

# Antioxidants in Endodontics: A Strategic Review

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## ABSTRACT

Over the past few decades, the free radicals and antioxidants have attracted tremendous importance in the field of dentistry. Depending on the level of free radicals in the body, they can either be harmful or helpful. Antioxidants are the substances that interact with and stabilize free radicals thereby protecting cells from the damage caused by free radicals. The indigenous antioxidants may be useful in preventing the harmful effects of oxidative stress. The present article presents a review on the free radicals formation, their role in disease pathogenesis, antioxidants and their potential role in endodontics.

**Keywords:** Free radicals, Reactive nitrogen species, Reactive oxygen species

## INTRODUCTION

Oxygen is an element indispensable for life, can act both so as to promote and deteriorate health of the body. The poisonous effects of oxygen were unknown until Gershman's free radical theory of oxygen toxicity in 1954, which states that the toxicity of oxygen is due to partially reduced forms of oxygen [1]. When cells use oxygen to generate energy in the form of ATP in the mitochondria, free radicals are created [2,3]. Free radicals are chemically active atoms that have a charge due to an excess or deficient number of electrons. They can be reactive oxygen species (ROS) or reactive nitrogen species (RNS) [4].

ROS and RNS are well recognized for playing a dual role as both deleterious and beneficial species [5]. ROS are highly reactive molecules derived from oxygen metabolism. *In vivo*, some of these ROS play constructive roles in cell physiology; however, they may also cause great destruction to cell membranes and DNA, by causing membrane lipid peroxidation, decreased membrane fluidity, and DNA mutations leading to cancer, degenerative, and other diseases [6-9].

ROS levels increase intensely and result in significant damage to cell structures during oxidative stress. ROS are cytotoxic and have been implicated in the aetiology of various human diseases [10]. Thus, ROS can kill bacteria but it also destroys the adjacent infected host tissues [11].

ROS scavengers/ antioxidants are of prime importance for preventing and controlling human diseases by counteracting ROS formation. Antioxidants are helpful in preventing the harmful effects of ROS by reacting with oxygen [12].

## Sources of Free Radical Insults in Dental Therapy [4]

In dentistry, many commonly used dental materials may form free radicals like.

1. Bleaching agents
2. Composite fillings
3. Dental cements
4. Ceramic restoration
5. Metals in restoration
6. Dental implants
7. Intracanal medicament

## Classification of anti-oxidants [13]

1. **Enzymatic:** Superoxide dismutase, Glutathione Peroxidase, Selenium, Catalase, Glutathione reductase, Glutathione transferase
2. **Non-enzymatic:** They are further subdivided into two.
  - Nutrient:** Alpha tocopherol,  $\beta$ - Carotene Ascorbate, Glutathione, Selenium, Proanthocyanidin, Lycopene, Green tea
  - NonNutrient:** Ceruloplasmin, Transferrin, uric acid, Peptides Camosine Anserine

Vitamin E, flavonoids, catechins, gallic acid derivatives, salicylic acid derivatives, cinnamic acid derivatives chlorogenic acid, resveratrol, folate, curcumin, caffeine, anthocyanins and tannins are examples of polyphenolic natural antioxidants derived from plant sources.

Non-phenolic secondary metabolites such as melatonin, carotenoids, retinal, thiols, jasmonic acid, ei-cosapentaenoic acid, ascopyrones and allicin that show excellent antioxidant activity [9].

## Mechanism of Action of Antioxidants [13]

Antioxidants end the electron stealing reaction of free radicals by donating one of their electrons. The antioxidant does not become a free radical by donating an electron because they are stable in either form. Antioxidants act by scavenging or chain breaking, such as Vitamin E (alpha tocopherol), Vitamin C (ascorbic acid), or Vitamin A (beta carotene) and preventative antioxidants that function basically by sequestering transition metal ions and preventing fenton reactions and are therefore predominantly proteins by nature (e.g., albumin, transferring, or lactoferrin).

## Commonly used antioxidants are:

### Carotenoids

Carotenoids belong to the tetraterpene family and over 600 natural structural variants exist. Of these only about 20 are found in human plasma and tissues, including lycopene,  $\alpha$ -carotene,  $\beta$ -carotene, lutein, cryptoxanthine, retinol (vitamin A1) and dehydroretinol (vitamin A2) [4].

Lycopene is one of the most potent antioxidants primarily present in tomatoes [14]. Lycopene, the red pigment of tomato, is a tetraterpene assembled from eight isoprene units composed entirely of carbon and hydrogen, containing 11 conjugated and two non-conjugated carbon-carbon double bonds [15]. Mageshwaran et al., determined the neutralizing effect of proanthocyanidin (grape

seed extract) and lycopene (tomato extract) on reactive oxygen species (ROS) generated by the mixture when used as an intra canal medicament. Lycopene showed more antioxidant potential than proanthocyanidin [16].

### Alpha-Tocopherol

Vitamin E is generally considered the most important and effective lipid soluble antioxidant, maintaining cell membrane integrity from lipid and decreasing wound-healing time [17]. Invitro study done by Sasaki RT, Flório FM, Basting RT revealed the shear bond strength of human enamel and dentin submitted to a bleaching treatment with 10% carbamide peroxide and treatment with antioxidant agents containing 10%  $\alpha$ -tocopherol and 10% sodium ascorbate formulated in solution and gel. Results showed antioxidant treatment with 10%  $\alpha$ -tocopherol solution was the only effective agent to revert the oxidizing effects of the bleaching treatment on enamel [18]. Other studies evaluated and compared composite bond strength to carbamide peroxide bleached enamel following the application of 10% and 25% sodium ascorbate and alpha-tocopherol solutions. Result showed 10 min application of 10% and 25% sodium ascorbate and 25% alpha tocopherol solutions significantly improves the shear bond strength of composite resin to enamel [19]. Alpha tocopherol allowed free-radical polymerization of the adhesive resin to proceed without premature termination by restoring the altered redox potential of the oxidized bonding substrate and improve resin bonding [18,20].

### Proanthocyanidin (Grape Seed Extract)

Oligomeric proanthocyanidin complexes (OPCs) are primarily known for their antioxidant activity. However, these compounds have also been reported to demonstrate anti-bacterial, anti-viral, anti-carcinogenic, anti-inflammatory, anti-allergic, and vasodilatory actions [21]. Manimaran et al., studied the application of proanthocyanidin agent (PA) that improves the bond strength of root dentin treated with sodium hypochlorite (NaOCl). The results demonstrated that 5.25% NaOCl caused significant reduction ( $p < 0.05$ ) in the bond strength, but this can be reversed by 5% PA significantly more than the 10% sodium ascorbate [22]. Study done by Abraham et al evaluated the effect of grape seed extract (oligomeric proanthocyanidin complexes {OPCs}) on the bond strength of composite resin to bleached enamel using 5<sup>th</sup> and 7<sup>th</sup> generations of bonding agents. Results showed that use of grape seed extract as an antioxidant after bleaching significantly improves the bond strength of composite resin to bleached enamel. 5<sup>th</sup> generation bonding agents have higher shear bond strength of composite resin to enamel [23]. A study investigated the long-term resin-dentin bond strength of dentin biomodified by proanthocyanidin-rich (PA) agents. Higher  $\alpha$ BS were observed for 6.5% grape seed extract (GSE) immediately (SB- 62.9MPa; OS- 51.9MPa) when compared to 6.5% cocoa seed extract ethanol-water (CSE-ET) (SB- 56.95MPa; OS- 60.28MPa), 6.5% cocoa seed extract acetone-water (CSE-AC) (SB- 49.97MPa; OS- 54.44MPa), and distilled water (CO) (SB- 52.0MPa; OS- 44.0MPa). GSE and CSE-ET agents provided enhanced immediate adhesion and stabilization to demineralized dentin after long-term storage, depending on adhesive system [24].

### Ascorbate

L-ascorbic acid (AA) is the white to light-yellow, water-soluble vitamin commonly known as vitamin C [25]. Sodium ascorbate (SA) is a salt of AA. Both AA and SA have antioxidant properties [26]. It is known that AA enhances the dentin bonding strength of adhesive resins when used on dentin surfaces as an experimental conditioner for C&B Metabond [25]. SA is known to enhance the bonding strength of adhesive resins on dentin surfaces treated with sodium hypochlorite (NaOCl) in endodontic and operative dentistry [27-31]. Reducing agents have the ability to donate two high-energy electrons to scavenge the free radicals that are formed during resin

polymerization [31]. Shrestha et al., investigated the effect of SA on degree of conversion (DC) and bond strength (BS) of RealSeal SE to sodium hypochlorite (NaOCl) treated root dentin. Results showed that NaOCl negatively affected DC and BS of RealSeal SE, which could be reversed with 10% SA [32]. Vongphan N et al studied the microtensile bond strengths of total etching adhesive systems to pulpal chamber wall dentine after treated with various irrigants. They concluded that SA significantly improved the bond strengths of NaOCl treated root dentine [33].

The bleaching process adversely but transitionally reduces enamel and dentin bond strength when immediate bonding is performed after bleaching [34-40]. Recent studies have shown that the reduction in bond strength can be reversed as a result of the application of SA as a biocompatible antioxidant agent [31,41-45].

Mazaheri and his co-workers concluded that application of 10% SA hydrogel or 1 week period elapsed after bleaching could significantly increase the enamel bond strength to a normal value [46]. Dabas et al., evaluated the effect of different concentrations of hydrogel of SA on bond strength of bleached enamel for varying periods of time and the mode of failure. They showed that there was no difference in bond strength with an increase in the concentration of SA hydrogel [47]. Morris MD et al., evaluated the effect of 5% NaOCl and RC-Prep treatment on the bond strength of resin cement, C&B Metabond. The results demonstrated that both 5% NaOCl and RC-Prep produced significantly ( $p < 0.05$ ) large reductions in resin-dentin bond strengths, and the reductions could be completely reversed by the application of either 10% ascorbic acid or 10% sodium ascorbate [28].

Prasansuttiporn T and his co-workers evaluated the effect of 10% sodium ascorbate solution, 100 $\mu$ M rosmarinic acid solution or Accel for 5 or 10s on microtensile bond strengths to sodium hypochlorite-treated dentin. The application of sodium ascorbate solution for 5 or 10 s did not significantly increase the compromised bonding to sodium hypochlorite-treated dentin ( $p > 0.05$ ). On the other hand, Accel and rosmarinic acid solution had significant reversal effects with the same application times [48]. Mohammadi Z studied effect of ascorbic acid on the antibacterial substantivity of Tetraclean in bovine root dentin pretreated with sodium hypochlorite (NaOCl). Results showed that Ascorbic acid prevents the decrease of residual antibacterial activity of Tetraclean in dentin samples pretreated with NaOCl [49].

Khoroushi and Aghelinejad compared the shear bond strength (sbs) of three different adhesives on bleached enamel immediately after bleaching, bleached/delayed for 1 week, and bleached/applied SA as antioxidantizing agent. Bond strength increased upto 25-35% higher than the initial bond strength subsequent to bleaching and the application of an antioxidant in their study [50].

Miranda and his colleagues found that bonding strength to bleached enamel was immediately restored with the application of sSA and exposure to human saliva in situ for at least seven days. Treatment with SA gel for 60 minutes may be recommended in cases patient cannot wait at least 7 days for adhesive techniques to be performed [51]. Khoroushi and Saneie compared the shear bond strength (SBS) of three different adhesives on bleached dentin immediately after bleaching, bleached/delayed for one week, and bleached/applied antioxidantizing agent. The findings suggest that bond strength of resin to bleached dentin may be affected with the adhesive system. Reduced SBS to bleached dentin can be amended by the use of SA as an antioxidantizing agent [52]. Muraguchi and his colleagues examined the effects of bleaching on the bond strength of an adhesive to bovine tooth surfaces; and explored the effectiveness of ascorbic acid application in preventing the deterioration of bonding ability due to bleaching. Results of this study suggested that ascorbic acid application was effective in preventing the reduction of bonding ability to bleached teeth [53].

Parnian Alizadeh Oskoei and his co-workers evaluated the

effect of 10% SA on bleached bovine enamel morphology and microhardness considering the possibility of its effect on enamel surface characteristics. The use of 35% carbamide peroxide alone or in conjunction with 10% SA does not have any detrimental effect on bovine enamel microhardness [54]. Eda Güler et al investigated the effect of SA on the microtensile bond strengths (MTBSs) of different composites to bovine enamel after vital bleaching with hydrogen peroxide (HP) or carbamide peroxide (CP). The application of 10% SA immediately after bleaching with 16% CP or 35% HP increased the enamel MTBS, regardless of the adhesive/composite resin used. They concluded that the application of SA for 10 minutes immediately after vital bleaching increases the enamel bond strength for dimethacrylate- and silorane-based composites [55]. Park et al., determined appropriate application duration of sodium ascorbate (SA) antioxidant gel in reducing microleakage of bonded composite restoration in intracoronaally-bleached teeth. Application of SA gel for three day after nonvital bleaching was effective in reducing microleakage of composite restoration in intracoronaally-bleached teeth [56].

## CONCLUSION

We see numerous studies advocating the use of antioxidants based supplements for their clinical usage in endodontics. Ascorbate, alpha-tocopherol, proanthocyanidin are one of few which are used till date. Yet further research and studies has to be done to know optimum level of anti-oxidants and their safest application in routine endodontic practice.

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