-toluidine and o-anisidine induce free radicals and intrachromosomal recombination in Saccharomyces cerevisiae. *Mutat Res* 1999;430**:**37-45.

7. Weisburger EK, Russfield AB, Homburger F, Weisburger JH, Boger E, Van Dongen CG, et al. Testing of twenty-one environmental aromatic amines or derivatives for long-term toxicity or carcinogenicity. *J Environ Pathol Toxicol* 1978;2**:**325-56.

8. Wang K, Yang D, Xiao M, Chen X, Lu F, Nie J. Sesamin as a co-initiator for unfilled dental restorations. *Acta Biomater* 2009;in press**:**in press.

9. Park YJ, Chae KH, Rawls HR. Development of a new photoinitiation system for dental light-cure composite resins. *Dent Mater* 1999;15**:**120-7.

10. Shi S, Nie J. Investigation of 3,4-methylenedioxybenzene methoxyl methacrylate as coinitiator and comonomer for dental application. *J Biomed Mater Res B Appl Biomater* 2007;82**:**487-93.

11. Liu S, Shi S, Hou G, Nie J. Benzodioxole derivative as coinitiator for dental resin. *Acta Odontol Scand* 2007;65**:**313-8.

12. Shi S, Nie J. A natural component as coinitiator for unfilled dental resin composites. *J Biomed Mater Res B Appl Biomater* 2007;82**:**44-50.

13. Hickey MJ. Investigation of the chemical constituents of Brazilian sassafras oil. *J Org Chem* 1948;13**:**443-6.

14. Micale N, Zappala M, Grasso S. Synthesis and antitumor activity of 1,3-benzodioxole derivatives. *Farmaco* 2002;57**:**853-9.

15. Micale N, Zappala M, Grasso S. Synthesis and cytotoxic activity of 1,3-benzodioxole derivatives. Note II. *Farmaco* 2003;58**:**351-5.

16. Regnier GH, Canevari RV, Laubie MV, Poignant JB, Regnier GH, Canevari RV, Laubie MV, Poignant JBRegnier GH, Canevari RV, Laubie MV, Poignant JBs; Science-Union Et Cie, France, assignee. Benzodioxoles Compounds. France patent 3,917,597. 1975 Nov. 4, 1975.

17. Shi SG, H.; Wu, G.; Nie, J. Cyclic acetal as coinitiator for biomolecular photoinitiating systems. *Polymer* 2007**:**2860-2865.

18. Lima G da S, Ogliari FA, da Silva EO, Ely C, Demarco FF, Carreno NL, et al. Influence of water concentration in an experimental self-etching primer on the bond strength to dentin. *J Adhes Dent* 2008;10**:**167-72.

19. Ogliari FA, de Sordi ML, Ceschi MA, Petzhold CL, Demarco FF, Piva E. 2,3-Epithiopropyl methacrylate as functionalized monomer in a dental adhesive. *J Dent* 2006;34**:**472-7.

20. Shinohara MS, Yamauti M, Inoue G, Nikaido T, Tagami J, Giannini M, et al. Evaluation of antibacterial and fluoride-releasing adhesive system on dentin--microtensile bond strength and acid-base challenge. *Dent Mater J* 2006;25**:**545-52.

21. Andrzejewska E. Photopolymerization kinetics of multifunctional monomers. *Progress in Polymer Science* 2001;26**:**605-665.

22. Ogliari FA, Ely C, Petzhold CL, Demarco FF, Piva E. Onium salt improves the polymerization kinetics in an experimental dental adhesive resin. *J Dent* 2007;35**:**583-7.

23. Prati C, Chersoni S, Mongiorgi R, Pashley DH. Resin-infiltrated dentin layer formation of new bonding systems. *Oper Dent* 1998;23**:**185-94.

24. Hara AT, Amaral CM, Pimenta LA, Sinhoreti MA. Shear bond strength of hydrophilic adhesive systems to enamel. *Am J Dent* 1999;12**:**181-4.

25. De Munck J, Van Landuyt K, Peumans M, Poitevin A, Lambrechts P, Braem M, et al. A critical review of the durability of adhesion to tooth tissue: methods and results. *J Dent Res* 2005;84**:**118-32.

26. Inoue S, Van Meerbeek B, Abe Y, Yoshida Y, Lambrechts P, Vanherle G, et al. Effect of remaining dentin thickness and the use of conditioner on micro-tensile bond strength of a glass-ionomer adhesive. *Dent Mater* 2001;17**:**445-55.

27. De Munck J, Van Meerbeek B, Satoshi I, Vargas M, Yoshida Y, Armstrong S, et al. Microtensile bond strengths of one- and two-step self-etch adhesives to bur-cut enamel and dentin. *Am J Dent* 2003;16**:**414-20.

28. Peumans M, Van Meerbeek B, Lambrechts P, Vanherle G. Two-year clinical effectiveness of a resin-modified glass-ionomer adhesive. *Am J Dent* 2003;16**:**363-8.

29. Turkun SL. Clinical evaluation of a self-etching and a one-bottle adhesive system at two years. *J Dent* 2003;31**:**527-34.

Figure Legends

Fig 1. Molecular structure of the co-initiators used in the present study.

Fig 2. Means in MPa (± standard deviations) for microtensile bond strength (μTBS) test in enamel and dentin.

Fig 3. SEM pictures of the bonding interfaces: A_{EDAB} (A,B), A_{BDO} (C,D), A_{PA} (E,F). The hybrid layer (delimited between arrows) for all groups was shallow (less than 2 μm thick). No appreciable differences in homogeneity and continuity were detected along the bonding interface. The resin tags observed for A_{EDAB} were cylindrical with a smooth, clean surface. For A_{BDO} and A_{PA}, the resin tags were trunk-conical. The surface of the tags for A_{BDO} and A_{PA} was rougher than for A_{EDAB}. For A_{PA}, the surface of the tags was slightly rougher compared with A_{BDO}, with the presence of accessory hybridized tubules (pointers).

Tables

Table 1. Composition of the binary photoinitiating systems used inthe experimental adhesive resins groups.

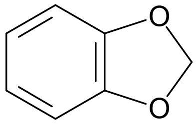
Composition (mol%)	Groups		
	A_{BDO}	A_{PA}	A_{EDAB}
Camphorquinone	1	1	1
1,3-benzodioxole (BDO)	4	-	-
Piperonyl alcohol (PA)	-	4	-
Ethyl 4-dimethylamino benzoate (EDAB)	-	-	1

Table 2. Failure analysis, according to tested groups.

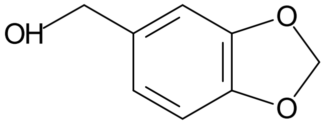
	Failure modes *									
Adhesive resins	Enamel					Dentin				
	CS	CA	AF	M	Total	CS	CA	AF	M	Total
A_{PA}	-	-	50 %	50 %	100 %	5 %	-	15 %	80 %	100 %
A_{BDO}	-	-	45 %	55 %	100 %	5 %	-	25 %	70 %	100 %
A_{EDAB}	-	-	40 %	60 %	100 %	10 %	-	30 %	60 %	100 %

*CS = cohesive within substrate; CA = cohesive within adhesive resin; AF = adhesive failure; M = mixed.

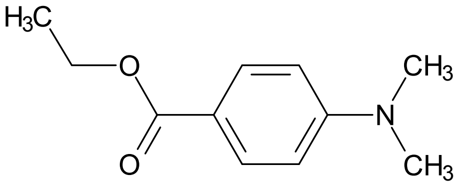
Figures



1,3-benzodioxole (BDO)

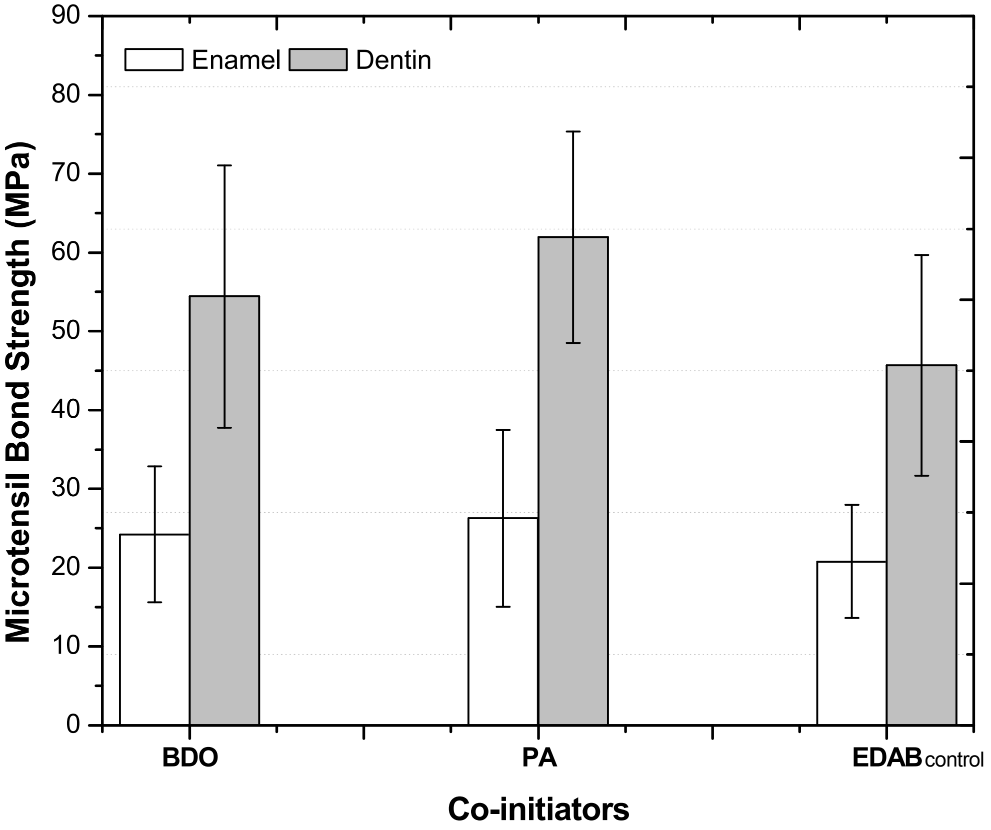


Piperonyl alcohol (PA)



Ethyl 4-dimethylamine benzoate (EDAB)

Figure 1



Enamel
Dentin
Microtensil Bond Strength (MPa)
0
10
20
30
40
50
60
70
80
90
BDO
PA
EDAB control
Co-initiators

Figure 2

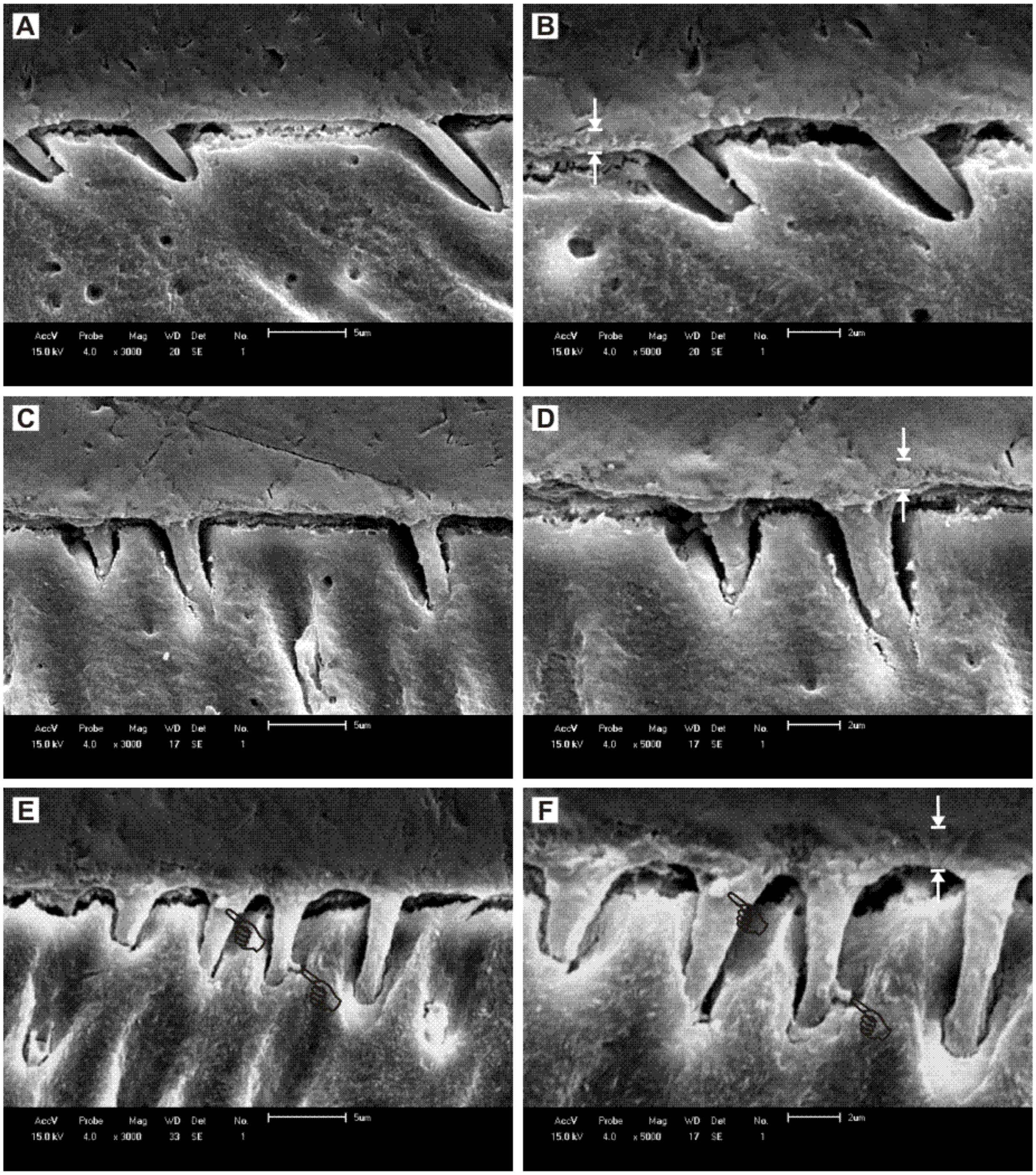

A
AccV Probe Mag WD Det No. 5um
15.0 kV 4.0 x 3000 20 SE 1
B
AccV Probe Mag WD Det No. 2um
15.0 kV 4.0 x 5000 20 SE 1
C
AccV Probe Mag WD Det No. 5um
15.0 kV 4.0 x 3000 17 SE 1
D
AccV Probe Mag WD Det No. 2um
15.0 kV 4.0 x 5000 17 SE 1
E
AccV Probe Mag WD Det No. 5um
15.0 kV 4.0 x 3000 33 SE 1
F
AccV Probe Mag WD Det No. 2um
15.0 kV 4.0 x 5000 17 SE 1

Figure 3

4 CONCLUSÕES

Utilizando os sistemas de fotoiniciação compostos por canforoquinona como fotoiniciador e derivados de benzodioxolas como co-iniciadoras da polimerização radicalar ativada por lâmpada halógena, foram obtidas formulações de adesivos autocondicionates experimentais viáveis.

O desempenho dos sistemas de fotoiniciação compostos por derivados de benzodioxolas mostrou-se efetivo apresentando características e propriedades físico-químicas, mecânicas e adesivas semelhantes àquelas obtidas quando empregada amina como co-iniciadora da polimerização nos adesivos experimentais avaliados.

Benzodioxolas são co-iniciadores alternativos a serem consideradas para aplicação em formulações de sistemas adesivos odontológicos.

5 REFERÊNCIAS

ALBRECHT, W. N.; STEPHENSON, R. L. Health hazards of tertiary amine catalysts **Scand J Work Environ Health**, v.14, n.4, p.209-219, 1988.

AMES, B. N. Dietary carcinogens and anticarcinogens. Oxygen radicals and degenerative diseases **Science**, v.221, n.4617, p.1256-1264, 1983.

ANDRZEJEWSKA, E. Photopolymerization kinetics of multifunctional monomers **Progress in Polymer Science**, v.26, n.4, p.605-665, 2001.

BRENNAN, R. J.; SCHIESTL, R. H. The aromatic amine carcinogens o-toluidine and o-anisidine induce free radicals and intrachromosomal recombination in Saccharomyces cerevisiae **Mutat Res**, v.430, n.1, p.37-45, 1999.

CHEN, Y. C.; FERRACANE, J. L.; PRAHL, S. A. Quantum yield of conversion of the photoinitiator camphorquinone **Dent Mater**, v.23, n.6, p.655-664, 2007.

CONDE, M. C.; ZANCHI, C. H.; RODRIGUES-JUNIOR, S. A.; CARRENO, N. L.; OGLIARI, F. A.; PIVA, E. Nanofiller loading level: Influence on selected properties of an adhesive resin **J Dent**, v.37, n.5, p.331-335, 2009.

COOK, W. D. Photopolymerization kinetics of dimethacrylates using the camphorquinone amine initiator system **Polymer**, n.33, p.600-609, 1992.

COOK, W. D. THERMAL Aspects of the kinetics of dimethacrylate photopolymerization. **Polymer**, v.33, n.10, p.2152-2161, 1992.

COX, C. F.; HAFEZ, A. A.; AKIMOTO, N.; OTSUKI, M.; SUZUKI, S.; TARIM, B. Biocompatibility of primer, adhesive and resin composite systems on non-exposed and exposed pulps of non-human primate teeth **Am J Dent**, v.11 Spec No, p.S55-63, 1998.

DA ROSA RODOLPHO, P. A.; CENCI, M. S.; DONASSOLLO, T. A.; LOGUERCIO, A. D.; DEMARCO, F. F. A clinical evaluation of posterior composite restorations: 17-year findings **J Dent**, v.34, n.7, p.427-435, 2006.

DE MUNCK, J.; VAN LANDUYT, K.; PEUMANS, M.; POITEVIN, A.; LAMBRECHTS, P.; BRAEM, M.; VAN MEERBEEK, B. A critical review of the durability of adhesion to tooth tissue: methods and results **J Dent Res**, v.84, n.2, p.118-132, 2005.

DE MUNCK, J.; VAN MEERBEEK, B.; SATOSHI, I.; VARGAS, M.; YOSHIDA, Y.; ARMSTRONG, S.; LAMBRECHTS, P.; VANHERLE, G. Microtensile bond strengths of one- and two-step self-etch adhesives to bur-cut enamel and dentin **Am J Dent**, v.16, n.6, p.414-420, 2003.

FARIA-E-SILVA, A. L.; CASSELLI, D. S.; LIMA, G. S.; OGLIARI, F. A.; PIVA, E.; MARTINS, L. R. Kinetics of conversion of two dual-cured adhesive systems **J Endod**, v.34, n.9, p.1115-1118, 2008.

GAENGLER, P.; HOYER, I.; MONTAG, R.; GAEBLER, P. Micromorphological evaluation of posterior composite restorations - a 10-year report **J Oral Rehabil**, v.31, n.10, p.991-1000, 2004.

GOLDBERG, M. In vitro and in vivo studies on the toxicity of dental resin components: a review **Clin Oral Investig**, v.12, n.1, p.1-8, 2008.

GUO, X.; SPENCER, P.; WANG, Y.; YE, Q.; YAO, X.; WILLIAMS, K. Effects of a solubility enhancer on penetration of hydrophobic component in model adhesives into wet demineralized dentin **Dent Mater**, v.23, n.12, p.1473-1481, 2007.

GUO, X.; WANG, Y.; SPENCER, P.; YE, Q.; YAO, X. Effects of water content and initiator composition on photopolymerization of a model Bis-GMA/HEMA resin **Dent Mater**, v.24, n.6, p.824-831, 2008.

HARA, A. T.; AMARAL, C. M.; PIMENTA, L. A.; SINHORETI, M. A. Shear bond strength of hydrophilic adhesive systems to enamel **Am J Dent**, v.12, n.4, p.181-184, 1999.

HICKEY, M. J. Investigation of the chemical constituents of Brazilian sassafras oil **J Org Chem**, v.13, n.3, p.443-446, 1948.

HIGASHI, C.; MICHEL, M. D.; REIS, A.; LOGUERCIO, A. D.; GOMES, O. M.; GOMES, J. C. Impact of adhesive application and moisture on the mechanical properties of the adhesive interface determined by the nano-indentation technique **Oper Dent**, v.34, n.1, p.51-57, 2009.

INOUE, S.; VAN MEERBEEK, B.; ABE, Y.; YOSHIDA, Y.; LAMBRECHTS, P.; VANHERLE, G.; SANO, H. Effect of remaining dentin thickness and the use of conditioner on micro-tensile bond strength of a glass-ionomer adhesive **Dent Mater**, v.17, n.5, p.445-455, 2001.

JAKUBIAK, J.; ALLONAS, X.; FOUASSIER, J. P.; SIONKOWSKA, A.; ANDRZEJEWSKA, E.; LINDEN, L. A.; RABEK, J. F. Camphorquinone-amines photoinitating systems for the initiation of free radical polymerization **Polymer**, v.44, n.18, p.5219-5226, 2003.

JOSEPHY, P. D.; EVANS, D. H.; PARIKH, A.; GUENGERICH, F. P. Metabolic activation of aromatic amine mutagens by simultaneous expression of human cytochrome P450

1A2, NADPH-cytochrome P450 reductase, and N-acetyltransferase in Escherichia coli **Chem Res Toxicol**, v.11, n.1, p.70-74, 1998.

JOSHI, R.; KUMAR, M. S.; SATYAMOORTHY, K.; UNNIKRISNAN, M. K.; MUKHERJEE, T. Free radical reactions and antioxidant activities of sesamol: pulse radiolytic and biochemical studies **J Agric Food Chem**, v.53, n.7, p.2696-2703, 2005.

KUMAGAI, Y.; LIN, L. Y.; SCHMITZ, D. A.; CHO, A. K. Hydroxyl radical mediated demethylenation of (methylenedioxy)phenyl compounds **Chem Res Toxicol**, v.4, n.3, p.330-334, 1991.

LIMA G. S.; OGLIARI, F. A.; DA SILVA, E. O.; ELY, C.; DEMARCO, F. F.; CARRENO, N. L.; PETZHOLD, C. L.; PIVA, E. Influence of water concentration in an experimental self-etching primer on the bond strength to dentin **J Adhes Dent**, v.10, n.3, p.167-172, 2008.

LISO, P. A.; VAZQUEZ, B.; REBUELTA, M.; HERNAEZ, M. L.; ROTGER, R.; SAN ROMAN, J. Analysis of the leaching and toxicity of new amine activators for the curing of acrylic bone cements and composites **Biomaterials**, v.18, n.1, p.15-20, 1997.

LIU, S.; SHI, S.; HOU, G.; NIE, J. Benzodioxole derivative as coinitiator for dental resin **Acta Odontol Scand**, v.65, n.6, p.313-318, 2007.

MATOS, M. A.; MONTE, M. J.; SOUSA, C. C.; ALMEIDA, A. R.; MORAIS, V. M. Thermodynamic study of sesamol, piperonyl alcohol, piperonylic acid and homopiperonylic acid: a combined experimental and theoretical investigation **Org Biomol Chem**, v.2, n.6, p.908-914, 2004.

MICALE, N.; ZAPPALA, M.; GRASSO, S. Synthesis and antitumor activity of 1,3-benzodioxole derivatives **Farmaco**, v.57, n.10, p.853-859, 2002.

MICALE, N.; ZAPPALA, M.; GRASSO, S. Synthesis and cytotoxic activity of 1,3-benzodioxole derivatives. Note II **Farmaco**, v.58, n.5, p.351-355, 2003.

MOREIRA, D. R. M.; LEITE, A. C. L.; FERREIRA, P. M. P.; COSTA, P. M.; LOTUFO, L. V. C.; MORAES, M. O.; BRONDANI, D. J.; PESSOA, C. O. Synthesis and antitumor evaluating of peptidyl-like derivatives containing the 1,3-benzodioxole system. **Eur J Med Chem**, v. 42, p.351-357, 2007.

MUSANJE, L.; FERRACANE, J. L.; SAKAGUCHI, R. L. Determination of the optimal photoinitiator concentration in dental composites based on essential material properties **Dent Mater**, 2009.

NEUMANN, M. G.; SCHMITT, C. C.; FERREIRA, G. C.; CORREA, I. C. The initiating radical yields and the efficiency of polymerization for various dental photoinitiators excited by different light curing units **Dent Mater**, v.22, n.6, p.576-584, 2006.

OGLIARI, F. A.; DE SORDI, M. L.; CESCHI, M. A.; PETZHOLD, C. L.; DEMARCO, F. F.; PIVA, E. 2,3-Epithiopropyl methacrylate as functionalized monomer in a dental adhesive **J Dent**, v.34, n.7, p.472-477, 2006.

OGLIARI, F. A.; ELY, C.; LIMA, G. S.; CONDE, M. C.; PETZHOLD, C. L.; DEMARCO, F. F.; PIVA, E. Onium salt reduces the inhibitory polymerization effect from an organic solvent in a model dental adhesive resin **J Biomed Mater Res B Appl Biomater**, v.86, n.1, p.113-118, 2008.

OGLIARI, F. A.; ELY, C.; PETZHOLD, C. L.; DEMARCO, F. F.; PIVA, E. Onium salt improves the polymerization kinetics in an experimental dental adhesive resin **J Dent**, v.35, n.7, p.583-587, 2007.

OGLIARI, F. A.; ELY, C.; ZANCHI, C. H.; FORTES, C. B.; SAMUEL, S. M.; DEMARCO, F. F.; PETZHOLD, C. L.; PIVA, E. Influence of chain extender length of aromatic dimethacrylates on polymer network development **Dent Mater**, v.24, n.2, p.165-171, 2008.

OGUNYINKA, A.; PALIN, W. M.; SHORTALL, A. C.; MARQUIS, P. M. Photoinitiation chemistry affects light transmission and degree of conversion of curing experimental dental resin composites **Dent Mater**, v.23, n.7, p.807-813, 2007.

OPDAM, N. J.; LOOMANS, B. A.; ROETERS, F. J.; BRONKHORST, E. M. Five-year clinical performance of posterior resin composite restorations placed by dental students **J Dent**, v.32, n.5, p.379-383, 2004.

PARK, Y. J.; CHAE, K. H.; RAWLS, H. R. Development of a new photoinitiation system for dental light-cure composite resins **Dent Mater**, v.15, n.2, p.120-127, 1999.

PELKA, M.; DANZL, C.; DISTLER, W.; PETSCHELT, A. A new screening test for toxicity testing of dental materials **J Dent**, v.28, n.5, p.341-345, 2000.

PEUMANS, M.; DE MUNCK, J.; VAN LANDUYT, K.; LAMBRECHTS, P.; VAN MEERBEEK, B. Five-year clinical effectiveness of a two-step self-etching adhesive **J Adhes Dent**, v.9, n.1, p.7-10, 2007.

PEUMANS, M.; VAN MEERBEEK, B.: LAMBRECHTS, P.; VANHERLE, G. Two-year clinical effectiveness of a resin-modified glass-ionomer adhesive. **Am J Dent,** n.16, p.363-8, 2003.

PRATI, C.; CHERSONI, S.; MONGIORGI, R.; PASHLEY, D. H. Resin-infiltrated dentin layer formation of new bonding systems **Oper Dent**, v.23, n.4, p.185-194, 1998.

REGNIER, G.H.; CANEVARI, R.V.; LAUBIE, M.V.; POIGNANT, J.B. Benzodioxoles Compounds. **United States Patent,** vol. 3917597. France: Science-Union Et Cie, France, 1975.

SCHNEIDER, L. F.; CONSANI, S.; SAKAGUCHI, R. L.; FERRACANE, J. L. Alternative photoinitiator system reduces the rate of stress development without compromising the final properties of the dental composite **Dent Mater**, v.25, n.5, p.566-72, 2008.

SCHNEIDER, L. F.; PFEIFER, C. S.; CONSANI, S.; PRAHL, S. A.; FERRACANE, J. L. Influence of photoinitiator type on the rate of polymerization, degree of conversion, hardness and yellowing of dental resin composites **Dent Mater**, v.24, n.9, p.1169-1177, 2008.

SCHROEDER, W. F.; COOK, W. D.; VALLO, C. I. Photopolymerization of N,N-dimethylaminobenzyl alcohol as amine co-initiator for light-cured dental resins **Dent Mater**, v.24, n.5, p.686-693, 2008.

SCHROEDER, W. F.; VALLO, C. I. Effect of different photoinitiator systems on conversion profiles of a model unfilled light-cured resin **Dent Mater**, v.23, n.10, p.1313-1321, 2007.

SCHWEIKL, H.; HILLER, K. A.; ECKHARDT, A.; BOLAY, C.; SPAGNUOLO, G.; STEMPFL, T.; SCHMALZ, G. Differential gene expression involved in oxidative stress response caused by triethylene glycol dimethacrylate **Biomaterials**, v.29, n.10, p.1377-1387, 2008.

SHI, S. G., H.; WU, G.; NIE, J. Cyclic acetal as coinitiator for biomolecular photoinitiating systems **Polymer**, n.48, p.2860-2865, 2007.

SHI, S.; NIE, J. A natural component as coinitiator for unfilled dental resin composites **J Biomed Mater Res B Appl Biomater**, v.82, n.1, p.44-50, 2007.

SHI, S.; NIE, J. Investigation of 3,4-methylenedioxybenzene methoxyl methacrylate as coinitiator and comonomer for dental application **J Biomed Mater Res B Appl Biomater**, v.82, n.2, p.487-493, 2007.

SHIN, D. H.; RAWLS, H. R. Degree of conversion and color stability of the light curing resin with new photoinitiator systems **Dent Mater**, 2009.

SHINOHARA, M. S.; YAMAUTI, M.; INOUE, G.; NIKAIDO, T.; TAGAMI, J.; GIANNINI, M.; DE GOES, M. F. Evaluation of antibacterial and fluoride-releasing adhesive system on dentin--microtensile bond strength and acid-base challenge **Dent Mater J**, v.25, n.3, p.545-552, 2006.

Site do Instituto Brasileiro de Plantas Medicinais (IBPM) < http://www.ibpm.org.br > acesso em 26 abril, 2009.

TAGASHIRA, M.; OHTAKE,Y. A new antioxidant 1.3-benzodioxole from Melissa officinalis. **Planta Med,** v. 64, p.555-558, 1998.

TAGASHIRA, M.; UCHIYAMA, K.; YOSHIMURA, T.; SHIROTA, M.; UEMITSU, N. Inhibition by hop bract polyphenols of cellular adherence and water-insoluble glucan synthesis of mutans streptococci **Biosci Biotechnol Biochem**, v.61, n.2, p.332-335, 1997.

TEIXEIRA, H. M.; DO NASCIMENTO, A. B.; HEBLING, J.; DE SOUZA COSTA, C. A. In vivo evaluation of the biocompatibility of three current bonding agents **J Oral Rehabil**, v.33, n.7, p.542-550, 2006.

TOLEDANO, M.; OSORIO, R.; OSORIO, E.; AGUILERA, F. S.; YAMAUTI, M.; PASHLEY, D. H.; TAY, F. Durability of resin-dentin bonds: effects of direct/indirect exposure and storage media **Dent Mater**, v.23, n.7, p.885-892, 2007.

TSENG, T. H.; TSHENG, Y. M.; LEE, Y. J. Cytotoxicity effects of di- and tri-hydroxybenzaldehydes as a chemopreventive potential agent on tumor cells **Toxicology**, v.161, n.3, p.179-187, 2001.

TURKUN, S.L. Clinical evaluation of a self-etching and a one-bottle adhesive system at two years. **J Dent**, n.31, p.527-34, 2003.

VAN DIJKEN, J. W. Durability of three simplified adhesive systems in Class V non-carious cervical dentin lesions **Am J Dent**, v.17, n.1, p.27-32, 2004.

WANG, K.; YANG, D.; XIAO, M.; CHEN, X.; LU, F.; NIE, J. Sesamin as a co-initiator for unfilled dental restorations **Acta Biomater**, v.in press, n.in press, p.in press, 2009.

WEISBURGER, E. K.; RUSSFIELD, A. B.; HOMBURGER, F.; WEISBURGER, J. H.; BOGER, E.; VAN DONGEN, C. G.; CHU, K. C. Testing of twenty-one environmental aromatic amines or derivatives for long-term toxicity or carcinogenicity **J Environ Pathol Toxicol**, v.2, n.2, p.325-356, 1978.

YOSHIDA, K.; GREENER, E. H. Effect of photoinitiator on degree of conversion of unfilled light-cured resin **J Dent**, v.22, n.5, p.296-299, 1994.

YOSHIDA, K.; GREENER, E. H. Effects of two amine reducing agents on the degree of conversion and physical properties of an unfilled light-cured resin **Dent Mater**, v.9, n.4, p.246-251, 1993.

APÊNDICES

Apêndice A

Relatórios de Ensaios - Resistência de união à microtração

UFPEL - Treinamento Evandro

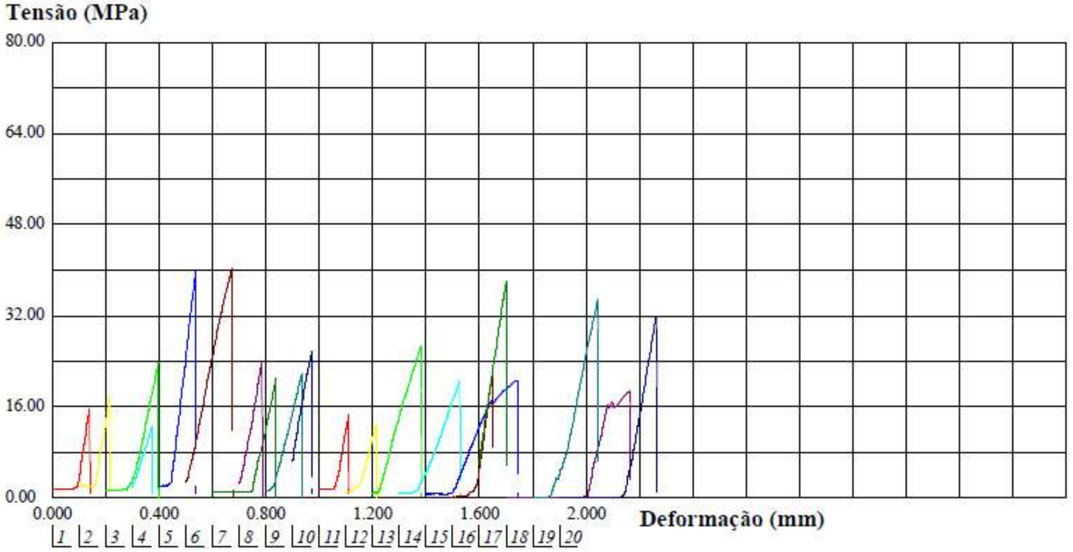
Relatório de Ensaio

Máquina: **Emic DL2000** Célula: **Trd 19** Extensômetro: - Data: **30/05/2009** Hora: **10:02:10** Trabalho n° **1685**

Programa: **Tesc versão 3.01** Método de Ensaio: **UFPEL Microtração Script_EP_Celula_100N**

Ident. Amostra: >>> Tipo: **Giana B4%**

Corpo de Prova	Largura (mm)	Espessura (mm)	Área (mm2)	Força Máxima (kgf)	Tensão Máxima (MPa)
CP 1	0.90	0.96	0.86	1.37	15.60
CP 2	0.75	0.84	0.63	1.15	17.94
CP 3	0.94	1.05	0.99	2.40	23.89
CP 4	0.84	1.62	1.36	1.76	12.71
CP 5	0.56	1.17	0.66	2.66	39.82
CP 6	0.95	0.97	0.92	3.79	40.30
CP 7	0.83	1.33	1.10	2.37	21.02
CP 8	0.76	0.99	0.75	1.84	24.00
CP 9	0.95	1.11	1.05	2.36	21.92
CP 10	0.87	1.05	0.91	2.41	25.92
CP 11	0.87	0.98	0.85	1.27	14.64
CP 12	1.05	1.42	1.49	1.97	12.95
CP 13	0.82	1.86	1.53	4.16	26.76
CP 14	0.78	1.92	1.50	3.14	20.55
CP 15	0.95	1.95	1.85	3.91	20.68
CP 16	0.73	0.96	0.70	1.60	22.39
CP 17	0.79	0.93	0.73	2.86	38.14
CP 18	0.80	1.08	0.86	1.66	18.80
CP 19	1.03	1.01	1.04	3.71	34.96
CP 20	0.89	1.04	0.93	3.01	31.93
Número CPs	20	20	20	20	20
Média	0.8530	1.212	1.036	2.470	24.25
Mediana	0.8550	1.050	0.9236	2.385	22.16
Desv.Padrão	0.1134	0.3516	0.3376	0.9201	8.642
Coef.Var.(%)	13.29	29.01	32.58	37.25	35.64
Mínimo	0.5600	0.8400	0.6300	1.153	12.71
Máximo	1.050	1.950	1.853	4.162	40.30



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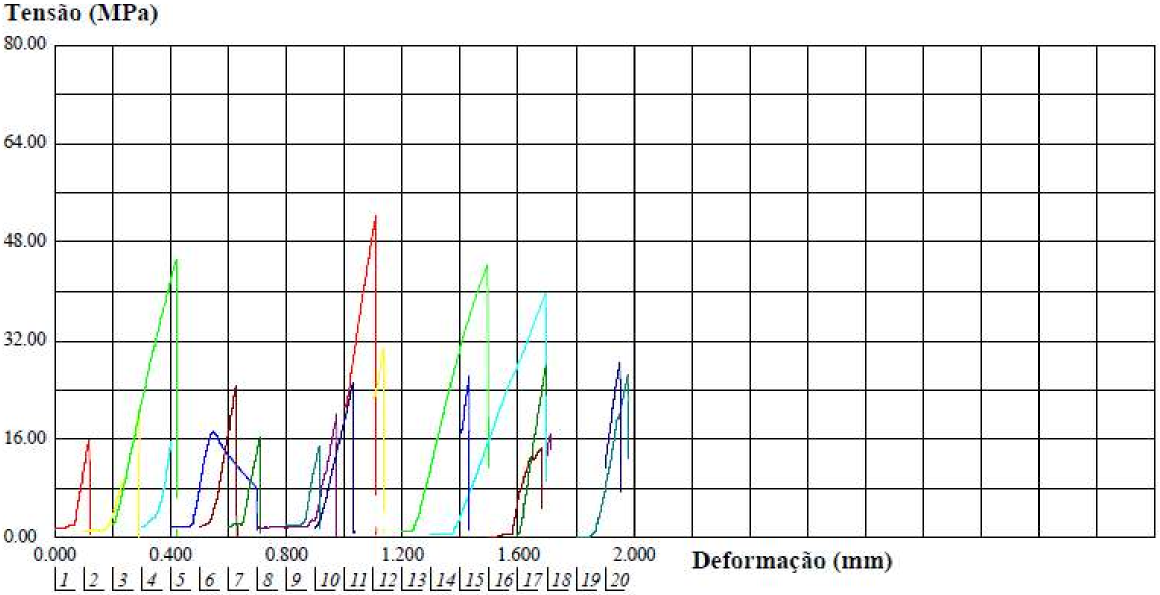
Relatório de Ensaio

Máquina: **Emic DL2000** Célula: **Trd 19** Extensômetro: - Data: **29/05/2009** Hora: **18:17:19** Trabalho nº **1684**

Programa: **Tesc versão 3.01** Método de Ensaio: **UFPEL Microtração Script_EP_Celula_100N**

Ident. Amostra: >> Tipo: **Giana AP 4%**

Corpo de Prova	Largura (mm)	Espessura (mm)	Área (mm2)	Força Máxima (kgf)	Tensão Máxima (MPa)
CP 1	0.84	1.10	0.92	1.50	15.88
CP 2	0.79	1.41	1.11	2.37	20.90
CP 3	1.00	1.11	1.11	5.12	45.27
CP 4	0.85	0.86	0.73	1.16	15.61
CP 5	0.85	0.86	0.73	1.29	17.31
CP 6	0.82	0.95	0.78	1.97	24.82
CP 7	0.89	0.95	0.85	1.42	16.42
CP 8	0.84	0.99	0.83	1.70	20.00
CP 9	0.73	0.86	0.63	0.95	14.86
CP 10	1.06	1.16	1.23	3.18	25.37
CP 11	0.80	1.07	0.86	4.56	52.29
CP 12	0.79	1.18	0.93	2.94	30.96
CP 13	0.87	1.49	1.30	5.86	44.31
CP 14	1.34	1.46	1.96	7.96	39.89
CP 15	0.89	0.92	0.82	2.20	26.34
CP 16	0.78	1.20	0.94	1.40	14.71
CP 17	1.09	0.81	0.88	2.53	28.11
CP 18	0.91	0.85	0.77	1.34	17.05
CP 19	0.91	0.85	0.77	2.09	26.56
CP 20	0.91	0.85	0.77	2.25	28.58
Número CPs	20	20	20	20	20
Média	0.8980	1.046	0.9461	2.691	26.26
Mediana	0.8600	0.9700	0.8508	2.147	25.10
Desv.Padrão	0.1382	0.2147	0.2940	1.832	11.24
Coef.Var.(%)	15.39	20.51	31.07	68.08	42.78
Mínimo	0.7300	0.8100	0.6278	0.9516	14.71
Máximo	1.340	1.490	1.956	7.957	52.29



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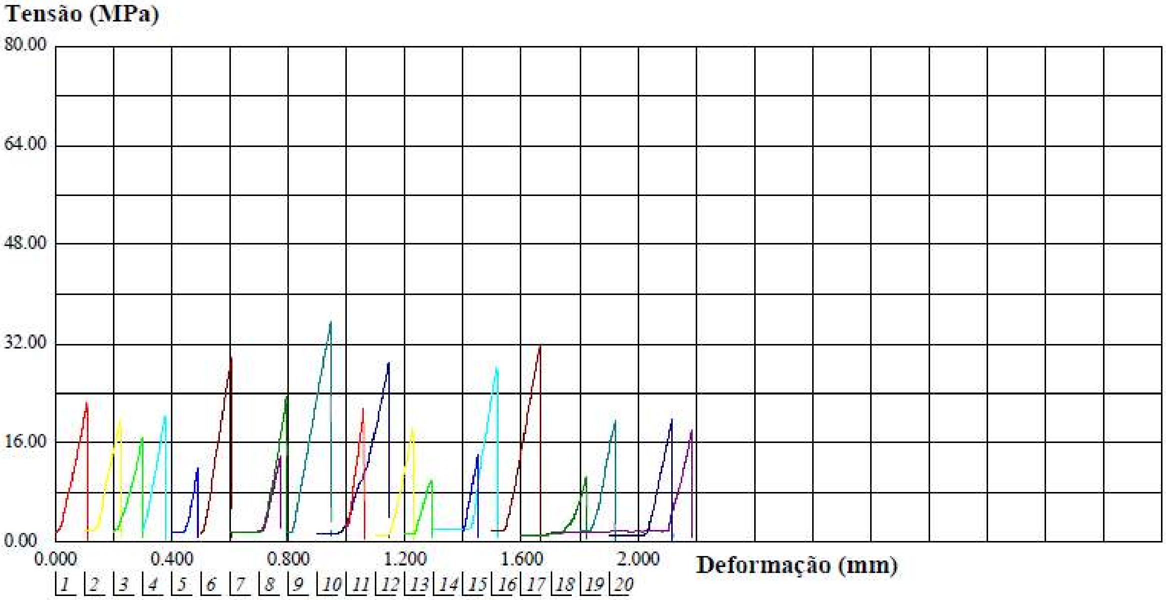
Relatório de Ensaio

Máquina: **Emic DL2000** Célula: **Trd 19** Extensômetro: - Data: **23/05/2009** Hora: **10:33:04** Trabalho nº **1665**
Programa: **Tesc versão 3.01** Método de Ensaio: **UFPEL Microtração Script_EP_Celula_100N**
Ident. Amostra: >>>Tipo: **EDAB2_esmalte_24h**

Corpo de Prova	Largura (mm)	Espessura (mm)	Área (mm2)	Força Máxima (kgf)	Tensão Máxima (MPa)
CP 1	0.88	0.92	0.81	1.86	22.50
CP 2	0.85	0.84	0.71	1.43	19.67
CP 3	0.95	0.95	0.90	1.55	16.86
CP 4	0.79	0.94	0.74	1.55	20.45
CP 5	0.93	0.86	0.80	0.98	12.03
CP 6	0.76	1.10	0.84	2.54	29.82
CP 7	0.93	0.82	0.76	1.84	23.67
CP 8	0.78	0.93	0.73	1.03	13.89
CP 9	0.79	1.00	0.79	2.87	35.68
CP 10	0.79	1.14	0.90	2.65	28.88
CP 11	0.87	0.79	0.69	1.51	21.56
CP 12	0.79	1.26	1.00	1.85	18.26
CP 13	0.74	1.24	0.92	0.94	10.06
CP 14	0.80	0.73	0.58	1.68	28.28
CP 15	0.88	1.01	0.89	1.28	14.11
CP 16	0.79	0.88	0.70	2.27	32.07
CP 17	0.87	1.16	1.01	1.09	10.55
CP 18	0.83	0.92	0.76	1.41	18.10
CP 19	0.80	0.96	0.77	1.53	19.59
CP 20	0.88	1.12	0.99	1.99	19.79
Número CPs	20	20	20	20	20
Média	0.8350	0.9785	0.8139	1.693	20.79
Mediana	0.8150	0.9450	0.7949	1.550	19.73
Desv.Padrão	0.06083	0.1487	0.1140	0.5543	7.203
Coef.Var.(%)	7.285	15.20	14.00	32.74	34.65
Mínimo	0.7400	0.7300	0.5840	0.9414	10.06
Máximo	0.9500	1.260	1.009	2.875	35.68

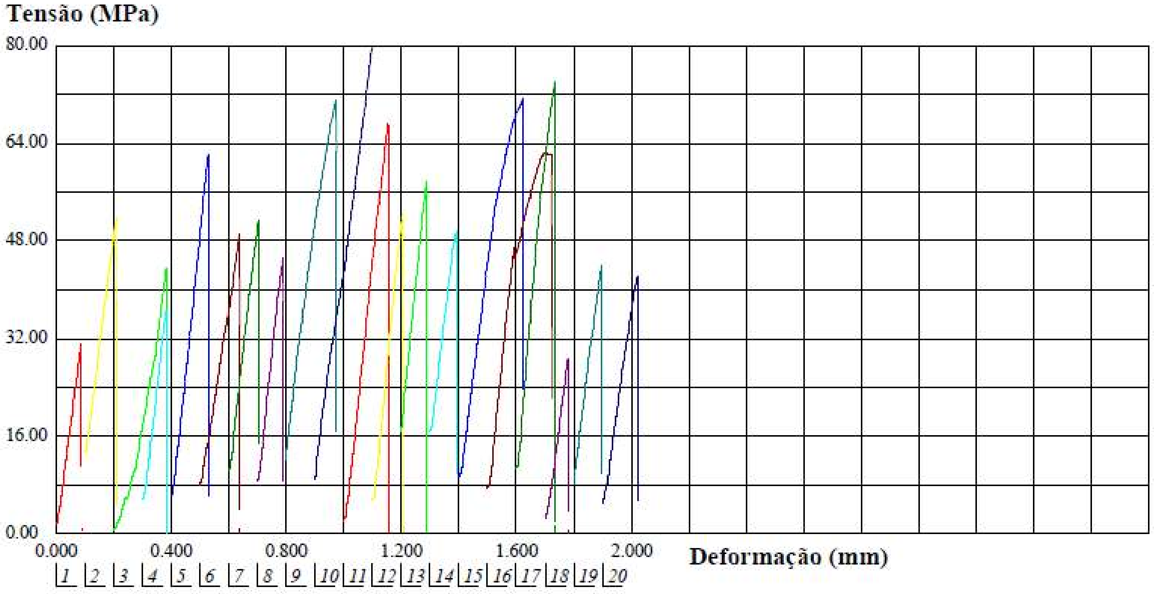


UFPEL - Treinamento Evandro

Relatório de Ensaio

Máquina: **Emic DL2000** Célula: **Trd 19** Extensômetro: - Data: **13/05/2009** Hora: **20:26:28** Trabalho nº **1641**
Programa: **Tesc versão 3.01** Método de Ensaio: **UFPEL Microtração Script_EP_Celula_100N**
Ident. Amostra: >>>Tipo: **B4%_24h**

Corpo de Prova	Largura (mm)	Espessura (mm)	Área (mm2)	Força Máxima (kgf)	Tensão Máxima (MPa)
CP 1	0.80	0.84	0.67	2.13	31.05
CP 2	0.81	0.85	0.69	3.63	51.77
CP 3	0.93	0.74	0.69	3.06	43.54
CP 4	0.89	0.86	0.77	2.87	36.73
CP 5	0.72	0.80	0.58	3.65	62.17
CP 6	0.80	0.81	0.65	3.25	49.24
CP 7	0.77	0.82	0.63	3.30	51.26
CP 8	0.76	0.80	0.61	2.80	45.22
CP 9	0.80	0.89	0.71	5.16	71.13
CP 10	0.90	1.02	0.92	9.23	98.55
CP 11	0.79	0.84	0.66	4.55	67.26
CP 12	0.78	0.79	0.62	3.30	52.50
CP 13	0.87	0.76	0.66	3.90	57.84
CP 14	0.82	0.79	0.65	3.27	49.52
CP 15	0.93	0.82	0.76	5.55	71.38
CP 16	0.81	0.79	0.64	4.08	62.48
CP 17	0.58	0.78	0.45	3.42	74.08
CP 18	0.85	0.97	0.82	2.41	28.65
CP 19	0.85	0.90	0.76	3.42	43.89
CP 20	0.85	0.95	0.81	3.47	42.16
Número CPs	20	20	20	20	20
Média	0.8155	0.8410	0.6874	3.823	54.52
Mediana	0.8100	0.8200	0.6678	3.421	51.52
Desv.Padrão	0.07877	0.07269	0.1010	1.516	16.64
Coef.Var.(%)	9.659	8.643	14.69	39.66	30.52
Mínimo	0.5800	0.7400	0.4524	2.128	28.65
Máximo	0.9300	1.020	0.9180	9.225	98.55

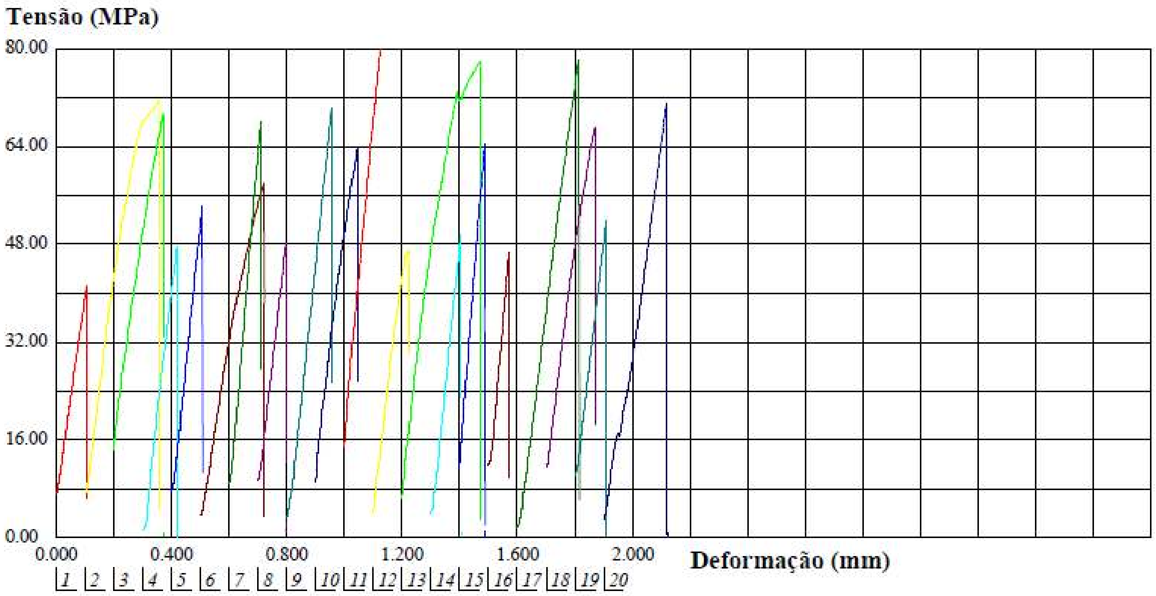


UFPEL - Treinamento Evandro

Relatório de Ensaio

Máquina: **Emic DL2000** Célula: **Trd 19** Extensômetro: - Data: **13/05/2009** Hora: **19:18:32** Trabalho n° **1640**
Programa: **Tesc versão 3.01** Método de Ensaio: **UFPEL Microtração Script_EP_Celula_100N**
Ident. Amostra: >>> Tipo: **AP4%_24h**

Corpo de Prova	Largura (mm)	Espessura (mm)	Área (mm2)	Força Máxima (kgf)	Tensão Máxima (MPa)
CP 1	0.82	0.87	0.71	3.00	41.22
CP 2	0.82	0.87	0.71	5.21	71.66
CP 3	0.80	0.85	0.68	4.82	69.45
CP 4	0.82	0.82	0.67	3.26	47.58
CP 5	0.82	0.78	0.64	3.55	54.39
CP 6	0.85	1.07	0.91	5.38	58.06
CP 7	0.71	0.78	0.55	3.84	68.05
CP 8	0.89	0.78	0.69	3.42	48.25
CP 9	0.77	0.79	0.61	4.36	70.29
CP 10	0.83	0.80	0.66	4.32	63.76
CP 11	0.76	0.69	0.52	4.93	92.27
CP 12	0.84	0.83	0.70	3.34	46.96
CP 13	0.78	0.95	0.74	5.90	78.08
CP 14	0.81	0.77	0.62	3.16	49.61
CP 15	0.78	0.71	0.55	3.64	64.37
CP 16	0.78	0.81	0.63	3.01	46.79
CP 17	0.80	0.86	0.69	5.49	78.29
CP 18	0.78	0.87	0.68	4.64	67.05
CP 19	0.76	0.79	0.60	3.18	52.01
CP 20	0.88	0.79	0.70	5.03	70.96
Número CPs	20	20	20	20	20
Média	0.8050	0.8240	0.6641	4.174	61.96
Mediana	0.8050	0.8050	0.6755	4.080	64.07
Desv.Padrão	0.04261	0.08230	0.08251	0.9430	13.42
Coef.Var.(%)	5.293	9.987	12.42	22.59	21.66
Mínimo	0.7100	0.6900	0.5244	2.999	41.22
Máximo	0.8900	1.070	0.9095	5.900	92.27



UFPEL - Treinamento Evandro

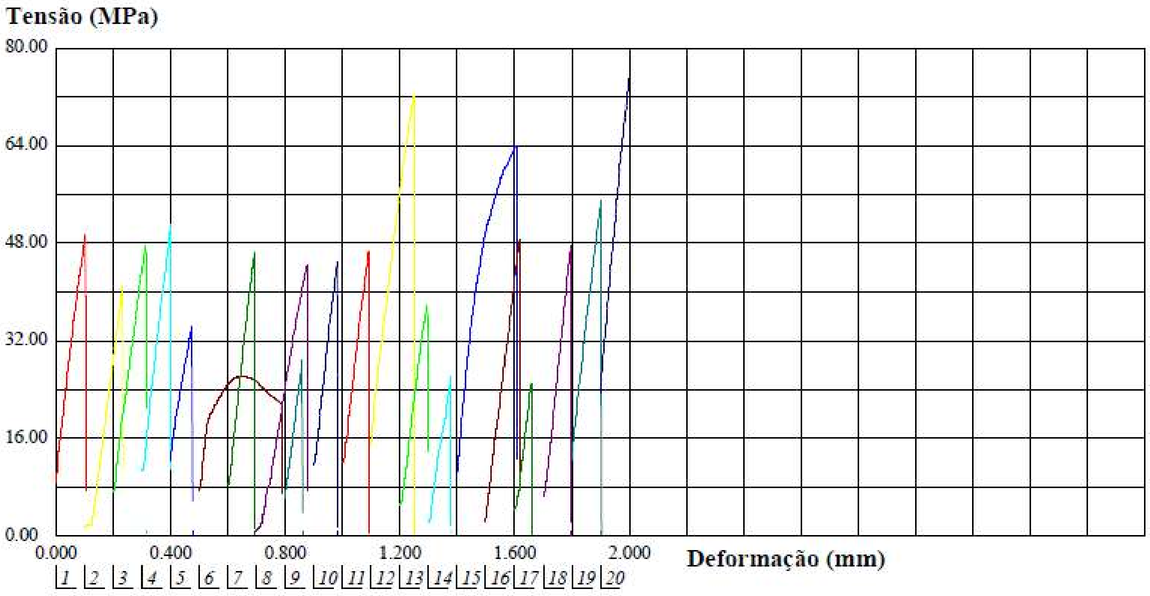
Relatório de Ensaio

Máquina: **Emic DL2000** Célula: **Trd 19** Extensômetro: - Data: **13/05/2009** Hora: **18:02:42** Trabalho n° **1639**

Programa: **Tesc versão 3.01** Método de Ensaio: **UFPEL Microtração Script_EP_Celula_ 0N**

Ident. Amostra: >>Tipo: **EDAB_24h**

Corpo de Prova	Largura (mm)	Espessura (mm)	Área (mm2)	Força Máxima (kgf)	Tensão Máxima (MPa)
CP 1	0.89	0.70	0.62	3.14	49.38
CP 2	0.81	0.81	0.66	2.75	41.16
CP 3	0.83	0.78	0.65	3.14	47.61
CP 4	0.80	0.83	0.66	3.46	51.07
CP 5	0.80	0.86	0.69	2.41	34.35
CP 6	0.80	0.80	0.64	1.72	26.34
CP 7	0.93	0.80	0.74	3.53	46.54
CP 8	0.85	0.90	0.76	3.47	44.44
CP 9	0.80	0.83	0.66	1.97	29.07
CP 10	0.86	0.78	0.67	3.08	45.00
CP 11	0.76	0.74	0.56	2.68	46.73
CP 12	0.81	0.79	0.64	4.72	72.36
CP 13	0.86	0.78	0.67	2.60	37.96
CP 14	0.83	0.87	0.72	1.93	26.18
CP 15	0.78	0.82	0.64	4.18	64.10
CP 16	0.79	0.79	0.62	3.11	48.79
CP 17	0.80	0.82	0.66	1.68	25.10
CP 18	0.82	0.83	0.68	3.31	47.76
CP 19	0.79	0.78	0.62	3.47	55.14
CP 20	0.81	0.79	0.64	4.90	75.07
Número CPs	20	20	20	20	20
Média	0.8210	0.8050	0.6607	3.062	45.71
Mediana	0.8100	0.8000	0.6560	3.121	46.63
Desv.Padrão	0.04012	0.04419	0.04549	0.8957	14.01
Coef.Var.(%)	4.887	5.489	6.885	29.25	30.66
Mínimo	0.7600	0.7000	0.5624	1.679	25.10
Máximo	0.9300	0.9000	0.7650	4.899	75.07



Apêndice B - Relatórios de Estatística – Sigma Stat 3.01 Systat INC.

Descriptive Statistics: terça-feira, junho 30, 2009, 01:39:23

Data source: Data 1 in Microtração dentina e esmalte-1

Column	**Size**	**Missing**	**Mean**	**Std Dev**	**Std. Error**	**C.I. of Mean**
DENTIN	60	0	54,061	15,977	2,063	4,127
ENAM	60	0	23,766	9,306	1,201	2,404
BDO	40	0	39,384	20,157	3,187	6,447
PA	40	0	44,108	21,815	3,449	6,977
EDAB	40	0	33,249	16,737	2,646	5,353

Column	**Range**	**Max**	**Min**	**Median**	**25%**	**75%**
DENTIN	73,450	98,550	25,100	50,340	44,720	67,155
ENAM	42,230	52,290	10,060	21,290	16,955	28,430
BDO	85,840	98,550	12,710	37,435	22,155	51,515
PA	77,560	92,270	14,710	46,030	25,095	64,065
EDAB	65,010	75,070	10,060	28,975	19,730	46,635

Column	**Skewness**	**Kurtosis**	**K-S Dist.**	**K-S Prob.**	**Sum**	**Sum of Squares**
DENTIN	0,400	0,0790	0,122	0,026	3243,670	190416,582
ENAM	0,996	0,681	0,121	0,029	1425,980	39000,054
BDO	0,809	0,369	0,109	0,257	1575,340	77888,545
PA	0,245	-1,042	0,137	0,058	1764,340	96382,777
EDAB	0,730	-0,0672	0,131	0,081	1329,970	55145,315

Two Way Analysis of Variance terça-feira, junho 30, 2009, 01:18:24

Data source: Data 1 in Microtração dentina e esmalte-1

Balanced Design

Dependent Variable: sqrt(col(21))

Normality Test: Passed (P = 0,417)

Equal Variance Test: Passed (P = 0,802)

Source of Variation	**DF**	**SS**	**MS**	**F**	**P**
Coinitiator	2	14,112	7,056	7,730	<0,001
Substrate	1	184,905	184,905	202,563	<0,001
Coinitiator x Substrate	2	1,897	0,948	1,039	0,357
Residual	114	104,063	0,913		
Total	119	304,977	2,563		

The difference in the mean values among the different levels of Coinitiator is greater than would be expected by chance after allowing for effects of differences in Substrate. There is a statistically significant difference (P = <0,001). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Substrate is greater than would be expected by chance after allowing for effects of differences in Coinitiator. There is a statistically significant difference (P = <0,001). To isolate which group(s) differ from the others use a multiple comparison procedure.

The effect of different levels of Coinitiator does not depend on what level of Substrate is present. There is not a statistically significant interaction between Coinitiator and Substrate. (P = 0,357)

Power of performed test with alpha = 0,0500: for Coinitiator : 0,924
Power of performed test with alpha = 0,0500: for Substrate : 1,000
Power of performed test with alpha = 0,0500: for Coinitiator x Substrate : 0,0543

Least square means for Coinitiator :
Group Mean
BDO6,078
PA 6,425
EDAB5,589
Std Err of LS Mean = 0,151

Least square means for Substrate :
Group Mean
DENTIN7,272
ENAM4,790
Std Err of LS Mean = 0,123

Least square means for Coinitiator x Substrate :
Group Mean
BDO x DENTIN7,305
BDO x ENAM4,852
PA x DENTIN7,827
PA x ENAM 5,023
EDAB x DENTIN6,685
EDAB x ENAM4,494
Std Err of LS Mean = 0,214

All Pairwise Multiple Comparison Procedures (Tukey Test):

Comparisons for factor: **Coinitiator**

Comparison	**Diff of Means**	**p**	**q**	**P**	**P<0,050**
PA vs. EDAB	0,836		35,534	<0,001	Yes
PA vs. BDO	0,347		32,297	0,240	No
BDO vs. EDAB	0,489		33,237	0,061	No

Comparisons for factor: **Substrate**

Comparison	**Diff of Means**	**p**	**q**	**P**	**P<0,050**
DENTIN vs. ENAM	2,483		220,128	<0,001	Yes

Descriptive Statistics: Sorpção e Solubilidade em água Monday, May 18, 2009, 11:34:50 PM

Data source: Data 1 in Notebook 1

Column	**Size**	**Missing**	**Mean**	**Std Dev**	**Std. Error**	**C.I. of Mean**
Sp_BDO	10	0	0.221	0.0115	0.00362	0.00819
Sl_BDO	10	0	0.206	0.0107	0.00337	0.00763

Sp_AP	10	0	0.227	0.0148	0.00469	0.0106
Sl_AP	10	0	0.211	0.0139	0.00439	0.00993
Sp_EDAB	10	0	0.226	0.0169	0.00534	0.0121
Sl_EDAB	10	0	0.211	0.0162	0.00511	0.0116

Column	**Range**	**Max**	**Min**	**Median**	**25%**	**75%**
Sp_BDO	0.0372	0.240	0.202	0.221	0.214	0.225
Sl_BDO	0.0352	0.224	0.188	0.206	0.199	0.210
Sp_AP	0.0510	0.256	0.205	0.229	0.216	0.234
Sl_AP	0.0479	0.238	0.190	0.213	0.201	0.218
Sp_EDAB	0.0502	0.246	0.196	0.231	0.220	0.238
Sl_EDAB	0.0485	0.230	0.182	0.216	0.205	0.223

Column	**Skewness**	**Kurtosis**	**K-S Dist.**	**K-S Prob.**	**Sum**	**Sum of Squares**
Sp_BDO	0.136	-0.411	0.150	0.653	2.210	0.490
Sl_BDO	0.140	-0.320	0.153	0.631	2.058	0.425
Sp_AP	0.320	0.351	0.173	0.484	2.273	0.519
Sl_AP	0.337	0.384	0.176	0.466	2.114	0.448
Sp_EDAB	-0.831	-0.152	0.209	0.239	2.260	0.513
Sl_EDAB	-0.853	-0.0912	0.211	0.230	2.112	0.448

One Way Analysis of Variance Monday, May 18, 2009, 11:35:51 PM

Data source: Sorpção em água

Normality Test: Passed (P = 0.236)

Equal Variance Test: Passed (P = 0.759)

Group Name	**N**	**Missing**	**Mean**	**Std Dev**	**SEM**
Sp_BDO	10	0	0.221	0.0115	0.00362
Sp_AP	10	0	0.227	0.0148	0.00469
Sp_EDAB	10	0	0.226	0.0169	0.00534

Source of Variation	**DF**	**SS**	**MS**	**F**	**P**
Between Groups	2	0.000220	0.000110	0.517	0.602
Residual	27	0.00573	0.000212		
Total	29	0.00595			

The differences in the mean values among the treatment groups are not great enough to exclude the possibility that the difference is due to random sampling variability; there is not a statistically significant difference (P = 0.602).

Power of performed test with alpha = 0.050: 0.879

One Way Analysis of Variance Monday, April 27, 2009, 12:33:05 AM

Data source: Data Módulo de Elasticidade.SNB

Normality Test: Passed (P = 0.704)

Equal Variance Test: Passed (P = 0.614)

Group Name	**N**	**Missing**	**Mean**	**Std Dev**	**SEM**

E_B_4%	10	1	1192.900	191.541	63.847
E_AP_4%	10	0	1062.212	184.836	58.450
E_EDAB	10	0	923.401	145.000	45.853

Source of Variation	**DF**	**SS**	**MS**	**F**	**P**
Between Groups	2	344850.523	172425.261	5.673	0.009
Residual	26	790208.174	30392.622		
Total	28	1135058.697			

The differences in the mean values among the treatment groups are greater than would be expected by chance; there is a statistically significant difference (P = 0.009).

Power of performed test with alpha = 0.050: 0.739

The power of the performed test (0.739) is below the desired power of 0.800.
Less than desired power indicates you are more likely to not detect a difference when one actually exists. Be cautious in over-interpreting the lack of difference found here.

All Pairwise Multiple Comparison Procedures (Tukey Test):

Comparisons for factor:

Comparison	**Diff of Means**	**p**	**q**	**P**	**P<0.050**
E_B_4% vs. E_EDAB	269.499	3	4.758	0.007	Yes
E_B_4% vs. E_AP_4%	130.688	3	2.307	0.251	No
E_AP_4% vs. E_EDAB	138.811	3	2.518	0.196	No

One Way Analysis of Variance Monday, May 18, 2009, 11:36:20 PM

Data source: Solubilidade em água

Normality Test: Passed (P = 0.223)

Equal Variance Test: Passed (P = 0.728)

Group Name	**N**	**Missing**	**Mean**	**Std Dev**	**SEM**
Sl_BDO	10	0	0.206	0.0107	0.00337
Sl_AP	10	0	0.211	0.0139	0.00439
Sl_EDAB	10	0	0.211	0.0162	0.00511

Source of Variation	**DF**	**SS**	**MS**	**F**	**P**
Between Groups	2	0.000199	0.0000994	0.526	0.597
Residual	27	0.00511	0.000189		
Total	29	0.00531			

The differences in the mean values among the treatment groups are not great enough to exclude the possibility that the difference is due to random sampling variability; there is not a statistically significant difference (P = 0.597).

Power of performed test with alpha = 0.050: 0.879

One Way Analysis of Variance Wednesday, June 24, 2009, 10:03:53 AM

Data source: Comparação co-iniciadores in Miniflexão .SNB

Normality Test: Passed (P = 0.681)

Equal Variance Test: Passed (P = 0.075)

Group Name	**N**	**Missing**	**Mean**	**Std Dev**	**SEM**
T_B_4%	10	1	84.843	14.021	4.674
T_AP_4%	10	0	84.573	6.209	1.963
T_EDAB_2	10	0	74.912	15.069	4.765

Source of Variation	**DF**	**SS**	**MS**	**F**	**P**
Between Groups	2	628.169	314.084	2.060	0.148
Residual	26	3963.386	152.438		
Total	28	4591.555			

The differences in the mean values among the treatment groups are not great enough to exclude the possibility that the difference is due to random sampling variability; there is not a statistically significant difference (P = 0.148).

Power of performed test with alpha = 0.050: 0.807

Two Way Analysis of Variance

Tuesday, June 23, 2009, 3:08:32 PM

Data source: Miniflexão e Módulo de Elasticidade.SNB

Balanced Design

Dependent Variable: Flexural strength

Normality Test: Passed (P = 0.486)

Equal Variance Test: Passed (P = 0.074)

Source of Variation	**DF**	**SS**	**MS**	**F**	**P**
Coinitiator	1	489.693	489.693	2.526	0.116
Concentration	4	6885.937	1721.484	8.879	<0.001
Coinitiator x Concentration	4	683.346	170.836	0.881	0.479
Residual	90	17448.798	193.876		
Total	99	25507.773	257.654		

The difference in the mean values among the different levels of Coinitiator is not great enough to exclude the possibility that the difference is just due to random sampling variability after allowing for the effects of differences in Concentration. There is not a statistically significant difference (P = 0.116).

The difference in the mean values among the different levels of Concentration is greater than would be expected by chance after allowing for effects of differences in Coinitiator. There is a statistically significant difference (P = <0.001). To isolate which group(s) differ from the others use a multiple comparison procedure.

The effect of different levels of Coinitiator does not depend on what level of Concentration is present. There is not a statistically significant interaction between Coinitiator and Concentration. (P = 0.479)

Power of performed test with alpha = 0.0500: for Coinitiator : 0.216

```
Power of performed test with alpha = 0.0500:  for Concentration : 0.999
Power of performed test with alpha = 0.0500:  for Coinitiator x Concentration : 0.0500

Least square means for Coinitiator :
Group     Mean
BDO       84.872
PA        80.446
Std Err of LS Mean = 1.969

Least square means for Concentration :
Group     Mean
1%        74.444
2%        72.270
4%        84.941
8%        86.919
16%       94.722
Std Err of LS Mean = 3.113

Least square means for Coinitiator x Concentration :
Group          Mean
BDO x 1%       81.384
BDO x 2%       72.865
BDO x 4%       85.309
BDO x 8%       90.080
BDO x 16%      94.722
PA x 1%        67.503
PA x 2%        71.675
PA x 4%        84.573
PA x 8%        83.758
PA x 16%       94.722
Std Err of LS Mean = 4.403


All Pairwise Multiple Comparison Procedures (Tukey Test):

Comparisons for factor: Concentration
Comparison     Diff of Means    p      q          P        P<0.050
16% vs. 2%          22.452      5   7.211     <0.001           Yes
16% vs. 1%          20.278      5   6.513     <0.001           Yes
16% vs. 4%           9.781      5   3.141      0.181            No
16% vs. 8%           7.803      5   2.506      0.396   Do Not Test
8% vs. 2%           14.649      5   4.705      0.011           Yes
8% vs. 1%           12.676      5   4.007      0.044           Yes
8% vs. 4%            1.978      5   0.635      0.992            No
4% vs. 2%           12.471      5   4.070      0.039            No
4% vs. 1%           10.498      5   3.372      0.129            No
1% vs. 2%            2.174      5   0.698      0.988            No
```

A result of "Do Not Test" occurs for a comparison when no significant difference is found between two means that enclose that comparison. For example, if you had four means sorted in order, and found no difference between means 4 vs. 2, then you would not test 4 vs. 3 and 3 vs. 2, but still test 4 vs. 1 and 3 vs. 1 (4 vs. 3 and 3 vs. 2 are enclosed by 4 vs. 2: 4 3 2 1). Note that not testing the enclosed means is a procedural rule, and a result of Do Not Test should be treated as if there is no significant difference between the means, even though one may appear to exist.

Two Way Analysis of Variance Tuesday, June 23, 2009, 3:32:24 PM

Data source: Miniflexão e Módulo de Elasticidade.SNB

Balanced Design

Dependent Variable: sqrt(col(7)) (Dados transformados por raiz quadrada)

Normality Test: Passed (P = 0.078)

Equal Variance Test: Passed (P = 0.366)

Source of Variation	**DF**	**SS**	**MS**	**F**	**P**
Coinitiator	1	57.579	57.579	4.790	0.031
Concentration	4	313.678	78.420	6.523	<0.001
Coinitiator x Concentration	4	383.646	95.912	7.978	<0.001
Residual	90	1081.937	12.022		
Total	99	1836.841	18.554		

The difference in the mean values among the different levels of Coinitiator is greater than would be expected by chance after allowing for effects of differences in Concentration. There is a statistically significant difference (P = 0.031). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Concentration is greater than would be expected by chance after allowing for effects of differences in Coinitiator. There is a statistically significant difference (P = <0.001). To isolate which group(s) differ from the others use a multiple comparison procedure.

The effect of different levels of Coinitiator depends on what level of Concentration is present. There is a statistically significant interaction between Coinitiator and Concentration. (P = <0.001)

Power of performed test with alpha = 0.0500: for Coinitiator : 0.475
Power of performed test with alpha = 0.0500: for Concentration : 0.978
Power of performed test with alpha = 0.0500: for Coinitiator x Concentration : 0.996

Least square means for Coinitiator :

Group	**Mean**
BDO	33.287
PA	31.770

Std Err of LS Mean = 0.490

Least square means for Concentration :

Group	**Mean**
1%	30.018
2%	30.904
4%	33.311
8%	33.697
16%	34.712

Std Err of LS Mean = 0.775

Least square means for Coinitiator x Concentration :

Group	**Mean**
BDO x 1%	32.327
BDO x 2%	33.049
BDO x 4%	34.140

BDO x 8%	35.227
BDO x 16%	31.694
PA x 1%	27.710
PA x 2%	28.760
PA x 4%	32.482
PA x 8%	32.168
PA x 16%	37.729

Std Err of LS Mean = 1.096

All Pairwise Multiple Comparison Procedures (Tukey Test):

Comparisons for factor: **Coinitiator**

Comparison	**Diff of Means**	**p**	**q**	**P**	**P<0.050**
BDO vs. PA	1.518	2	3.095	0.031	Yes

Comparisons for factor: **Concentration**

Comparison	**Diff of Means**	**p**	**q**	**P**	**P<0.050**
16% vs. 1%	4.693	5	6.054	<0.001	Yes
16% vs. 2%	3.807	5	4.911	0.007	Yes
16% vs. 4%	1.400	5	1.806	0.706	No
16% vs. 8%	1.014	5	1.308	0.887	Do Not Test
8% vs. 1%	3.679	5	4.745	0.010	No
8% vs. 2%	2.793	5	3.603	0.089	No
8% vs. 4%	0.386	5	0.498	0.997	Do Not Test
4% vs. 1%	3.293	5	4.247	0.028	No
4% vs. 2%	2.407	5	3.105	0.191	Do Not Test
2% vs. 1%	0.886	5	1.142	0.928	No

Comparisons for factor: **Concentration within BDO**

Comparison	**Diff of Means**	**p**	**q**	**P**	**P<0.05**
8% vs. 16%	3.533	5	3.222	0.162	No
8% vs. 1%	2.900	5	2.645	0.341	Do Not Test
8% vs. 2%	2.178	5	1.987	0.626	Do Not Test
8% vs. 4%	1.087	5	0.991	0.956	Do Not Test
4% vs. 16%	2.446	5	2.231	0.516	Do Not Test
4% vs. 1%	1.813	5	1.654	0.769	Do Not Test
4% vs. 2%	1.091	5	0.995	0.955	Do Not Test
2% vs. 16%	1.355	5	1.236	0.906	Do Not Test
2% vs. 1%	0.722	5	0.658	0.990	Do Not Test
1% vs. 16%	0.633	5	0.577	0.994	Do Not Test

Comparisons for factor: **Concentration within PA**

Comparison	**Diff of Means**	**p**	**q**	**P**	**P<0.05**
16% vs. 1%	10.019	5	9.138	<0.001	Yes
16% vs. 2%	8.970	5	8.181	<0.001	Yes
16% vs. 8%	5.562	5	5.073	0.005	No
16% vs. 4%	5.247	5	4.786	0.009	No
4% vs. 1%	4.772	5	4.352	0.023	No
4% vs. 2%	3.723	5	3.395	0.125	No
4% vs. 8%	0.315	5	0.287	1.000	Do Not Test
8% vs. 1%	4.458	5	4.066	0.040	No
8% vs. 2%	3.408	5	3.108	0.190	Do Not Test

2% vs. 1%	1.050	5	0.957	0.961	No

Comparisons for factor: **Coinitiator within 1%**

Comparison	**Diff of Means**	**p**	**q**	**P**	**P<0.05**
BDO vs. PA	4.617	2	4.211	0.004	Yes

Comparisons for factor: **Coinitiator within 2%**

Comparison	**Diff of Means**	**p**	**q**	**P**	**P<0.05**
BDO vs. PA	4.289	2	3.912	0.007	Yes

Comparisons for factor: **Coinitiator within 4%**

Comparison	**Diff of Means**	**p**	**q**	**P**	**P<0.05**
BDO vs. PA	1.658	2	1.512	0.288	No

Comparisons for factor: **Coinitiator within 8%**

Comparison	**Diff of Means**	**p**	**q**	**P**	**P<0.05**
BDO vs. PA	3.059	2	2.790	0.052	No

Comparisons for factor: **Coinitiator within 16%**

Comparison	**Diff of Means**	**p**	**q**	**P**	**P<0.05**
PA vs. BDO	6.035	2	5.505	<0.001	Yes

A result of "Do Not Test" occurs for a comparison when no significant difference is found between two means that enclose that comparison. For example, if you had four means sorted in order, and found no difference between means 4 vs. 2, then you would not test 4 vs. 3 and 3 vs. 2, but still test 4 vs. 1 and 3 vs. 1 (4 vs. 3 and 3 vs. 2 are enclosed by 4 vs. 2: 4 3 2 1). Note that not testing the enclosed means is a procedural rule, and a result of Do Not Test should be treated as if there is no significant difference between the means, even though one may appear to exist.

Relatório de Experimento

Metodologia de microdureza Knoop

O ensaio de microdureza, disposto no projeto de tese não foi incluído nos resultados. A resina adesiva modelo não apresentava características de opacidade, inviabilizando a visualização e mensuração das edentações realizadas nos corpos de prova, dificultando a obtenção de medidas precisas e confiabilidade dos dados, representando um sério viés na obtenção de dados. Adicionalmente, uma vez que as propriedades mecânicas (resistência à flexão e módulo de elasticidade) dos adesivos foram avaliadas em associação à cinética de polimerização, esta análise foi considerada desnecessária.

Relatório de artigos no período de Pós-graduação, 2006 a 2009.

1. Artigos publicados em Periódicos

1.1 LIMA, G. S., OGLIARI, F.A., SILVA, E.O., ELY, C., DEMARCO, F.F., PETZHOLD, C.L., CARRENO, N. L. V., PIVA, E. Influence of the water concentration in an experimental self-etching primer on the bond strength to dentin. The Journal of Adhesive Dentistry. 10 (2008) No. 3 (13.06.2008).

1.2 LIMA, G. S., FONTES, S. T., TARQUÍNIO, S.B.C., ETGES, A., ARAUJO, L. M. A., GOMES, A. P. N. A survey of oral and maxillofacial biopsies in children. The 20-year CDDB/SD - UFPel study. Journal of Applied Oral Science (Online). *J Appl Oral Sci*. v.16, n.6, p.397-402, 2008.

1.3 FARIA-E-SILVA, A. L.; CASSELLI, D.S. ; LIMA, G. S. ; OGLIARI, F. A.; PIVA, E.; MARTINS, L.R. Kinetics of conversion of two dual-cured adhesive systems. *Journal of Endodontics,* v. 34, p. 1115-1118, 2008.

1.4 OGLIARI, F.A., ELY, C., LIMA, G. S., CONDE, M. C. M., DEMARCO, F.F., PETZHOLD, C. L., PIVA, E. Onium salt reduces the inhibitory polymerization effect from an organic solvent in a model dental adhesive resin. *Journal of Biomed Mater Res B, Appl Biomater.*, v. 86, n.1,p.113-118, 2008.

1.5 OGLIARI, F.A. , SILVA, E.O., LIMA, G. S., MADRUGA, F. C., HENN, S., BUENO, P., PETZHOLD, C. L., PIVA, E. Synthesis of phosphate monomers and bonding to dentin: Esterification methods and use of phosphorus pentoxide. *Journal of Dentistry*. v.36, p.171 - 177, 2008.

1.6 LUND, R.G., OGLIARI, F.A., LIMA, G. S., DEL-PINO, PETZHOLD, C. L., EVANDRO PIVA. Diametral tensile strength of two brazilian resin-modified glass ionomers cements: Influence of the powder/liquid ratio. *Braz J of Oral Science* (Online). , v.6, p.1354 - 1356, 2007.

2 Artigos Submetidos para publicação

2.1 FONTES, S. T., OGLIARI, F. A., LIMA, G.S., BUENO, M., Schneider,L. F. J., PIVA, E. Tetrahydrofuran as alternative solvent in dental adhesive systems. *Dental Materials*. Manuscript Number: DEMA-D-09-00043. (Aceito, junho 2009)

2.2 LIMA, G. S., SOUZA E SILVA, M. G., OGLIARI, F.A., LIMA, G. B., PETZHOLD, C. L., CARREÑO, N. L. V., DEMARCO, F. F., PIVA, E. Influence of acidic functional monomers in the composition of experimental self-etching adhesive systems on bond strength to dentin and enamel. *Journal of Adhesive Dentistry.* MS 906.

2.3 LIMA, G. S., OGLIARI, F. A., MORAES, R. R., MATTOS, E. M., SILVA, A. F., CARREÑO, N. L. V., PETZHOLD, C. L., PIVA, E. Water content in self-etching primers affects their aggressiveness and strength of bonding to ground enamel. *Journal of Adhesive Dentistry.* MS 946.

2.4 COLLARES, F. M., OGLIARI, F. A., LIMA, G. S., FONTANELLA, V., PIVA, E., SAMUEL, S. Ytterbium Trifluoride as a Radiopaque Agent for Dental Cements. *Journal of Endodontics.* JOE-S-09-00374.

2.5 FONTES, S. T., MONTEMEZZO, M. L., FLORES, J. B., LIMA, G. S., OGLIARI, F. A., BUENO, M., PIVA, E. Evaluation of six two-step etch-and-rinse adhesive systems – degree of conversion and microtensile bond strength to dentin. *Journal of Applied Oral Science.* JAOS-752

2.6 LIMA, G.S., SILVA, G. F., GOMES, A. P., ARAÚJO, L. M. A., SALUM, F.G. DICLOFENAC IN HYALURONIC ACID GEL: AN ALTERNATIVE TREATMENT FOR ACTINIC CHEILITIS. *Journal of Applied Oral Science.* JAOS-1027.

2.7 HENN, S., LIMA, G. S., MASOTTI, A. S., DEMARCO, F. F. Clareamento dental caseiro através de fitas adesivas (White strips): relato de caso e discussão. At-homa tooth whitening using white strips: A case report and discussion. *Clínica (International Journal of Brazilian Dentiistry).*

2.8 PIVA, E., LIMA, G. S., OGLIARI, F. A., COLLARES, F.M., LEAL, F. B., SAMUEL, S. M. W., PETZHOLD, C. Onium salt increase bond strength of a model self-etching adhesive. *Journal of Dentistry.*

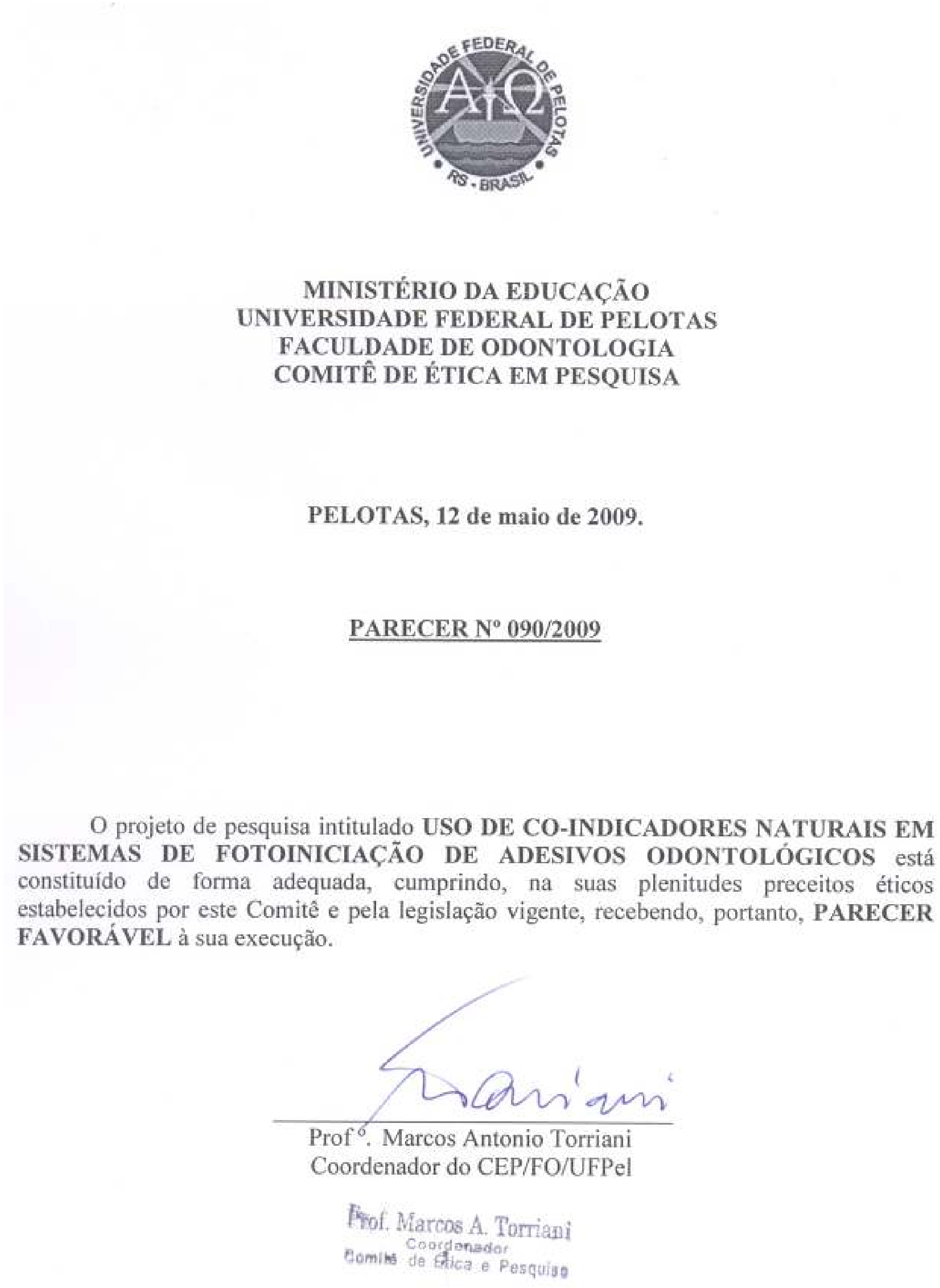
2.9. ELY, C., OGLIARI, F.A., SCHNEIDER, L.F.J., LIMA, G.S., PETZHOLD, C.L., PIVA, E. Thioxanthone and iodonium salt derivatives as initiators of a model dental adhesive. *Acta Biomaterialia.*

3 Patente (produto tecnológico)

3.1 PIVA, E., LINE, S., OGLIARI, F. A., PARDO, A. P., SILVA, A. F., LIMA, G.S., CARVALHO, R. V., PETZHOLD, C. L. **Patente***: Composição para adesivos odontológicos com capacidade de inibição de metaloproteases,* 2008. Com pedido de depósito de patente no INPI, junto a UFPel. Patente tipo: Modelo de Utilidade. Código: 000022080389. Pedido para Depósito 22/06/2008.

ANEXOS

Anexo A



MINISTÉRIO DA EDUCAÇÃO
UNIVERSIDADE FEDERAL DE PELOTAS
FACULDADE DE ODONTOLOGIA
COMITÊ DE ÉTICA EM PESQUISA

PELOTAS, 12 de maio de 2009.

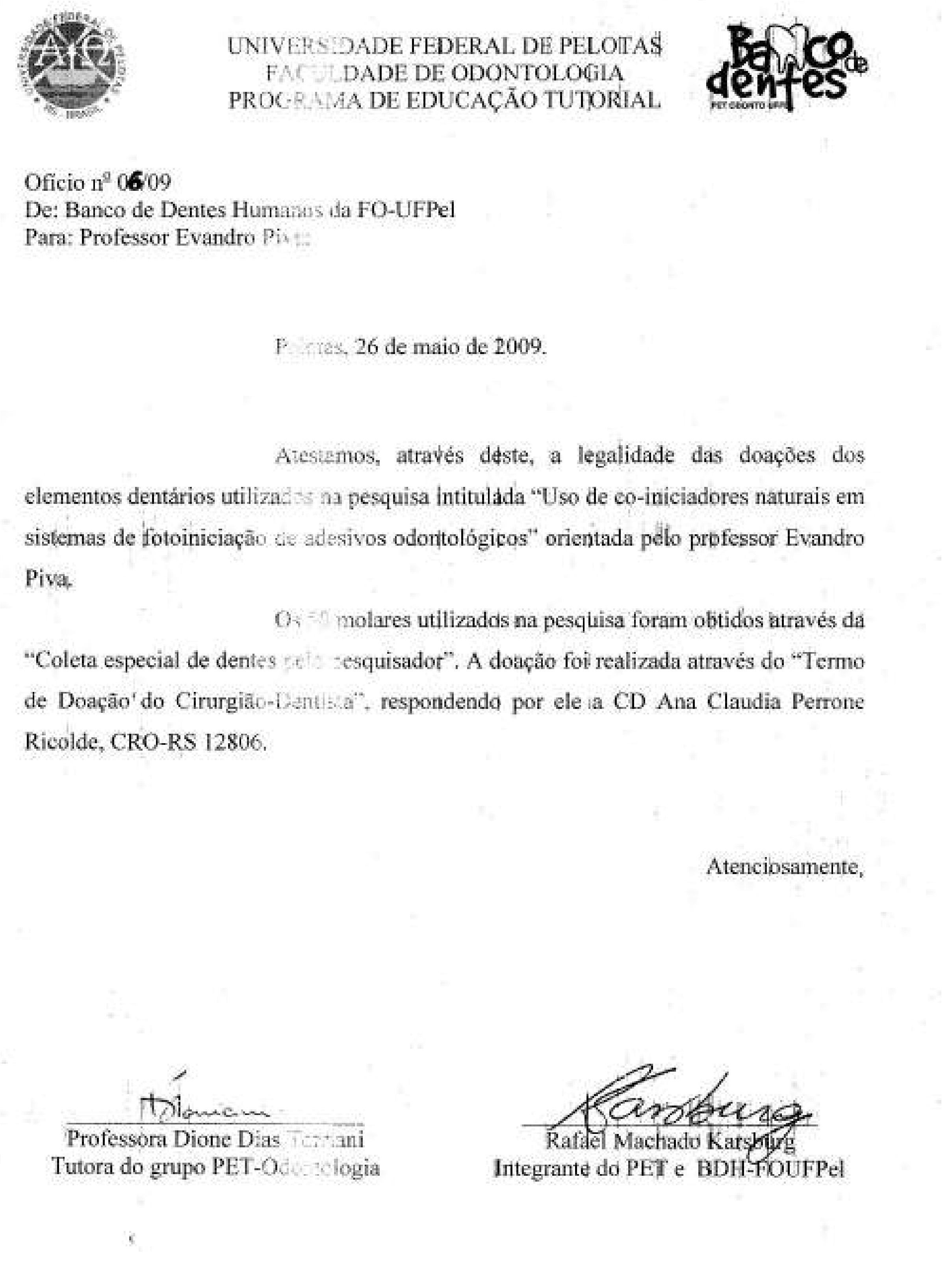
PARECER Nº 090/2009

O projeto de pesquisa intitulado **USO DE CO-INDICADORES NATURAIS EM SISTEMAS DE FOTOINICIAÇÃO DE ADESIVOS ODONTOLÓGICOS** está constituído de forma adequada, cumprindo, na suas plenitudes preceitos éticos estabelecidos por este Comitê e pela legislação vigente, recebendo, portanto, **PARECER FAVORÁVEL** à sua execução.

Prof º. Marcos Antonio Torriani
Coordenador do CEP/FO/UFPel

Prof. Marcos A. Torriani
Coordenador
Comitê de Ética e Pesquisa

Anexo B



UNIVERSIDADE FEDERAL DE PELOTAS
FACULDADE DE ODONTOLOGIA
PROGRAMA DE EDUCAÇÃO TUTORIAL

Oficio nº 06/09
De: Banco de Dentes Humanos da FO-UFPel
Para: Professor Evandro Piva

Pelotas, 26 de maio de 2009.

Atestamos, através deste, a legalidade das doações dos elementos dentários utilizados na pesquisa intitulada "Uso de co-iniciadores naturais em sistemas de fotoiniciação de adesivos odontológicos" orientada pelo professor Evandro Piva.

Os 50 molares utilizados na pesquisa foram obtidos através da "Coleta especial de dentes pelo pesquisador". A doação foi realizada através do "Termo de Doação do Cirurgião-Dentista", respondendo por ele a CD Ana Claudia Perrone Ricolde, CRO-RS 12806.

Atenciosamente,

Professora Dione Dias Torriani
Tutora do grupo PET-Odontologia

Karsburg
Rafael Machado Karsburg
Integrante do PET e BDH-FOUFPel