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Effect of flavonoids on the mechanical properties of demineralised dentine

D.J. Epasinghe [a], C.K.Y. Yiu [a,*], M.F. Burrow [b], J.K.H. Tsoi [c], F.R. Tay [d]

[a] Paediatric Dentistry and Orthodontics, Faculty of Dentistry, The University of Hong Kong, Prince Philip Dental Hospital, 34 Hospital Road, Pokfulam, Hong Kong, China

[b] Oral Diagnosis and Polyclinics, Faculty of Dentistry, The University of Hong Kong, Prince Philip Dental Hospital, 34 Hospital Road, Pokfulam, Hong Kong, China

[c] Oral Rehabilitation, Faculty of Dentistry, The University of Hong Kong, Prince Philip Dental Hospital, 34 Hospital Road, Pokfulam, Hong Kong, China

[d] Department of Endodontics, College of Dental Medicine, Georgia Regents University, Augusta, GA, USA

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ABSTRACT

Objectives: This study compared the effect of three flavonoids: proanthocyanidin, naringin and quercetin on the modulus of elasticity (MOE) and ultimate tensile strength (UTS) of demineralised dentine.

Methods: Thirty teeth were sectioned into 0.5 mm × 1.7 mm × 7 mm beams for MOE measurement. Another 30 non-carious molars were sectioned into 0.5 mm × 0.5 mm thick dentine beams for UTS testing. Demineralised specimens were divided into three groups according to treatments: 6.5% proanthocyanidin, 6.5% quercetin and 6.5% naringin. Specimens were kept in their respective solutions and tested at baseline, 10 min, 30 min, 1 h and 4 h. The MOE of each specimen was determined using a three-point bending test at a crosshead speed of 0.5 mm/min. For UTS evaluation, each specimen was tested in tension until failure using a crosshead speed of 1 mm/min. Means and standard deviation were calculated. Two-way ANOVA and Tukey test were used to evaluate the effect of flavonoid treatment and treatment duration on MOE and UTS.

Results: Both MOE and UTS were significantly affected by flavonoid treatment ($p < 0.001$) and treatment duration ($p < 0.001$). Interaction of the two factors was significant for MOE ($p < 0.001$), but not for UTS ($p > 0.05$). Flavonoid treatment improved the mechanical properties of demineralised dentine in the order: proanthocyanidin > quercetin > naringin. naringin. It took a longer time for the flavonoids to produce a significant change in UTS, when compared to MOE.

Conclusion: Proanthocyanidin was more effective than quercetin and naringin in improving biomechanical properties of dentine matrix, thereby enhancing preventive and reparative dental therapies.

Clinical Significance: Despite its larger molecular size, proanthocyanidin was more effective than quercetin and naringin, in enhancing the biomechanical properties of demineralised dentine.

* *Corresponding author*. Tel.: +852 28590251; fax: +852 25593803.
E-mail address: ckyyiu@hkucc.hku.hk (C.K.Y. Yiu).

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1. Introduction

Despite the immense advances in adhesive dentistry over the years, resin–dentine bonds are less durable than resin–enamel bonds.[1–3] The decline in resin–dentine bond strength over time could be attributed to degradation of either the hydrophilic resin components following water sorption, or denuded collagen fibrils within the bonded interface by dentine-bound enzymes, such as matrix metalloproteinases (MMPs) and cysteine cathepsins.[4]

Biomodification of dentine has recently become a popular approach to enhance the reparative potential of the dentine substrate. Proanthocyanidin (PA), a plant flavonoid which belongs to the flavanol group, has been shown to improve the mechanical properties of the demineralised dentine collagen matrix[5,6] and increase its resistance to biodegradation by collagenolytic enzymes via its cross-linking effects.[7] Apart from its effects on collagen fibrils, PA also interacted with proteoglycans resulting in a remarkable reduction of GAGs content.[7] Furthermore, PA inhibited both soluble and matrix-bound MMPs and cathepsin K.[8] Hence, PA has been suggested as a potential dentine tissue modifier for enhancing the reparative potential of dentine.[7]

Being an oligomeric molecule, PA has a large molecular size, which may limit its ability to penetrate into the demineralised collagen matrix.[9] This may adversely affect the effectiveness of PA as a dentine tissue modifier. Quercetin (QC) and naringin (NR) are both flavonoids, with molecular structures similar to PA (Fig. 1).[10] They both have the same catechin/epicatechin building units as PA, but have smaller molecular sizes.[10,11]

Quercetin belongs to the flavonol group and is commonly found in onions, apples, tea and red wine.[12] As a result of its cross-linking properties, QC has been shown to enhance the mechanical properties and thermal denaturation temperature of the extracelluar matrix of heart valves.[13,14] Naringin, which belongs to the flavonone glycoside group, is a flavonoid found mainly in grapefruit and other citrus fruits.[15] Naringin has been shown to increase local bone formation using a rabbit bone defect model.[16] It also promoted bone formation in a titanium particle-induced diabetic murine calvarial osteolysis model[17] and improved bone quality in orchidectomized rats.[18] Furthermore, NR also has an inhibitory effect on osteoclast formation and bone resorption by inhibiting RANK-mediated NF-κB and ERK signalling.[19] The hydroxyl groups in flavonoid molecules form hydrogen bonds with amide groups of protein molecules.[20] Cross-linking of protein molecules by flavonoids occurs via this mechanism,[21] which enhances the biomechanical properties of the collagen matrix.[5,6,13,14]

Thus, the present study evaluated the effects of three flavonoids, PA, QC and NR on the modulus of elasticity and ultimate tensile strength of demineralised dentine matrix. The null hypotheses tested were that there was no difference among the three flavonoids on the mechanical properties of demineralised dentine.

2. Materials and methods

2.1. *Modulus of elasticity*

Thirty freshly extracted human caries-free third molars stored in 0.9% NaCl solution supplemented with sodium azide to prevent bacterial growth at 4 °C for no longer than six months were used in this study. The study was approved by the local Institutional Review Board (UW 11-242). The teeth were

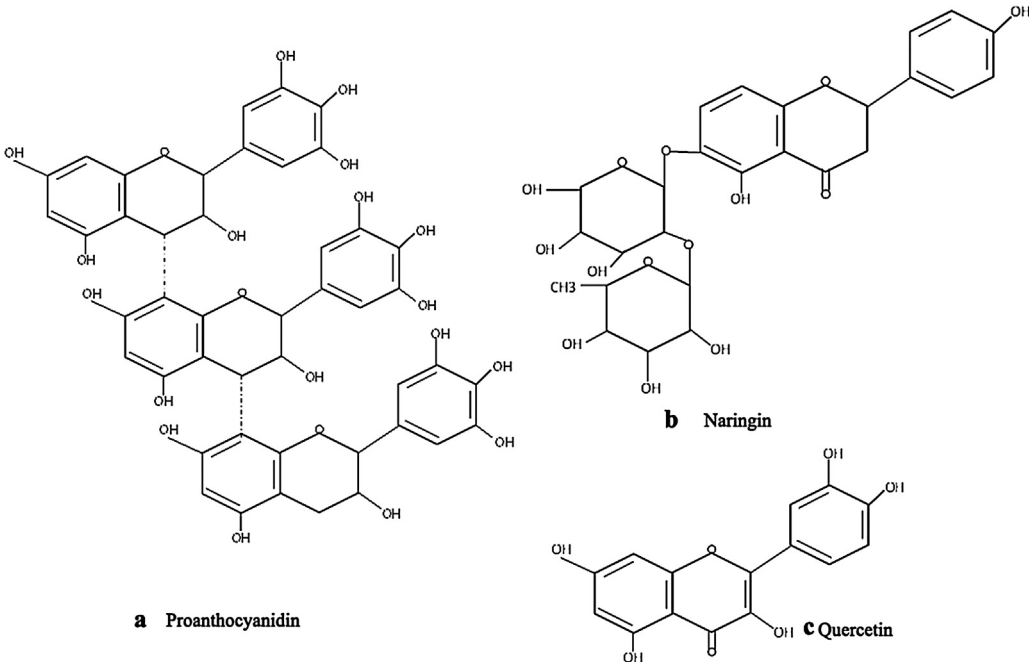

Fig. 1 – Chemical structures of the three tested flavonoids. (a) Molecular structure of proanthocyanidin. Multiple molecules are polymerized to form oligomeric molecules, (b) molecular structure of naringin and (c) molecular structure of quercetin.