

Evaluation of Photosensitizers on Root Canal Disinfection and Bonding Interface of Fiber Post Cementation System: An *In Vitro* Study

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ABSTRACT

Aim and objective: The current study aimed to assess and compare the effectiveness of photosensitizers rose Bengal (RB), riboflavin, and curcumin (CP) to the conventional canal disinfection regime sodium hypochlorite (NaOCl) on the bonding interface of a fiber post cementation system.

Materials and methods: Sixty nontraumatic, extracted, and closed apex human mandibular second premolars were gathered and disinfected. All specimens were decoronated and embedded perpendicularly in heat cure acrylic resin. Shaping and cleaning of the canal were done and obturated with gutta-percha and sealer. Using peeso-reamer canal space was prepared. To assess the effectiveness of various disinfectants, post space was cleansed with four different types of disinfectants ($n = 15$) in each group. Group I riboflavin + 17% mixture of tetracycline, acid, and detergent (MTAD); group II CP + 17% MTAD; group III RB + 17% MTAD; and group IV 2.5% NaOCl + 17% MTAD. Fiber post was luted within radicular dentin of each sample with dual-cure self-etch resin cement. The specimen's radicular portions were vertically segregated into apical, middle, and coronal dentinal post portions and positioned over the universal testing machine. Modes of failure were assessed. Analysis of variance (ANOVA) was applied for the means of independent unrelated groups. Mean differences were calculated using Tukey multiple comparison tests ($p = 0.05$).

Results: Group II canal disinfected with CP + 17% MTAD at all three levels of root demonstrated the highest PBS score. Group IV (control) in which samples were disinfected by the conventional method (2.5% NaOCl + 17% MTAD) showed the lowest PBS at all root portions. Intergroup comparison unveiled PBS at all three root levels for group II (CP + 17% MTAD) and group I (riboflavin + 17% MTAD) than group III (RB + 17% MTAD) ($p > 0.05$). The intragroup assessment demonstrated a significant inclination in values of PBS from coronal to apical direction in all examined groups.

Conclusion: The use of photosensitizers curcumin, rose Bengal, and riboflavin as canal disinfectant demonstrated better PBS compared to the conventional method of canal disinfectant NaOCl at all three root levels coronal, middle, and apical.

Clinical significance: In a nonvital tooth, radicular disinfection is necessary. Loss of structure in endodontically treated teeth requires post which improves strength and prognosis of treatment. The bonding of glass fiber post to radicular dentin necessitates good clinical outcomes.

Keywords: Curcumin, Riboflavin, Rose Bengal, Photodynamic therapy, Pushout bond strength.

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INTRODUCTION

Restoration of nonvital teeth with extensive coronal destruction is a foremost challenge for dental personnel as it involves distinctive knowledge of endodontics, periodontics, prosthodontics, and operative dentistry.¹ An absolute eradication of endodontic microbiota from the canals is imperative to prevent reinfection and uphold the longevity of the restoration.²

Endodontically treated teeth possess a considerable loss of tooth structure which can be strengthened by post and core implementation for structural restoration and occlusal load resistance.³ Pre fabricated fiber reinforced composite (PFRC) posts are progressively accepted for repair and consolidation of the structural foundation of root canal-treated teeth plummeting the risk of root fracture.^{1,4} During instrumentation of smear layer (3–15 μm thick), a constitution of dentin chips, microbial colonies, salivary protein, and dentinal collagen may prevent adhesive-bulk dentin affiliation, in turn, compromising the bond strength and sealing capacity.⁵ Thus, removal of smear layer and other associated bond-weakening factors should be assessed properly and suitable root space desensitization should be executed.⁶

Disinfectants have been used for effective cleansing of radicular canal space.⁷ Sodium hypochlorite (NaOCl), a

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remarkable nonspecific proteolytic negotiator and traditional cleanser upsurge the solubilization of organic components during the endodontic procedure.⁸ However, the resin cement's polymerization reaction and resin-infiltrated hybrid layer formation are altered due to NaOCl's oxidation process, thus disturbing the post-dentin bond efficacy.⁹ Apart from NaOCl, a cross-linking agent exhibiting antioxidative ability "riboflavin" has also been recommended for dentin-post

bond fortification.^{10–12} However, the application of this recent approach is still not operated as a canal disinfectant.

An unparalleled and innovative endodontic canal disinfecting approach “photodynamic therapy” utilizes varied photosensitizers, activated to definite wavelength emits reactive oxygen species (ROS), and potentiates photo-activated cell death maximizing canal decontamination.^{13,14} An anionic, polyphenolic hydrophobic compound “curcumin photosensitizer (CP)” is proved to be a puissant canal disinfecting dye, enhancing dentin collagen–post bond strength by its antioxidative action.⁷ Besides CP, rose Bengal (RB) a halogen-derived photosensitizer from iodine fluorescein convincingly embraces an eminent antimicrobial behavior at a concentration of 5–10 µg/mL, sustaining dentinal structure and fiber post bond integrity.¹⁵

Nonetheless, it has been demonstrated from current indexed literature that studies on the disinfection of radicular dentin using different photosensitizers are scarce and doubtful. The prevalent data’s heterogeneity sanctions the need to perform *in vitro* exploration, paralleling several endodontic disinfection protocols and their influence on dentin–post bond strength. Accordingly, it has been hypothesized that 2.5% NaOCl will unveil superior radicular dentin disinfection property and will demonstrate the highest push out bond strength (PBS) in contrast to different photosensitizers (curcumin, rose Bengal, and riboflavin). Therefore, the present study aimed to assess and compare the effectiveness of photosensitizers to the conventional regime (NaOCl) on PBS of PFRC post cemented to root dentin.

MATERIALS AND METHODS

The current study design anticipated the period of 3 months (90 days) from June to August 2021 and complied with the guidelines of checklist reporting *in vitro* study (CRIS). During this period of 90 days, 60 nontraumatic, closed apex human mandibular second premolars were extracted for orthodontic and periodontal reasons were considered. Attached periodontal fibers, plaque, or calculi were disengaged with the ultrasonic scaler (Woodpecker Dental Ultrasonic Piezo Scaler UDS-J, Zodex, India) then disinfected with the Chloramine T trihydrate (Sigma, Aldrich, London, UK) solution for 48 hours at 4°C. Cleansed specimens were decoronated via diamond bur (Medelec Instrument, New Delhi, India) under irrigation preserving 12 mm of radicular portion. All samples were embedded perpendicularly in heat cure acrylic resin (Acrylic Heat Cure, Mr. Dental, UK) using a Teflon mold (4 mm radius) to perform an endodontic procedure.

Canal shaping and cleaning was performed with a patency K-file #10 (Premier Dental Products Romano Drive, Plymouth Meeting, Pennsylvania) 1 mm less of working length (apical constriction) and broadened till 25 K-file. Further root space expansion and finishing of canals were premeditated by protaper universal NiTi system (Intellodent, Rampur, Roorkee, Dist. Haridwar) using the crown down method simultaneously, rinsing with 1% NaOCl. Drying of prepared canals was done with paper points (Diadent Paper Point Pro T, Hundal Dental Traders, India) and successively obturated with gutta-percha (Dentsply Protaper Universal Gutta Percha Points, India) and sealer Adseal (Resin Based Sealer S.M. Overseas, India). All specimens were preserved in 100% humidity for 7 days at 37°C.

Post space was prepared after partial removal of gutta-percha (approximately 6 mm) via peeso reamers (World Link Traders Sahpurjat, New Delhi). To assess the effectiveness of various disinfectants, post space was cleansed with four different types of disinfectants ($n = 15$) in each group.

Group I: Riboflavin + 17% MTAD

In group I, the canals of each specimen were disinfected with 6 mL of 1% riboflavin, injected via an open-ended needle 27 gauge for 120 seconds, photo-activated for 20 seconds with a wavelength of 660 nm with 150 mW power and 23.43 J/cm² power density, and then washed with 17% mixture of tetracycline, acid, and detergent (MTAD) and air-dried without desiccation.

Group II: CP + 17% MTAD

For 180 seconds, 2.5 mg/mL solution of CP dye was injected into the canal and activated via light-emitting diode (LED) curing unit (LED Rainbow Curing Light, New Delhi, India) at 1600 mW/cm intensity and wavelength of 480 nm. Subsequently, the canals were rinsed with 17% MTAD and then dried off.

Group III: RB + 17% MTAD

In group III, samples were cleansed with 5 µM RB (Innovative, Kalbadevi, Mumbai, Maharashtra) and photo-irradiated for 180 seconds exploiting red-light-emitting diode (LED) at a wavelength of 480 nm and 200 mW power yield and power density of 526 (mW/cm²) then sanitized via 17% MTAD for 60 seconds and dried rigorously.

Group IV: 2.5% NaOCl + 17% MTAD

In group IV, 5 mL of 2.5% NaOCl (Hipos Sodium Hypo Chlorite, Suvehdi Industries, India) for 1 minute was irrigated in the control group for cleansing the canals. For smear layer removal, 5 mL of 17% MTAD for 1 minute via 30 minutes gauge needle irrigating syringe was used.

After successive photo-irradiation and conventional cleansing, the canals of all investigated samples were washed with distilled water and dried with paper points (Diadent Paper Point Pro T, Hundal Dental Traders, India). Repeatedly, PFRC post was checked and placed within the sterilized post space. PFRC was cleaned with 70% ethyl alcohol before luting, then dried and luted with Rely X Unicem (Self-Etch Resin Cement, 3M, USA) dual-cure self-etch resin cement and finally polymerized using a light-emitting diode curing light (iLed—Guilin Woodpecker Medical Instrument Co Ltd, UK) within the allotted space. For 72 hours after post-cementation, all specimens were deposited in a 100% temperature-controlled setting at 37°C before evaluating and comparing PBS. All samples were thermocycled (1100/THE-1200—SD Mechatronik) at 5–60°C closely 10,000 cycles with a dwell time of 40 seconds.

Examination of PBS

The specimen’s radicular portions were vertically segregated into apical, middle, and coronal dentinal post portions 30 slices using a diamond bur (Medelec Instrument, New Delhi, India) under a constant water coolant mechanism. These sliced roots were introduced into a specific jig, associated with universal testing equipment (Testometric UK, Great Britain, UK) in metallic mold subjected to increasing load at a crosshead speed of 1 mL/minute to assess the PBS in the apical-cervical direction. The formula used to calculate the PBS in megapascals (MPa) is stated below

$$\text{Debond stress} = N/A$$

where,

N signifies force that results in post–dentin bond failure

A represents the area taken off the cemented post

Investigation of Failure Modes

A stereomicroscope at 40× magnification (ESAW Advance Student Binocular Microscope, The Engineering Science Apparatus Workshop,

India) was utilized for fracture analysis. Modes of failure were assessed and categorized as adhesive, cohesive, and admixed.

Statistical Analysis

Analysis for shear bond strength (SBS) and failure mode was performed using a statistical program for social science (IBM SPSS Statistics 21.0). Comparison of means and standard deviations (SD) of investigated groups was performed using analysis of variance (ANOVA) for intergroup comparison. Tukey's *post hoc* test for multiple comparisons ($p = 0.05$).

RESULTS

In the present study, the proportionate distribution of data was achieved by Levene's test. Table 1 and Figure 1 reveal PBS values means and standard deviation (SD) among investigated groups at all three root levels. The maximum PBS scores were observed in group II canal disinfected with CP and 17% MTAD at all radicular levels, coronal (8.83 ± 0.41), middle (7.87 ± 0.45), and apical (5.35 ± 0.31). Group IV (control) in which samples were disinfected by conventional method (2.5% NaOCl + 17% MTAD) showed the lowest PBS at all root portions, (6.12 ± 0.54), (5.46 ± 0.84), and (3.00 ± 1.88), respectively.

The intragroup assessment demonstrated a significant inclination in values of PBS from crown to apex in all investigated groups. No significant difference in PBS was observed in cervical and middle root thirds ($p > 0.05$), excluding the apical third ($p < 0.05$).

Intergroup assessment showed similar PBS at all root levels (coronal, middle, and apical) for group II (CP + 17% MTAD), group I (riboflavin + 17% MTAD) than group III (RB + 17% MTAD) ($p > 0.05$). Nevertheless, a noteworthy difference was observed in group IV 2.5% NaOCl + 17% MTAD (control) at the coronal level compared to other experimental groups at the same level ($p < 0.05$).

Outcomes of failure analysis of fiber post–dentin bond among different investigated groups were displayed in Figure 2. The most prevalent failure mode was adhesive (luting cement–dentin collagenous network bond) followed by the admixed type failure. However, lowermost failure percentages were revealed by cohesive (cement–fiber post bond).

Better PBS was noted in all photosensitizers (RB, CP, and riboflavin) when used as a cavity disinfectant with adhesive failure type.

Table 1: PBS values in means and standard deviations (SD) among different investigational groups at three root levels coronal, middle, and apical

Examined groups	Cervical (MPa ± SD)	Middle (MPa ± SD)	Apical (MPa ± SD)
Group I: Riboflavin and 17% MTAD	$8.15 \pm 0.04^{c,D}$	$7.11 \pm 0.76^{c,D}$	$5.33 \pm 0.41^{c,H}$
Group II: CP and 17% MTAD	$8.83 \pm 0.41^{c,D}$	$7.87 \pm 0.45^{c,D}$	$5.35 \pm 0.31^{c,H}$
Group III: RB 5 μM and 17% MTAD	$7.99 \pm 0.24^{c,D}$	$7.24 \pm 1.11^{c,D}$	$5.64 \pm 0.34^{c,H}$
Group IV: 2.5% NaOCl and 17% MTAD	$6.12 \pm 0.54^{d,D}$	$5.46 \pm 0.84^{d,D}$	$3.00 \pm 1.88^{c,H}$

MTAD, a mixture of tetracycline, acid, and detergent; CP, curcumin photosensitizer; RB, rose Bengal; NaOCl, sodium hypochlorite; $p < 0.05$: statistically significant differences within the same column dissimilar superscript lowercase alphabets; $p < 0.05$: statistically significant differences within each row dissimilar uppercase alphabets

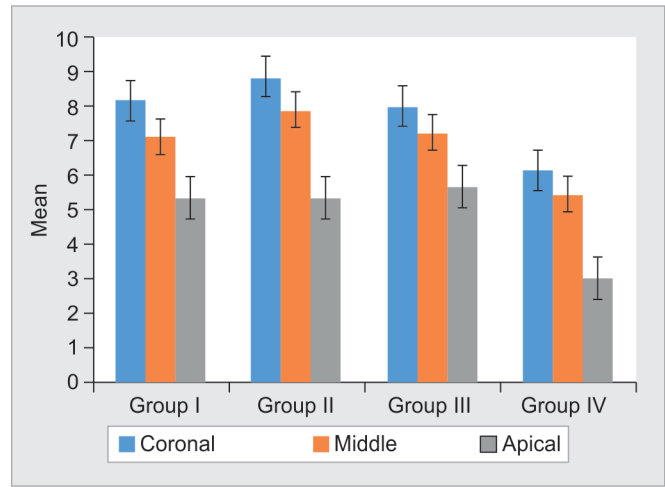


Fig. 1: PBS values in means and standard deviations (SD) among different investigational groups at three root levels coronal, middle, and apical

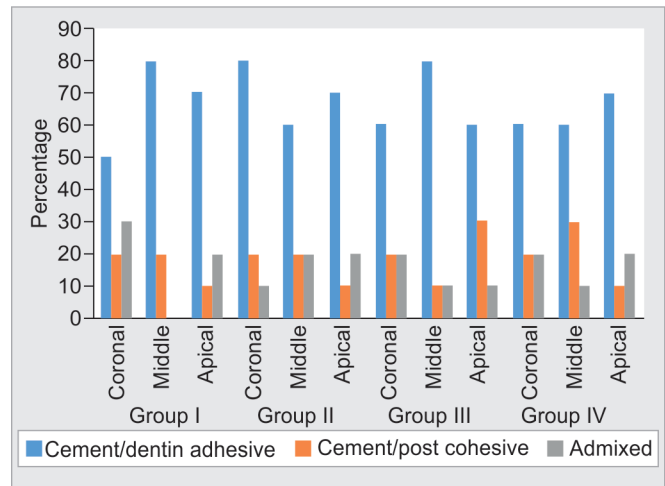


Fig. 2: Percentage of failure mood against different experimental groups

DISCUSSION

The present study intended to assess and compare the effectiveness of photosensitizers to the conventional regime (NaOCl) on PBS of PFRC post cemented to radicular dentin. The prevalent analysis was outlined on the reference that 2.5% NaOCl will unveil superior radicular dentin disinfection property and will demonstrate the highest PBS in contrast to different photosensitizers (CP, RB, and riboflavin). Nevertheless, reviewing the outcomes of our lab-based study, the supposition was completely rejected as it was manifested that PFRC post cemented to dentinal collagen of root via SERC has highest PBS when photo-activated with CP and lowest with NaOCl at all three root levels (coronal, middle, and apical) while riboflavin and RB also signified enhanced PBS.

The binding capacity of PFRC post to radicular dentin may also rely on the adhesive mechanical bond type and property.¹⁶ This study, therefore, preferred self-etch resin (SERC) cement for luting action. The SERC bonded to radicular dentin and post has low fracture resistance than conventional cement due to various organic resin monomers and inorganic fillers that upsurge the bond stability and maintain marginal integrity by reducing polymerization shrinkage.¹⁷ SERC application exhibits subsequent

properties: Minimum application time, notable flow rate, and deep tubular dentin penetration promoting effective bond.¹⁸ To examine the data related to the measurement of bond vitality at the interface of dentinal structure and resin matrix, a shear stress test of fiber pushout bond strength was executed. The test is reproducible and analyzes root-filled material's bond strength to root dentin at low levels through permeation of luting cement into the canal tubular dentin and collagenous network, thus augmenting adhesion, fracture resistance, and reduction in interfacial stress to overcome occlusal forces.^{19,20}

In the present study, the conventional method of disinfection demonstrated the lowest PBS at all three levels. An apparent elucidation to this finding is that NaOCl (2.25–5.25%) in water dissociates into Na^+ and hypochlorous acid (OCl^-) exhibiting robust antioxidative properties.²¹ Emission of oxygen (O_2) and chlorine (Cl^-) establishes O_2 enriched film over dentinal surface and destroys organic collagen with uneven obliteration of smear layer, therefore, diminishing the bond efficacy.^{2,22} To subjugate this, final cleansing with 17% MTAD (a calcium chelator) was encouraged in this study resulting in the removal of the smear layer with negligible erosion of dentinal tubules²³ and boosting mechanical merging of the cement into the canal walls, optimizing the bond.^{2,24} However, the use of MTAD in conjugation with NaOCl shows oxidative properties and diminishes the advantage of NaOCl making dentin less receptive to bonding compromising bond integrity. The result of the present study is in harmony with the findings of Paulson et al. who claimed that 2.5% NaOCl with MTAD as canal disinfectant showed low PBS due to substandard smear layer eradicating properties.²⁰

In credit to improved impregnation of SERC into the opened tubular dentin, smear layer should be eradicated proficiently to augment fiber post bond strength.^{25,26} The outcome of the current study confirmed the above rationale as samples in group II demonstrated the highest PBS when photo-irradiated with CP with 17% MTAD at all three root levels as compared to the conventional disinfecting method. CP as a canal disinfectant has been reckoned to have superior post space disinfection potentials as it releases singlet O_2 on activation, its hydrophobic nature, low wavelength photo liability, and anionic affinity for cationic molecules. A plausible validation for improved post-dentin bond efficacy to SERC.^{27,28} The findings are in accordant with the provisions of Hamdan et al. that curcumin has the highest PBS as being anionic, binding to calcium (Ca^{++}) in dentinal structure resulting in enhanced micromechanical attachment of the resin cement to the dentin.⁷ Canal disinfected with RB demonstrated comparable PBS to CP and riboflavin at all three root levels. Apparent explanations for this conclusion are the sensitivity, specificity, and disposition of the gram-positive bacteria against RB, the concentration of PS, the wavelength of LED light for provocation, and extent of the photo-irradiated period.^{15,29} However, the use of CP and RB on affected dentin and as an endodontic canal disinfectant still needs to be reconnoitered.

A reinforced reactive oxygen species (ROS) producer and a potent MMP inhibitor, "riboflavin" was used as a canal disinfectant in group I, which inherently promotes telopeptidase action and reinforces collagenous meshwork by photo-activation. Its superior cross-linking and biomechanical rigidity by 4.5 times more improves fiber post receptivity to dentin structure.¹⁰ These properties are coordinated with the study by Al-Kheraif et al., in which riboflavin displayed comparable PBS to CP.^{11,30}

Considering the results, PBS exhibited a declining tendency from coronal to apical third in all groups. Several factors may account for this, such as a distance of the curing light, nonpatent apical collagenous dentin (less sealer infiltration), dentinal tubule density, high c-factor, increased dissolution of apatite crystals, inadequate apical cleaning and shaping of the radicular portion having a discernable impact on bond integrity, and mechanical strength of dentinal apparatus.^{31–33} This is explained by the already published work Alireza et al., which specified that dentinal tubules are denser in the coronal and middle root areas incomparable to apical radicular dentin. This could be the probable cause for high PBS in the coronal compared to the apical root level.³⁴ Reflecting the outcomes of fracture mode analysis, adhesive failure at cement and dentin interface was the most common failure type followed by the admixed type. The number, structure, and concentration of dentinal tubules in different segments of radicular dentin are responsible for the bond failure tendency.

The present *in vitro* study is subjected to some inherent limitations; the results are due to a lack of topographical characterization of dentin. Configuration of dentin structure, dentin type and nature, the flow of tubular fluid, projections of odontoblast, smear layer, and quality of adhesive of cement momentarily may influence the current study's findings. Furthermore, scanning electron microscopy (SEM) and atomic force microscopy should be introduced to further evaluate the PBS of varying concentrations of NaOCl and disinfectant at different concentrations CP, RB, and riboflavin on different zones of radicular dentin.

CONCLUSION

The use of photosensitizers curcumin, rose Bengal, and riboflavin as canal disinfectant demonstrated better pushout bond strength compared to the conventional method of canal disinfectant NaOCl at all three root levels coronal, middle, and apical.

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