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Data Article

Dataset on the effect of flavonoids on the stabilization of the resin-dentin interface

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ABSTRACT

Data in this article are associated with our research article "Effect of Myricetin on Odontoblast-like Cells and its Potential to Preserve Resin–Dentin Bonds." Both a poor infiltration of resin monomers into the demineralized dentin matrix and hydrolytic degradation of the adhesive could lead to the instability of the resin–dentin interface. The degradation of collagen is caused by matrix metalloproteinases (MMP) and cysteine cathepsins. These collagenolytic enzymes are contained in their latent form as pro-MMPs in the dentinal structure, and undergo activation during the adhesive process. Given that the integrity of the collagen matrix is essential for the preservation of the dentin bond strength in both the medium and long term, the inhibition of these proteases is necessary to improve the durability of adhesive restorations. Among the different strategies suggested to improve both the behavior of the substrate against enzymatic degradation and the biomechanical behavior of the adhesive interface, the use of protease inhibitors and collagen crosslinking agents has been recommended, such as polyphenols. Research has focused on flavonoids such as proanthocyanidins (PAC), a class of phenolic compounds found in a variety of plants such as blueberry and grape whose chemical structure favors their action as cross-linking agents. However, the focus has recently shifted towards myricetin (MYR) due to its chemical structure: a greater amount of hydroxyl groups at the substitution positions, which form bonds with the carbonyl groups of the side

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chains of collagen amino acids and generate interfiber bonds. Our previous study has shown the efficacy of MYR both as a cross-linking agent and as a MMP inhibitor without any immediate effects on microtensile bond strength (μTBS) and preserving it for six months after storage, and maintaining the odontoblastic phenotype without affecting cell viability. The objective of this article is to present a dataset on the effect of flavonoids PAC and MYR on the resin–dentin interface. Given that durability of the resin–dentin bond holds great importance for the clinical longevity of adhesive restorations, our data aims to show the effects of these flavonoids on resin–dentin μTBS after 18-month storage. Test groups for the μTBS assay were set as follows: G1 (negative control), conventional adhesion technique; G2 (vehicle control), 100% ethanol (EtOH) for 120 s; G3, 0.2% chlorhexidine (CHX) for 60 s; G4, 1% glutaraldehyde (GA) for 60 s; and G5, 600 μM myricetin (MYR) for 120 s. Datasets were exported to SPSS software, version 21.0 (SPSS, Chicago, IL, USA) for analysis using the Shapiro–Wilk, a two-way analysis of variance including factor interactions (treatment and storage time). Data are presented as mean ± standard deviation (SD). Differences with p-values < 0.05 were considered significant. Our data can be used as a basis for comparison among other natural and synthetic substances that could work as MMP inhibitors and crosslinking agents. These findings could be useful for designing an effective strategy towards the stabilization of the hybrid layer in a relevant clinical protocol.

Specifications Table

Subject	Dentistry, Oral Surgery and Medicine
Specific subject area	Adhesive dentistry
Type of data	Table Figure
How data were acquired	μTBS was evaluated using the universal testing machine Shimadzu AG-IS 5kN (Shimadzu, Tokyo, Japan). Samples were subjected to a tensile force at a crosshead speed of 0.5 mm/min. μTBS was calculated as the maximum load at failure divided by the cross-sectional area of the stick (1 mm^2), and was expressed in MegaPascals (MPa). Data were processed with the TRAPEZIUM X software (Microsoft Corporation, Redmond, Washington, USA).
Data format	Raw Analyzed
Parameters for data collection	Test groups for the μTBS assay were set as follows: G1 (negative control), conventional adhesion technique; G2 (vehicle control), 100% EtOH for 120 s; G3, 0.2% CHX for 60 s; G4, 1% GA for 60 s; and G5, 600 μM MYR for 120 s. Freshly extracted human molars were randomized into the test groups. The demineralized dentin was treated with the test substances prior to the conventional bonding protocol. Adhesive treatment was performed using Adper Single Bond 2 (3M ESPE, St. Paul, MN, USA) and Filtek Z-350 XT composite (3M ESPE). The restored teeth were sectioned perpendicular to the bond interface to obtain sticks (cross-sectional area, 1.0 ± 0.1 mm^2). The bonded sticks originated from the same teeth and were randomly divided and assigned to be tested immediately (24 h) or after 18 months of storage in artificial saliva at 37 °C in a Hygrobath (Whip Mix, Louisville, KY, USA). Each bonded stick was subjected to a tensile force using the universal testing machine Shimadzu AG-IS 5kN (Shimadzu, Tokyo, Japan) to obtain the μTBS values in MPa.

Description of data collection	Microsoft Office Excel 2010 (Microsoft Corporation, Redmond, Washington, USA) was used for the μTBS databases construction. Datasets were exported to SPSS software, version 21.0 (SPSS, Chicago, IL, USA) for analysis using the Shapiro–Wilk, a two-way analysis of variance including factor interactions (treatment and storage time). Data are presented as mean ± SD. Differences with p-values < 0.05 were considered significant.
Data source location	Universidad Nacional de Colombia, Bogotá, Colombia.
Data accessibility	The raw data are deposited in New Folder, Supporting Data V.3. Mendeley Data, V3, https://doi.org/10.17632/p3g3zdv6zb.3. [1]. https://data.mendeley.com/datasets/p3g3zdv6zb/3
Related research article	P.A. Baldion, C. C. Cortes, J. E. Castellanos, D. E. Betancourt. Effect of Myricetin on Odontoblast-like Cells and its Potential to Preserve Resin–Dentin Bonds. J Mech Behav Biomed. 117 (2021) 104392 [2].

Value of the Data

- Our data provide novel information on the effect of flavonoids on the resin–dentin interface and show the effect of MYR on the biomechanical behavior of the resin-dentin interface and on the dentin bonding after 18 months.
- Mainly, these data can be useful to researchers in the area of dental materials exploring different treatment possibilities to improve the durability of polymeric adhesive restorations, whose frequent replacement may have implications for the oral health of patients, and in turn, have an impact on public health policies.
- Our data can be used as a basis for comparison among other natural and synthetic substances that could work as MMP inhibitors and crosslinking agents.
- Given that the durability of the resin-dentin holds great importance for the clinical longevity of adhesive restorations, our data provide sound grounds for research in dental adhesion and give some suggestions for the development of new techniques and materials that guarantee a better performance of adhesive restorations.
- An understanding of the biomechanical and biological behavior of crosslinking agents such as flavonoids on dental structure could enable a better estimation of the advantages associated to the clinical use of flavonoids within an adhesive protocol, allowing for the development of improved strategies for the aesthetic restoration of dental structures.

1. Data Description

The whole dataset of the original article and this article is available on the online Mendeley Data repository under the title “Myricetin dataset” [1]. This dataset contains raw data of the cell response to flavonoids obtained on dentin and on an in vitro cell model of odontoblast-like cells (OLC) differentiated from hDPSC [3].

Regarding the effect of flavonoids on dentin and on the stabilization of the resin–dentin interface, the raw data were gathered in several worksheets as follows: the “MMP activity” worksheet displays the inhibitory effect of MYR on the matrix-bound endogenous MMPs in dentin; the “μTBS 6 months and μTBS 18 months” worksheets show the characterization of resin–dentin interface assessed through the μTBS after six and 18 months of storage; the “Failure mode” worksheet presents the corresponding failure mode data analysis; and the “TS-E” worksheet shows the effect of MYR on the tensile strength and elastic modulus of dentin.

Regarding the effect of flavonoids on odontoblast-like cells, the raw data were gathered in several worksheets as follows: the “Resazurin” worksheet shows the cell viability of OLC exposed to different MMP inhibitors and cross-linking agents evaluated by resazurin assay; the “DMP-1 qPCR” worksheet displays the maintenance of the odontoblast phenotype assessed through

Table 1
Data description available at Mendeley Data repository.

Excel worksheet	Method	Test Groups*	n	Data	Measurement units
MMP activity	Generic MMP assay kit (fluorometric green Abcam)	DD APMA H_2O_2 EtOH EDTA PAC MYR	6	Measurement of the MMP activity	Fluorescence intensity expressed in relative fluorescence units (RFU)
uTBS six months	Microtensile bond strength	Control EtOH CHX MYR	15	μTBS immediate and after six months of storage	MegaPascals (MPa)
uTBS 18 months	Microtensile bond strength	Control EtOH CHX GA PAC MYR	15	μTBS immediate and after 18 months of storage	MegaPascals (MPa)
Failure mode six months	Failure mode after μTBS test	Control EtOH CHX MYR	15	Type of fracture	Percentage of failures classified as adhesive, mixed, or cohesive
Failure mode 18 months	Failure mode after μTBS test	Control EtOH CHX GA PAC MYR	15	Type of fracture	Percentage of failures classified as adhesive, mixed, or cohesive
TS - E	Dentin tensile strength - Elastic modulus	DD EtOH CHX GA MYR	15	Maximum load and strain-stress curve's slope	MegaPascals (MPa)
DMP-1 qPCR	Evaluation of gene expression by reverse transcription quantitative polymerase chain reaction	Control + EtOH CHX PAC GA MYR	3	Relative quantification of DMP-1 RNA messengers	Relative Fold Change/β-Act
Resazurin	Resazurin technique	Control - Control + EtOH CHX PAC GA MYR	3	Metabolic activity evaluation	Fluorescence intensity (expressed in RFU), which is proportional to the number of viable cells

* DD: demineralized dentin with 10 % phosphoric acid (H_3PO_4) overnight; APMA: 1 mM 4-aminophenylmercuric acetate for 2 h; H_2O_2: 1 mM hydrogen peroxide for 2 h; EtOH: 100% ethanol for 120 s; CHX: 0.2% chlorhexidine for 60 s; EDTA: 5 mM ethylenediaminetetraacetic acid; GA: 1% glutaraldehyde for 60 s; PAC: 6% proanthocyanidin for 6 min; MYR: 600 μM myricetin for 120 s.

the evaluation of the expression of a specific marker for odontoblast phenotype Dentin Matrix Protein-1 (DMP-1) [4] before and after exposure to the test substances.

Raw data on the biomechanical and biological behavior of flavonoids on dentin and on the OLC response assessed for each assay are shown in tables where the following information is at display: group or treatment, time points (only for the Resazurin assay), intraexperiment and inter-experiment replicas, mean ± SD. Analyzed data on the aforementioned methods are displayed in research article [2] and summarized in Table 1, except for the μTBS at 18 months after storage, which are to be found below.

Table 2
Microtensile bond strength of resin–dentin sticks under different treatments, immediate and after 18 months of storage.

Treatment	Storage time	Mean*	SD	p–value
Control (-)	Immediate	25.27	6.3	0.000[a]
	18 months	9.11	3.36	
100% EtOH	Immediate	31.75	5.5	0.000[a]
	18 months	15.47	4.71	
0.2% CHX	Immediate	28.67	7.6	0.024[a]
	18 months	20.77	2.98	
1% GA	Immediate	26.08	2.93	1.000
	18 months	24.94	2.79	
6% PA	Immediate	25.31	2.75	1.000
	18 months	26.65	2.87	
600 µM MYR	Immediate	27.76	6	0.996
	18 months	25.16	1.98	

[a] Statistical difference between the immediate value and after 18 months of storage within each group ($p<0.05$).
* Values in MPa. ($n = 15$).

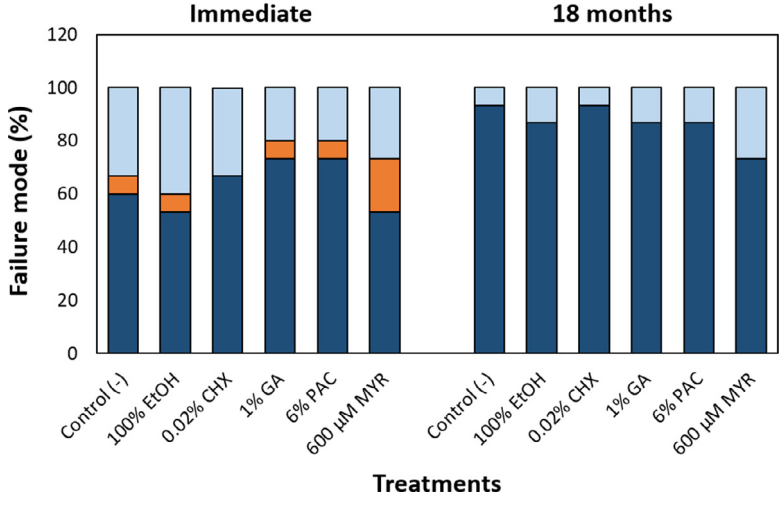

Fig. 1. Failure mode of resin–dentin interfaces. After the µTBS test, failure mode was evaluated immediately and after 18 months of storage under stereomicroscopy (50X), and it was classified as adhesive (dark blue), cohesive (orange) and mixed (light blue). The failure mode was recorded according to the portion of the interface affected by the fracture. The number of failures per group was converted into a percentage, setting the 15 samples as 100%.

Analyzed data on the effect of several dentin pretreatments on the µTBS, immediate and after 18 months of storage, are displayed in Table 2. The data of µTBS mean values (MPa) were obtained from 24 freshly human restored molars sectioned in sticks with 1.0 ± 0.1 mm2 of bonded area. From each test group, half of the samples were tested immediately and the other half were stored for 18 months in artificial saliva at 37 °C in a Hygrobath (Whip Mix, Louisville, KY, USA). The failure modes were evaluated under stereomicroscopy (SMZ 800, Nikon, Tokyo, Japan) at a 50X magnification and classified as cohesive, adhesive, or mixed. The data are presented as a percentage relative to the total number of samples in each group ($n = 15$) (Fig. 1).

2. Experimental Design, Materials and Methods

Freshly extracted non-carious human third molars were obtained with informed consent from 18 to 25-year-old patients under a protocol approved by the Ethics Committee of Facultad de Odontología of Universidad Nacional de Colombia (CIE-233-14). The teeth were stored in 0.9% sodium chloride and 0.02% sodium azide at 4 °C, and were used within 2 months after extraction. The enamel and superficial dentin of human molars were removed with a low-speed saw (Isomet, Buehler Ltd., Lake Bluff, IL, USA) under water cooling. The dentin surfaces were sanded with 600-grit wet silicon-carbide paper to standardize the smear layer [5], and then etched with 37% H_3PO_4 (Scotchbond etchant, 3M ESPE, St. Paul, MN, USA) for 10 s, and rinsed with distilled water for 20 s.

The teeth were randomly allocated into the control and five experimental groups by a person not involved in the research protocol. Test substances were applied as a primer before using the adhesive system as follows: G1 (control group), conventional adhesive treatment with Adper Single Bond 2; G2, 100% EtOH for 120 s; G3, 0.2% CHX for 60 s; G4, 1% GA for 60 s; G5, 6% PAC for 6 min; and G6, 600 µM MYR for 120 s. The adhesive protocol was performed according to the manufacturer's instructions. A 6-mm-high composite resin block was built with increments of up to 2 mm using Filtek Z-350 XT composite (3M ESPE) polymerized with Bluephase light-curing unit (Ivoclar Vivadent, AG Schaan, Liechtenstein) at 800 mW/cm^2. The light intensity was checked using a radiometer (Demetron LED Radiometer, Kerr Sybron, Middleton, WI, USA).

Restored molars were longitudinally sectioned, perpendicular to the resin–dentin interface, to obtain sticks with an adhesive cross-sectional area of 1± 0.1 mm^2, using a diamond saw mounted in a cutting machine (Isomet). The cross-sectional area of each stick was measured using a digital caliper (Mitutoyo, Tokyo, Japan). The sticks were stored in distilled water at 37 °C for 24 h. They were either tested for µTBS immediately or after 18 months of storage in artificial saliva at 37 °C (n= 15) in a Hygrobath (Whip Mix, Louisville, KY, USA). The artificial saliva was changed weekly.

Each bonded stick was glued to a jig for microtensile testing using cyanoacrylate resin, and then subjected to a tensile force using the universal testing machine Shimadzu AG-IS 5kN (Shimadzu, Tokyo, Japan) at a crosshead speed of 0.5 mm/min. The failure mode was assessed under stereomicroscopy (SMZ 800, Nikon, Tokyo, Japan) at 50X magnification, and each failure was classified as adhesive (at the resin–dentin interface), cohesive (within dentin or resin), or mixed (at the resin–dentin interface and partial cohesive failure).

µTBS was calculated as the maximum load at failure divided by the cross-sectional area of the stick (1 mm^2), and was expressed in MPa. Data were processed using the TRAPEZIUM X software (Microsoft Corporation, Redmond, Washington, USA). The data were analyzed performing a Shapiro–Wilk test and a two-way ANOVA with factor interactions. Multiple comparisons were made in order to evaluate the effect of storage time within each group. Data are expressed as mean ± SD, and differences with p-values < 0.05 were considered significant.

Ethics Statement

Project approved by the ethics committee of the Facultad de Odontología, Universidad Nacional de Colombia [B.ClEFO-229-18].

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships which have or could be perceived to have influenced the work reported in this article.

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