Critical Review
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Bonding efficiency and durability: current possibilities

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Abstract: Bonding plays a major role in dentistry nowadays. Dental adhesives are used in association with composites to solve many restorative issues. However, the wide variety of bonding agents currently available makes it difficult for clinicians to choose the best alternative in terms of material and technique, especially when different clinical situations are considered. Moreover, although bonding agents allow for a more conservative restorative approach, achieving a durable adhesive interface remains a matter of concern, and this mainly due to degradation of the bonding complex in the challenging oral environment. This review aims to present strategies that are being used or those still in development which may help to prevent degradation. It is fundamental that professionals are aware of these strategies to counteract degradation as much as possible. None of them are efficient to completely solve this problem, but they certainly represent reasonable alternatives to increase the lifetime of adhesive restorations.

Keywords: Dentin-Bonding Agents; Dental Cements; Adhesives; Dental Enamel.

Introduction

The advent of adhesives and the understanding of their interaction mainly with dentin have recognizably become landmarks for the practice of operative and conservative dentistry. In addition to direct resin restorations of teeth compromised by fractures, carious or non-carious lesions, adhesives enable several other procedures, such as bonding of indirect restorations, intra radicular posts and orthodontic brackets, repair of failed restorations, control of dentin hypersensitivity and correction of aesthetic impairments.

For a long time, amalgam was the material of choice for directly restoring posterior damaged teeth, leading to the preparation of large and geometric macro-retentive cavities. However, as recently presented by Alexander et al.,[1] the United Nations Environment Programme (UNEP), supported by the World Health Organization (WHO), has urged for policies that could play down the use of mercury, and consequently of dental amalgam. This trend was also corroborated by the Minamata Convention, which strengthened the so-called phase-down of amalgam.[2] Despite many controversies about its suitability as a real alternative to amalgam, resin composites gradually turned out to be the most indicated restorative material, also for posterior

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teeth. This procedure, however, always requires an intermediate bonding agent which penetrates enamel and/or dentin, primarily establishing what is called micromechanical bonding.[3] Basically, the adhesive procedure consists of removing minerals from the dental substrate by acid etching it to create micro-retentive porosities, where resin monomers infiltrate and polymerize.[4,5,6] Specifically, on dentin, the acid etching procedure not only removes possible superficial debris, the so-called smear layer, but also exposes a net of collagen fibers besides opening the dentin tubules. By penetrating this collagen mesh and dentin tubules, the infiltrating resin will form two well-defined structures which are known as hybrid layer and resin tags, respectively. This polymer-collagen biocomposite layer is in large part responsible for the bonding effectiveness, which does not actually rely on its thickness but rather on its quality.[7] In other words, an effective bonding relies mostly on the ability of the bonding agent to completely infiltrate the exposed collagen mesh, ideally sealing and protecting it from all sorts of degradation pathways. In the same way, it has been suggested that the bonding effectiveness does not depend on the number nor on the length of resin tags.[7]

Contemporary adhesives are formally categorized into two main types according to their mechanism of action, namely etch-and-rinse and self-etch systems.[4,5,8] The first requires prior application of phosphoric acid as an initial step. After mandatory rinsing, smear layer is completely removed and the tooth substrate is demineralized, resulting in profound pores in the highly-mineralized enamel and exposed collagen mesh in dentin in a depth that may range from 5 μm to 10 μm.[4,5,9,10] When using etch-and-rinse systems, proper hybrid layer is classically achieved through the infiltration of resin monomers into the exposed collagen mesh by using the so-called wet-bonding technique. In this protocol, water remaining from the rinsing step maintains the collagen network expanded, allowing resin monomers to properly infiltrate it; excessive dehydration would cause fibrils to collapse, impairing resin penetration and hybrid layer formation.[11,12,13] However, it is noteworthy that the wet-bonding technique is a major challenge for clinicians, who do not count on ultimate parameters to determine ideal moisture. Although it is suggested that dentin should clinically present a shiny aspect, this judgement is subjective and not precise enough.[11,12] Etch-and-rinse adhesives are subcategorized according to their presentation, more specifically according to the number of steps needed to accomplish the adhesive protocol. Three-step systems are comprised of etchant, primer and bonding resin while their two-step counterparts consist of etchant and a single bottle containing both primer bonding resin chemical components.[14] Primers contain water, ethanol, and/or acetone as solvents which act dissolving hydrophilic monomers such as HEMA, for example. While chasing water from the wet tooth substrate, they prevent collagen network from collapsing, therefore allowing for proper impregnation of the solvent-free, hydrophobic bonding resin.[4,6]

Conversely, self-etch adhesives contain monomer molecules with carboxylate or phosphate acidic groups which concurrently etch and infiltrate dental substrates in such a manner that smear layer is not removed, but incorporated into the adhesive interface.[5,15,16] Even if the thickness of demineralization/impregnation area is smaller than that promoted by etch-and-rinse adhesives, this does not necessarily reflect on lower bond strength.[16] Self-etch adhesives differ from each other in the amount of intrinsic water and potential of hydrogen (pH).[17] Their formulation is available in one or two separate bottles/compartments and their application protocol can be carried out in one or two steps. Both presentations can be subdivided according to their acidity into 'strong' (pH≤1), 'intermediate' (pH ≈ 1.5), 'mild' (pH ≈ 2) and 'ultra-mild' (pH ≥ 2.5).[5] Those having a pH lower than or near to 1.5, contain functional monomers which mainly demineralize dental hard tissues instead of chemically bonding to hydroxyapatite; those having a pH greater than 1.5, only demineralize the dental substrate partially, chemically interacting with the remaining hydroxyapatite.[5] The latter thus only exposes dentin collagen very superficially, creating a nanoscaled hybrid layer also referred to as nano-interdiffusion zone.[18] Higher bonding durability obtained with these "mild" self-etch bonding agents can be ascribed, among other aspects, to the formation of a rather thin hybrid layer which is less prone to hydrolysis.

Partial demineralization of dentin and consequent bonding to remaining hydroxyapatite also counts for a more stable and durable bonding interface.[5,19] Such interaction is derived from the presence of specific functional monomers in the adhesive composition: 10-methacryloyloxydecyl dihydrogen phosphate (10-MDP), 4-methacryloyloxyethyl trimellitate anhydride (4-META) and 2-metacriloxil ethyl phenyl hydrogen phosphate (Phenyl-P).[20]

A new family of adhesives, named "universal" or "multi-mode", was recently introduced in the dental market aiming for further simplification of the adhesive procedure and rationalization of the inventory at the dental office.[21] The definition of a universal bonding agent is still controversial. Although many commercially available brands claim their universality, a real "multi-mode" adhesive should: a) perform equally well in both etching modes (etch-and-rinse and self-etch); b) bond to enamel and dentin as well as to different restorative materials (composites, ceramics, metal, zirconia, and so on); and c) be suitable for use in both direct and indirect restorative techniques irrespective of the curing mode of the luting cement (light-cure or dual-cure). Despite their versatility in terms of etching mode (etch-and-rinse or self-etch[21,22] such adhesives are essentially single-step self-etch bonding agents. Some of them contain copolymers of polyalkenoic acid which, in combination with MDP, show contradictory results regarding their bonding effectiveness.[23,24] Considering the concept of "multi-mode application", one must assume that bond strength would not be compromised by the chosen etching strategy. However, it has been noticed that enamel etching with phosphoric acid favors the bonding performance of these bonding agents.[17] For dentin, similar immediate performance was observed regardless of the application mode, but significant reduction in bond strength to dentin, both in dry and wet-bonding techniques, was reported in the long-term when phosphoric acid was used.[22] It has been suggested that phosphoric acid demineralizes dentin in a greater depth than self-etch adhesive systems can infiltrate. Moreover, the adhesive interface will not benefit from the advantages of chemical bonding in case the dentin surface is extensively demineralized and residual apatite crystals are not left in sufficient amount to promote the desirable interaction with the adhesive's functional monomers.[25,26] Finally, post-operative sensitivity is less likely to occur when the self-etch protocol is adopted.[22,27]

As another restorative alternative, glass-ionomers are self-adhering materials that can bond to tooth structure without an intermediate adhesive layer.[5,28,29] However, a short polyalkenoic acid pre-treatment is recommended, resulting in a two-step approach. Such acidic conditioner removes the smear layer and demineralizes the underlying dentin up to a depth of about 0.5–1 μm.[30] Glass ionomer components are then able to infiltrate the exposed collagen mesh, establishing a micro-mechanical retention which follows the principle of hybridization.[31] Within this hybrid layer, a chemical bonding is also obtained by ionic interaction of the carboxyl groups of the polyalkenoic acid with calcium of hydroxyapatite that remains attached to the collagen fibrils.[20] This two-fold bonding mechanism which combines micro-mechanical interlocking and chemical interaction may explain the clinical success boasted by glass ionomers in specific clinical indications such as non-stress bearing restorations[28] and atraumatic restorative treatment.[32] Likewise, mild self-etch adhesives, the chemical bonding promoted by glass ionomers has also been described as an important aspect against hydrolytic degradation of the adhesive interface.[5,28] Despite its excellent clinical performance in terms of retention and microleakage in non-stress bearing areas, glass ionomers commonly present lower aesthetic characteristics and polishing maintenance over time due to their lower mechanical properties when compared to resin-based restorative materials.[33] Nevertheless, a modern high-viscosity glass ionomer associated with a resin-based protective coating has been showing promising clinical performance in posterior permanent restorations,[34,35] therefore deserving further attention.

Dental adhesive technology, as impeccably stressed by Peumans et al.,[28] "evolves quickly and continuously with a rapid turnover of commercial adhesives, a high number of laboratory studies on adhesive materials and a high demand for laboratory techniques and data in potential prediction of clinical effectiveness". Despite the importance of laboratory studies, clinical trials

remain the ultimate source to collect scientific data on the effectiveness of different restorative materials and protocols. All things considered, the practice of restorative dentistry should base itself on qualified evidence-based dentistry to better understand the intrinsic limitations of current restorative materials, especially those of bonding agents, aiming to overcome them in favor of a successful clinical performance.

The key to success in adhesive dentistry relies on the effectiveness of the adhesive interfaces over time. By developing new restorative products and operative strategies, one can increase the long-term performance of aesthetic restorations, thus minimizing the need for replacement of failed restorations in the daily clinical practice. This review aims to explain the mechanisms of degradation of adhesive interfaces, suggesting strategies to overcome the intrinsic limitations of bonding agents and their application protocols.

Why do adhesive interfaces fail?

Sano[36] hypothesized that the biodegradation of the adhesive interface involves a sequence of events. The first stage towards biodegradation starts when dentin is acid etched for smear layer removal and exposure of the underlying collagen network for subsequent hybrid layer formation. Once depleted of minerals, the exposed collagen fibers become more prone to prospective deterioration. In the second stage, resins that infiltrated the dentin matrix are leached out and replaced by water creating nanometer-sized voids within the hybrid layer. The third stage involves enzymatic degradation of the exposed collagen fibrils. During bonding procedures with both etch-and-rinse and self-etch systems, demineralized dentin activates proteolytic enzymes (MMPs) that are also responsible for the degradation of unprotected collagen fibrils.[37,38] Therefore, we might need to counteract the adverse effects of enzymes such as esterases and matrix metalloproteinases (MMPs) at the bonding interface.

If biodegradation of the adhesive interface is to be avoided, a complete and intimate infiltration of resin monomers into the collagen network is fundamental, as well as an efficient polymerization of these monomers in situ. Both goals are clear and reasonable, but not so easily attained.

Acid etching of dentin removes its inorganic components, exposing collagen fibrils which should be subsequently protected through an intimate and complete infiltration of resin monomers from the bonding agent. However, the depth of demineralization can be greater than the infiltrating potential of resin monomers, leaving a zone of unprotected collagen fibers. This area is highly vulnerable to both hydrolytic and enzymatic degradation.[6] For the adhesives to properly impregnate its hydrophobic monomers into the intrinsically humid dentin, hydrophilic monomers are included in their composition. As a counter effect, the adhesive interface becomes more prone to water sorption and consequent hydrolysis[39,40,41,42,43,44] Hydrophilic adhesives can also attract water from hydrated dentin resulting in water-filled channels within the polymer matrices.[45] Besides promoting the degradation of collagen fibers, water also presents a degradative effect on resin monomers. Most of the monomers used in bonding agents nowadays do contain chemical groups such as ester, urethane and hydroxyl groups and ether linkages that are susceptible to hydrolytic cleavage. Water within the adhesive layer is therefore also responsible for the elution of unreacted monomers within the adhesive layer[46] and plasticization of the polymer network.[47] Porosity and intermolecular spaces in the polymer network further contributes for the degradation of the hybrid layer due to hydrolysis or exposure of additional collagen fibrils to activated MMPs.[48]

Interaction between polymeric dental materials and dentin organic components seems to be rather complex from a sub-micrometer perspective, therefore impairing a hermetic encapsulation of dentin collagen fibrils. According to its structural relation with collagen, hydroxyapatite in dentin can be classified into extrafibrillar mineral, located among the fibrils, and intrafibrillar mineral, mainly in the gaps within the fibrils extending between collagen molecules.[49] The synthetic monomers used nowadays are meant to infiltrate the space among collagen fibrils. Nevertheless, they do not seem to be small enough to penetrate the nanometric voids between collagen molecules within the demineralized collagen fibers.[50] Moreover, it has been shown that collagen molecules are surrounded by water molecules to form highly

ordered and multilayered cylinders of hydration[51] which may favor the hydrolytic degradation of resin monomers in a sub-micrometer level.

To reduce the degradation process detailed above, some recent strategies can be explored, such as: inactivation or blockage of MMP's; reduction of dentin water content without increasing hydrophilic components of adhesive systems; addition of new chemical functional monomers into bonding agents; and an increase of collagen fibers stiffness to improving their resistance.

Strategies to counteract degradation of adhesive interfaces

Caries management from the restorative point of view

Minimal intervention approach recommends complete removal of infected dentin and maintenance of caries-affected dentin (CAD) on the pulpal cavity floor. Although most the literature related to the performance of bonding agents refers to sound dentin as a standard substrate,[52] clinicians are frequently challenged by the presence of caries-affected and/or sclerotic dentin in their daily practice. In the same way, manufacturers base their new technologies on the adhesion to sound dentin which is far from representing a clinically relevant substrate. The main common goal among researchers and manufacturers should be the development of a bonding agent that could equally bond to sound, caries-affected, eroded and sclerotic dentin.[53]

The caries process induces different chemical, biological and physical modifications on the affected substrate, rendering it less favorable for interaction with polymeric restorative materials.[54] Bonding to infected dentin is contra-indicated due to its high bacterial count, lower resistance due to high minerals loss, and advanced deterioration of collagen structure. Consequently, such substrate should be completely removed.[55] Caries-affected dentin, in turn, is more permeable than sound dentin due to its lower mineral content, besides presenting a higher water ratio and important changes in the secondary structure of collagen.[54] Despite these unfavorable features, CAD still stands high chances of remineralization, which justifies its maintenance at the pulpal floor of the cavity. It should be considered, however, that a hybrid layer of poorer quality is obtained on CAD, and that a lower bonding performance may be expected irrespective of the kind of bonding agent employed.[56]

While manufacturers do not create an adhesive system that performs equally well on different sorts of modified dentin, it is highly recommended to finish the margins of the restoration in sound tooth structure to assure an optimal marginal sealing.[56] In the light of a more conservative approach, caries-affected dentin should be maintained at the pulpal cavity floor and should be covered with a thin layer of glass ionomer cement. There are indications that glass ionomer cements may promote remineralization of caries affected dentin,[57] although further studies are necessary to evaluate the reincorporation of mineral content within demineralized dentin and the recovery of its mechanical properties. The bonding agent must be applied over the remaining cavity walls and margins after application of the glass ionomer cement as part of the procedure for adhesive composite restorations.

Chemical bonding

Chemical bond in SE adhesives is achieved through specific functional monomers that bind calcium ions of hydroxiapatite within the hybrid layer.[5,58] Different functional monomers may be found in this kind of bonding agents such as 10- methacryloyloxydecyl dihydrogen phosphate (10 MDP), 4- methacryloxyethyl trimellitic acid (4-MET) and 2 (methacryloyloxyethyl) and phenyl hydrogenphosphate (Phenyl-P).[5,58] Their physico-chemical properties play an important role on the bonding effectiveness and durability of self-etch adhesives.[5,59]

Such chemical bond is mostly observed in mild and ultra-mild SE adhesives (pH < 2) that partially demineralizes dentin producing sub-micrometer hybrid layers[14] in which substantial HAp-crystals remain around partially exposed collagen fibers.[3,18,60,61] Besides protecting collagen fibers against degradation, such remaining mineral content also serves as receptor for additional chemical bonding with the respective functional monomer,[14,18] by which their two-fold bonding mechanism (*i.e.* micro-mechanical and chemical adhesion) closely resembles that of glass-ionomers.[62]

The additional chemical bonding of mild self-etch adhesives and glass-ionomers is supposed to be beneficial in terms of bonding durability.[18,20] However, the ability to chemically bond to HAp is not sufficient on its own. The formed ionic bond should also be stable in an aqueous environment. It has been demonstrated that the chemical bonding generated by 10-MDP is not only more effective, but also more stable in water than that of 4-MET and phenyl-P.[18] The dissolution rate of the calcium salts formed by these three monomers was inversely related to their chemical bonding potential. This chemical phenomenon is known as the adhesion-decalcification concept (AD-concept) that defines if molecules will either decalcify or adhere to mineralized tissues.[63] The more effective bonding promoted by 10-MDP-containing adhesives has recently been demonstrated via bond strength tests.[64] From a chemical point of view, the ionic interaction of 10-MDP with HAp has been revealed by X-ray diffraction (XRD) complemented by Transmission Electron Microscopy (TEM), presenting it as a "nanolayered" structure at the tooth-adhesive interface, and this being more evidently observed on dentin than on enamel.[64] Each layer of this self-assembled nano-layered structure constitutes of two 10-MDP molecules with their methacrylate groups directed towards each other and their functional hydrogen phosphate groups directed away from each other. In between the layers, calcium salts are deposited.[65]

Apparently, the weakest zone in self-etch adhesives is located below the hybrid layer, where the adhesive penetration and/or polymerization are not sufficient to protect collagen against hydrolysis and enzymatic degradation.[6,66,67,68] Despite its superior performance as compared to other functional acidic monomers, the salt formed by 10-MDP may also degrade with time, as it is also somehow sensitive to hydrolytic dissolution.[61] According to Kim et al.[67] the hydrolytic degradation of the collagen fibers still occurs at the base of the hybrid layer, characterized by voids and nanoleakage between the intact top of the hybrid layer and the mineralized dentin base, as detected by SEM and Confocal microscopy analysis.

Considering the chemical bonding approach, a strategy based on the selection of 10-MDP-based adhesives could be an additional advantage to improve bonding durability of esthetic restorations.

Enzymatic inhibitors

As mentioned before, one of the foremost impediment for the longevity of adhesive interfaces is degradation of exposed collagen fibrils at the base of the hybrid layer: preservation of their integrity is tough pivotal to improve bonding durability.[68]

Since Pashley et al.[37] and Armstrong et al.[39] auspiciously linked such degradation to the action of host derived endogenous matrix metalloproteinases (MMPs),[69] several approaches to make them inactive, hence slowing down or nullifying the phenomenon, started to be considered.[40,68,70,71,72] It is well-known that certain substances are promising MMPs inhibitors, but the most studied in adhesive dentistry is chlorhexidine (CHX).[37,58,73,74] It is, in addition, also capable of inhibiting cysteine cathepsins (CCs)[75] the other key class of proteolytic enzymes identified in dentin.[76]

CHX was accordingly shown to partially conserve the integrity of the hybrid layer,[77], favoring bond durability. CHX can be used as an antiproteolytic "primer" solution directly applied on the dentin surface, or after phosphoric acid etching and rinsing[74,78,79,80,81,82] or incorporated into the acid etching agent[83,84] or within the adhesive system composition.[74,85,86,87]

The use of 2% CHX solution as an effective and nonspecific protease inhibitor primer, after acid-etching, is attractive from a clinical point of view, since it is already used in other oral health situations as an antimicrobial agent.[88] Compared to control, aged CHX-treated dentin relates to higher bond strength values[73,74,77,78,79,80,89,90,91,92] which are interestingly equivalent to each other in most of the immediate evaluations.[93] Especially when two step etch-and-rinse adhesives are used, reduction of bond strength values for conventionally treated dentin over one to two years is of approximately 50%; when CHX is applied after acid etching, total reduction is around 20%[72] to 84.9%.[88]

Even though the presence of carious-affected dentin, as well as artificially eroded dentin, relates to inferior bond strength values,[53,94,95] CHX can preserve adhesive interface stablished by two-step etch-and-rinse adhesives, although not indefinitely.[77,96,97,98,99] Higher bond strength durability of a two-step self-etch adhesive over 2-year aging in artificial saliva and under simulated intrapulpal pressure was also described

in the literature when caries-affected dentin was pretreated with CHX.[100] Besides, the degradation of the adhesive interface in primary teeth may be counteracted or blocked by the application of CHX as a therapeutic primer.[92,101]

Avoiding to add another step in the adhesive treatment of dentin,[6,88] some investigators have studied the beneficial effects of incorporation CHX into the etchant[83,84] or the adhesive.[87,97,102,103,104] First strategy was alleged to reduce the effectiveness of CHX in the inhibition of MMPs and CCs, due to the limited contact time and the little concentration of CHX at the moment of adhesive application. It should be considered that CHX is leached from demineralized and mineralized powdered human dentin with water rinsing.[67] However, Loguercio et al.[105] have identified CHX inside a 5-year aged hybrid layer after application of a CHX-containing etchant. The high substantivity of CHX on dentin may explain inefficiency of the rinsing step in eliminating substantial amounts of it.[106]

CHX incorporated into experimental adhesives, in turn, was proved not to jeopardize immediate bond strength to dentin and to partially reduce the degradation of the resin-dentin bonds after aging.[103] A reservoir of CHX for controlled release would be derived from its inclusion into primers and/or adhesives:[87] addition of relatively low CHX concentrations to commercial adhesive, chiefly in ethanol-solvated hydrophobic resins, was shown feasible in this sense.[85,86,87] Controversial results look as if they were related to the concentration of CHX added to the adhesive formulation and/or to the chemical compositions of the adhesives,[93,102,107,108] which may influence resin water sorption, and hence its solubility, degree of conversion, and mechanical properties.[85,109] To endorse CHX incorporation into adhesives, Stanislawczuk et al.[87] advised the conduction of further studies in more clinically relevant conditions. In vitro studies have already attested that CHX-containing primers of two-step self-etch and simplified etch-and-rinse adhesives are capable of, respectively, inhibiting MMPs[108] and reducing nanoleakage,[104] precluding time-related bond strength loss.[102,104] An adhesive system containing 0.2% chlorhexidine (Peak Universal Bond, Ultradent Products Inc, South Jordan, USA) was then just introduced in the market.[6]

As an alternative, Abu Nawareg et al.[110] successfully used a monomer named CHX-methacrylate (Ivoclar Vivadent, Schaan, Liechtenstein) as a primer to improve hybrid layer long-term stability. In this way, CHX would both bind to demineralized dentin[67] and copolymerize with adhesive monomers: CHX-methacrylate would be much more than electrostatically bound to the dentin matrix, it would be sealed inside it, maybe for many years.[110] Then CHX-methacrylate could inhibit matrix proteases probably the same way CHX digluconate can do.[110]

CHX is a cationic bisbiguanide which inhibits collagenase/gelatinase activity of dentin matrices.[74,37,111,112] It is believed that it acts by cation chelation, sequestrating calcium and zinc ions that are essential for the activation of the MMPs catalytic domains.[111,113,114] Even low CHX concentrations (0.002–0.02%) and short-time applications (15 to 30 seconds)[86] are effective in minimizing the degradation of adhesive interfaces, but the association between the concentration and the bond strength is not linear.[73] Lower percent of failure mode in the hybrid layer, especially at the bottom part, found after six-month aging when CHX was applied, was one of the first indicative of its effectiveness as a protease inhibitor in adhesive dentistry.[78] Later, after CHX application, improved collagen network formation (better adhesive penetration), reduced collagen degradation, and distinct gold-labeling signals were identified by field emission scanning electron microscopy and immuno-gold staining; a better resin and dentin tube combination was detected in the surface micromorphology of the fractured dentin resin restoration as well.[38] About long-term effectiveness, it would be explained by the high substantivity of CHX.[106] Regardless of its concentration, CHX has a strong affinity to the dental structure, binding to phosphate groups of mineralized dentin crystallites and to negative carboxyl groups of the collagen matrix (electrostatic forces between NH_3^+ in the CHX molecule and $COOH^-$ or OH^- in dentin).[106] After oversaturating proteases binding sites, if still available (higher concentrations), it can remain bound to collagen fibrils for later release.[115]

Adversely, a limitation of CHX indication as an enzymatic inhibitor is that its effect seems not to be

indefinitely long.[85] Because CHX molecule is large and water soluble, it may be gradually leached out from the adhesive interface,[92] especially when in contact with an external environment (through marginal gaps, for instance).[105] Remaining concentration then becomes no longer appropriate to exert noticeable antiproteolytic effects.[92]

Usage of CHX on etched dentin before priming plus bonding together, thus when two-step etch-and-rinse adhesives are applied,[116] is strongly recommended; by the way, demineralization increases CHX binding to dentin.[67,106] Its effect on the durability of the bonding when three-step etch and-rinse are used, in turn, appears to be only slightly favorable. Adhesive interfaces determined by using non-simplified etch-and-rinse systems are, per se, more stable than that determined by using the simplified counterparts.[73] Regarding the effectiveness of self-etch adhesives, and even the moment or how CHX must be applied, are conflicting.[74,90,117,118,119] Recent systematic reviews show that an aging-associated decline in dentin bond strength of both categories of adhesives can be lessened by CHX application.[74,118,119] In this sense, a growing predisposition among clinicians in properly applying 0.2–2% CHX for 15–60 s for re-wetting the collagen network on acid-etched dentin[77,79,89,120] to minimize degradation of resin-dentin bonds, can be currently appreciated.[115,119] Tough medium-term randomized clinical trials (18-36 months) does not show significant beneficial results from incorporating CHX to adhesive treatment of dentin.[121,122,123] Loguercio et al.,[105] incite researchers to evaluate the only one proven clinically and easy to adopt strategy[88] after long-term follow-ups, when the benefits of CHX would possibly turn out to be detectable.

Inhibitors of endogenous dentin proteases other than CHX, such as the quaternary ammonium surface-acting benzalkonium chloride (BAC)[124,125,126,127] the tetracyclines (TCs) and their antimicrobially inactive analogs (minocycline, for now),[71,84,128,129,130] bisphosphonates (batimastat, galardin, and zoledronate),[93,129,131,132] green tea polyphenol epigallocatechin-3-gallate (EGCG),[40,133,134] and the chelating agent ethylenediaminetetraacetic acid (EDTA) [135,136,137] are being studied as better alternatives associated with dentin hybrid layer preservation. The same happens to adhesives containing zinc [114,138,139,140,141] or MMP-inhibiting monomers or solvents: polymerizable quaternary ammonium methacrylates (QAMs; *e.g.* 12-Methacryloyloxydodecylpyridinium bromide / MDPB),[66,68,93,125,142,143] incorporated into the primer of Clearfil Protect Bond, Kuraray,(6) and dimethyl sulfoxide (DMSO),[144,145,146] correspondingly. As further efforts are required to ratify related initial encouraging results, for compiled deeper information, one should refer to Perdigão et al.[40] and Tjäderhane.[6]

Authors claim that,[6,41,68] although CHX may not be unfailing, it can and should be indicated (often as an optional step before application of etch-and-rinse, or even self-etch, primer / primer plus bonding) until new strategies have been recognized harmless and just as effective in preserving collagen fibrils integrity, thus favoring bonding durability for some time.

Addition of a separate hydrophobic adhesive resin

Simplified adhesive systems, both two-step etch-and-rinse and one step self-etch adhesives, are composed of high concentrations of hydrophilic resin monomers and a higher amount of water than their more complex counterparts, namely three-step etch-and-rinse and two-step self-etch adhesives. This composition is necessary as water is required to dissociate the weak acidic methacrylate monomers into ionized forms for permeation into dentin.[16] However, the excess water may prevent the optimal polymerization of the adhesive monomers, thereby reducing the mechanical properties of the adhesive layer and the resulting resin–dentin bond strength. Additionally, due to their high hydrophilicity, such simplified adhesives remain permeable upon polymerization, permitting movement of water from both the host tooth as well as from the outer oral cavity across the interface.[147]

To counteract this limitation of simplified adhesives, it has been proposed that the application of an additional layer of hydrophobic fluid resin should be incorporated to their bonding protocol.[24,148,149,150,151,152] This strategy proved to be effective in minimizing nanoleakage and improving the polymerization of simplified adhesives[150] as well as their immediate[40,146,150] and long-term[146] bond strength. It could also improve the sealing ability of

two-step etch-and-rinse adhesives by reducing fluid conductance across the adhesive interface.[151]

All things considered, an extra coating of hydrophobic resin should be applied over dentin treated with both simplified adhesive systems, etch-and-rinse or self-etch. Advantages of this technique can be the improvement of marginal sealing, bond strength and degree of conversion, together with a reduced post-operative hypersensitivity.[6]

Selective enamel acid-etching

Contrary to the possible shortcomings of etching dentin with phosphoric acid, its selective application on enamel seems to have been recognized as a key step toward the clinical success of self-etch protocol,[153] especially when mild and ultra-mild adhesives are considered.[154,155,156] The pH of self-etch and self-adhesive materials directly relates to their reduced potential, as compared to that of phosphoric acid,[157,158] to etch the highly-mineralized enamel.[25,155,156,157,159,160,161,162,163,164] Hence, increasing its surface area and creating deep etch-pits remains important for the achievement of an effective and durable bond to enamel.[4,165,166,167,168] According to the buffering capacity that such substrate offers to the action of self-etch systems in general, it can be expected that they do have a lower demineralizing effect on the hydroxyapatite-rich enamel, which ultimately compromise their bonding effectiveness.[169,170]

Several in vitro studies proved that higher bond strength of self-etch systems to enamel is achieved with the selective phosphoric acid etching technique.[161,171,172,173,174] The same proved true, in recent times, for universal adhesives.[17,168,170,175,176] Performing selective enamel etching implies in less marginal discoloration and better marginal adaptation.[24,177,178,179,180,181] Likewise, an interesting meta-analysis communicated by Peumans et al.[28] showed that selective etching reduces the clinical annual failure rates of mild self-etch systems, notwithstanding this was not true when clinical trials were evaluated separately. Also, retention rates of restorations of NCCLs might be improved by selective application of phosphoric acid on enamel and further employment of self-etch adhesives.[175]

It is suggested, therefore, that prior acid etching of enamel with phosphoric acid should be a routine procedure when using any self-etch protocol, to achieve higher retention of the resinous material,[5,172,173] especially in more critical situations due to the structure of the substrate: unground/aprismatic enamel; or due to the lack of inherent form of retention of certain cavity profiles a sin the case of Class IV cavities, fractures in anterior teeth, and luting laminate veneers, for instance.[9,14,153,160,182] Selective acid-etching is increasingly more suitable the greater the amount of remaining enamel.[182]

Cross-linkers

Type I collagen represents the major component of the organic matrix of hard tissues and its stiffness depends on the formation of endogenous and exogenous cross-links.[183] Collagen is a heterotrimeric molecule composed of two $\alpha 1$ and one $\alpha 2$ chains that are comprised of three domains: NH2–terminal (N-telopeptide), the central triple helix and the COOH-terminal non-triple helical (C-telopeptide) domain.[184]

Endogenous collagen cross-links are mediated by both non-enzymatic and enzymatic reactions. Non-enzymatic collagen cross-links are mediated by oxidation and glycation processes,[185] while enzymatic reactions happen between the telopeptide and adjacent triple helical chains,[186] which is mediated by chemical reactions through lysyl oxidase covalent bonding.[187,188] This results in the formation of inter and intra-molecular and inter-microfibrillar cross-links.[187,188,189,190,191] Such enzymatic reactions are the basis of tissue stability, viscoelasticity and strength of collagen fibrils.[184,192] In particular, intra-molecular cross-links provide primarily biostability to the collagen molecule, while inter-molecular and inter-microfibrillar cross-links enhance mechanical properties, in addition to fibril biostability.[193]

Exogenous cross-linking agents have been proposed to mimic physiological cross-links. They increase the intrinsic properties of the collagen against collagenases degradation through inducing additional formation of inter and intra-molecular cross-links.[194,195] Exogenous cross-links also improve biomechanical and biostability properties of collagen fibrils[196] and are mediated by non-enzymatic reaction sources, such as chemical agents (*i.e.* glutaraldehyde, carbodiimide hydrochloride and natural resources) and physical methods.[196]

Physical methods are mediated by photo-oxidative reaction, usually by light exposure, such as ultraviolet radiation (UVA).[197] Riboflavin is the most common cross-link of this class that can be induced by UVA. However, the use of UVA in clinical practice is unfeasible.

Among the great variability of synthetic chemical agents, the most widely known is Glutaraldehyde (GA).[68,196] GA can induce cross-links in collagen, consequently enhancing its mechanical properties, such as hardness, and maintaining its mechanical stability. GA may also prevent matrix degradation by crosslinking the binding and/or active sites of endogenous dentin MMPs, therefore blocking the access of such enzymes.[198] However, its high cytotoxicity makes its clinical use inappropriate.[196]

Carbodiimide hydrochloride (EDC) has also been proposed as an effective collagen cross-linker as it has been shown to improve the durability and structural integrity of the adhesive interface, thus preserving its bond strength over time, and this through the formation of inter- and intra-molecular cross-links.[199,200,201,202] Moreover, EDC is one of the least cytotoxic and most stable cross linkers.[58] EDC also seems to be capable of inactivating dentinal gelatinases.[200,201,202] It should be considered, however, that it is currently designed to be used just after acid-etching, in etch-and-rinse adhesives.[58]

Genipin is a natural cross-linking agent that can react with the amino groups of lysine, hydroxylysine or arginine to form intra or inter-molecular cross-links within collagen molecule or between adjacent collagen molecules.[195,203] However, the rates of induced exogenous cross-links are slow, which represents a limiting factor as far as treatment of dentin is taken into account.[204]

Proantocianidin (PA) is a polyphenolic compound that can be extracted from several fruits, nuts, vegetables and barks.[195,205] Their interaction with collagen type I depends on the type of PA, chemical structure, stereochemistry pattern and concentration of these natural extracts.[206,207] PA is a natural cross-linking agent that has been widely studied in recent years due to their ability to biomodify the dentin matrix[184] and enhance its mechanical properties and resistance against biodegradation, finally favoring the resin/dentin bond strengths.[199,204] PA can interact with collagen tissue and induce non-enzymatic collagen cross-linking[193] increasing collagen stiffness and dentin bond strength by keeping the bonding stable over time.[207]

Even though crosslinkers proved to be efficient in enhancing mechanical properties of dentin, and impairing degradation of the dentin-resin interface, researchers did not reach the exact point to recommend its clinical use. More studies are necessary to turn this promising strategy into a clinically effective protocol.

Ethanol – wet Bonding

The development of a promising bonding procedure on dentin is still challenging due to its humid and porous intrinsic biological features. The formation of a stable hybrid layer depends on an efficient penetration of resin monomers for a tight encapsulation of the exposed collagen matrix.[208] However, this ideal hybrid layer is difficult to obtain, being therefore prone to hydrolytic degradation of collagen fibrils and adhesive polymers.[40,46,209]

The "ethanol wet bonding" technique consists of gradually replacing water from interfibrillar and intrafibrillar spaces by ethanol, starting with the application of lower concentrations of ethanol solutions and slowly progressing to higher concentrations up to a complete dehydration of the exposed collagen network.[13,210,211,212]. Ethanol has a higher vapor pressure than water, enhancing its evaporation and creating wider interfibrilar spaces for impregnation of hydrophobic monomers to form a more stable hybrid layer.[213,214,215]

Basically, ethanol dehydrates the demineralized collagen matrix and coaxes hydrophobic monomers into it.[216] This technique also prevents phase separation of hydrophobic resin monomers in the presence of water,[217,218,219] since the latter is completely replaced by ethanol prior to the application of the ethanol-soluble monomers.[220,221] Additionally, the elimination of residual water seems to contribute to decrease or even eliminate hydrolytic enzymatic degradation of collagen fibrils,[212,222] thereby increasing bond durability and stability.[213]

However, this technique is very sensitive, time consuming and requires the application of many

steps to achieve the desired dehydration, which becomes inappropriate. More studies are necessary to improve this protocol for clinical use.

Conclusion

Our main objective towards the formation of a strong and durable bond to tooth structure is to create on dentin a hybrid layer which is completely free of voids so that the collagen network becomes fully protected against hydrolytic and enzymatic degradation. However, based on the current state of the art, this task seems impossible to be fully achieved. Basically, the adhesive interface as we know today is the antithesis of a completely successful bonding, especially from a nanoscale and molecular standpoint. On the other hand, we do believe that there are materials and strategies that can certainly help to prevent degradation within the adhesive interface. It is fundamental that professionals are aware of these strategies to counteract degradation as much as possible. None of them are efficient to completely solve the problem, but they certainly represent reasonable alternatives to increase the lifetime of adhesive restorations. In a nutshell, we could mention:

a. Preserve as much as possible enamel on cavity margins;
b. Remove all infected dentin;
c. Remove caries-affected dentin from surrounding walls, maintaining it at pulpal and axial walls;
d. Maintain the correct humidity of dentin, according to the type of bonding agent used;
e. Use selective acid etching for self-etch agents when cavities margins are on enamel;
f. Clean cavities with anionic detergent before proceeding with the restorative protocol;
g. Make sure your curing unit is effective with the right wavelength per photo initiators of the bonding systems. Thus, follow manufacturer`s instructions;
h. When opting for self-etching strategies, give preference to those promoting chemical adhesion to the dental substrate, especially those based on 10-MDP.
i. An extra coating of hydrophobic resin should be applied over dentin treated with any of the currently available simplified bonding agents.

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