Comparative Study on the Effect of Propolis and Sodium Fluoride on Exposed Dentinal Tubules

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List of Abbreviations

ACC	Arginine-Calcium Carbonate
ANOVA	Analysis of Variance
CO_2	Carbon Dioxide
CP	Carbamide Peroxide
CPP-ACPF	Casein Phosphopeptide- Amorphous Calcium Phosphate Fluoride
CPP-ACP	Casein Phosphopeptide-Amorphous Calcium Phosphate
DH	Dentin Hypersensitivity
DPSCs	Dental Pulp Stem Cells
EEP	Ethanolic Extract of Propolis
Er:YAG	Erbium: Yttrium Aluminum Garnet
FEG	Field Emission Gun
GaAIAs	Gallium–Aluminum–Arsenide Laser
GTFs	Glucosyltransferases
HBSS	Hank’s Balanced Salt Solution
HNSCC	Human Head and Neck Squamous Cell Carcinoma

IBM	International Business Machine
MTA	Mineral Trioxide Aggregate
NaF	Sodium Fluoride
Nd-YAG	Neodymium: Yttrium Aluminum Garnet
(p)	Probability
PDL	Periodontal Ligament
PG	Placebo Gel
RPE	Red Propolis Extract
S.D.	Standard Deviation
SEM	Scanning Electron Microscope
SPSS	Statistical Package for Scientific Studies
t-test	Two Sample Location Test

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Abstract

Dentin hypersensitivity (DH) occurs on exposed dentin and is dependent on the patency of dentinal tubules. Occlusion of tubules openings may decrease DH according to the hydrodynamic theory. **Aim of the study:** Evaluate and compare the effect of Propolis and NaF containing agent in occluding exposed dentinal tubules viewed by scanning electron microscope. **Materials and Methods:** 21 recently extracted caries-free human premolars were used. Teeth were sectioned with a diamond saw**,** perpendicular to the long axis of the tooth, to create dentin discs with a thickness of 1.0 ± 0.1 mm from the mid coronal dentin. All the specimens were etched with acid etch for 30 sec. to remove the smear layer, in order to expose the tubules. Dentin discs were divided into 3 groups, 7 dentin discs each, according to the treatment solution used as follows: Group I: pretreated with acid etch with no additional treatment. Group II: treated with NaF containing agent. Group III: treated with ethanolic extract of propolis (EEP). All samples were examined with SEM to evaluate the effect of the tested agents on occlusion of dentinal tubules and analyzed using ImageJ Digital Analyzer. The statistical analysis was performed using the arithmetic mean, S.D., unpaired t test and ANOVA. **Results:** Discs treated with NaF demonstrated tubular occlusion with crystal like deposits on the surface. Discs treated with propolis formed homogenous resinous layer and showed interlocking within the tubules openings. Statistical results showed no significant difference between group II and group III, regarding the number of dentinal tubules and the area percentage of open tubules occupied in the total area. **Conclusion:** Propolis was as effective as NaF in occluding dentinal tubules.

Introduction
and Review of Literature

Dentin hypersensitivity (DH) is a significant global clinical oral health problem in the adult population. It is one of the most commonly encountered clinical conditions in the day to day dental practice. DH is defined as "short, sharp pain arising from exposed dentin in response to stimuli typically thermal, evaporative, tactile, osmotic or chemical, which cannot be ascribed to any other dental defect or pathology". It is a symptom that arises from exposed dentin which causes pain that may disturb a person during eating, drinking, tooth brushing, and sometimes even breathing. It changes the quality of life of people, because of the pain experienced which causes severe and frequent discomfort ***(Addy, 1990; Miglani et al., 2010 and Naidu et al., 2014).***

Generally, a slightly higher prevalence of DH has been reported in women than in men and all ages can be affected. However, epidemiological data suggest that DH is more often diagnosed in individuals from 20 to 50 years of age, with the peak occurrence found at the end of the third decade. This observation may be attributable to the natural processes of aging. It was also reported that maxillary teeth seem to be more affected than the mandibular teeth ***(Splieth & Tachou, 2013)***.

In the literature, it is reported that DH prevalence ranges from 4 to 57%, depending on the population samples studied, the difference in the selection criteria used for each study and the diagnostic approaches. In patients affected by periodontitis, DH prevalence was even higher ranging between 60 to 98%. However, DH prevalence is likely to increase in next years since more adults keep their teeth into later life ***(West et al., 2013b and Haneet & Vandana, 2016).***

Histology of dentin pulp complex

Dentin is the most voluminous structural component of human tooth. It protects pulp tissue from microbial and other noxious stimuli. It also provides essential support to enamel and enables highly mineralized and fragile enamel to withstand occlusal and masticatory forces without fracturing. Furthermore, it is the first vital tissue to meet external irritation, and instead of being merely a passive mechanical barrier, dentin may in many ways participate in dentin–pulp complex defensive reactions ***(Tjäderhane et al., 2012).***

Dentin tissue is formed of the odontoblasts and their odontoblastic processes which secrete dentin matrix building the dentinal tubules, the peritubular and the intertubular dentin. The functional cellular unit of the dentin complex is the odontoblast. This cell is responsible for the main dentinal functions: dentin formation; including protein secretion and mineral deposition, and with the dentinal fluid, it helps in sensation to painful stimuli. And more importantly, dentin is capable of repair as odontoblasts can be stimulated to deposit more dentin as the occasion demands ***(Garcés-Ortíz et al., 2013).***

Odontoblastic processes run in canaliculi that traverse the dentin layer, which are known as "Dentinal Tubules". They are actually the extensions of odontoblasts. The presence of the tubules in dentin makes the tissue permeable, especially when the outer protective layer of enamel or cementum is removed. The odontoblastic processes are surrounded by dentinal fluid inside the tubules. Dentinal fluid forms around 22% of total volume of dentin ***(Orchardson & Cadden, 2001).***

Dentinal tubules are tapered structures measuring approximately 2.5 µm in diameter near the pulp, 1.2 µm in the mid portion of the dentin and 900 nm near the dentino-enamel junction**.** The number of dentinal tubules per mm^2 varies markedly depending on the location within the dentin. The normal range in tubule density per mm^2 is from about 5,000 to 90,000. However, the number of tubules in the coronal part of the dentin layer which is close to the pulp is 45,000-65,000/mm^2; this number is higher if compared to the outer dentin areas (15,000–20,000/mm^2) ***(Harran Ponce et al., 2001 and Nanci & Cate, 2008).***

Due to the difference in the surface area of the dentin at the amelo- and cemento-dentinal junction and that at the predentin border in the fully formed and functional teeth, the tubules will be more densely packed on the pulpal aspect of the dentin than peripherally. The highest density of tubules is found at the pulpal aspect subjacent to occlusal fissures, incisal edges and cusp tips. The lowest density of tubules is found peripherally and it does not vary much in the different parts of the tooth ***(Mjör & Nordahl, 1996).***

Pulp is integrally connected to dentin, i.e., physiologic and/or pathologic reactions in dentin will also affect the pulp. Innervation of the pulp and dentin is linked by the fluid and by its movement between the dentinal tubules and peripheral receptors, and thus to the sensory nerves of the pulp proper ***(Pashley et al., 2002).***

Mechanism of dentin sensitivity

Three theories have been proposed to explain the mechanism of action of dentin sensitivity: the transducer theory, the neural theory and the hydrodynamic theory. Of these theories, the hydrodynamic theory is currently believed to be the most responsible for the transmission of dentinal sensation ***(Yadav et al., 2015).***

1. Transducer theory

This theory was suggested and termed as the "odontoblast transducer" mechanism. Based on that odontoblasts are embryologically of neural crest derived mesenchymal cells and can act as receptor cells mediating changes via synaptic junctions with nerves. This theory of dentinal sensation takes into consideration the "synaptic like" relationship between the odontoblastic processes. This could result in the sensation of pain from the nerve endings located in the pulpodentinal border ***(West et al., 2013a).***