Journal of Dental Research
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J DENT RES 1994; 73; 44
DOI: 10.1177/00220345940730010601

J Dent Res 73(1):44-55, January, 1994

In vitro Bond Strengths and SEM Evaluation of Dentin Bonding Systems to Different Dentin Substrates

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Abstract. In comparison to enamel, bonding to normal dentin is a greater challenge because of its organic constituents, fluid-filled tubules, and variations in intrinsic composition. Bonding to sclerotic dentin is even more difficult. To evaluate the shear bond strengths of four adhesive systems to dentin substrates with different levels of mineralization, 120 extracted human teeth were randomly assigned to three groups (n = 40). After mid-coronal dentin was exposed, groups of specimens were artificially hypermineralized by immersion in a remineralizing solution, demineralized by means of an acetic acid demineralizing solution, or stored in distilled water to model sclerotic, carious, and normal dentin, respectively. Resin composite was bonded to dentin by use of commercial adhesive systems. After the specimens were thermocycled, shear bond strengths were determined in an Instron universal testing machine. Dentin substrates and resin/dentin interfaces were examined by SEM. For each adhesive system, the mean shear bond strength to normal dentin was significantly higher than that to either of the other substrates. Shear bond strengths to hypermineralized dentin were significantly higher than those to demineralized dentin with all adhesives except Prisma Universal Bond 3.

Key words. Dental Bonding, Dentin.

Introduction

Strong, durable bonds between dental biomaterials and tooth substrates are essential, not only from a mechanical standpoint, but also from biologic and esthetic perspectives. Good marginal adaptation of restorative materials reduces microleakage, staining, pulpal irritation, and recurrent caries (Nakabayashi, 1992).

Buonocore (1955) demonstrated that acid-etching of enamel with 85% phosphoric acid increased the retention of resin to enamel. Bonding is micromechanical, because resin forms tag-like extensions into the etched enamel surface (Gwinnett and Matsui, 1967). Dentin is a less favorable substrate than enamel for resin bonding. Many factors contribute to the difficulty in bonding, including the high organic content of dentin, variations in its intrinsic composition, the presence of fluid and odontoblastic processes in the tubules, the presence of the smear layer, and the inherent wetness of the surface (Pashley, 1989; Ten Cate, 1989; Söderholm, 1991).

Bonding to hypermineralized dentin surfaces is even more difficult than bonding to normal dentin (Duke and Lindemuth, 1990). Hypermineralized dentin occurs in several situations. For example, peritubular dentin is more mineralized than intertubular dentin (Takuma, 1960). In addition, dentin changes throughout the life of an individual, since deposition of calcified tissue continues with function (Mendis and Darling, 1979; Duke and Lindemuth, 1991). Dentin in naturally desensitized areas is also highly mineralized, and most of the tubules are occluded with rhombohedral crystals (Yoshiyama *et al.*, 1989). Under carious lesions, deposition of beta tricalcium phosphate crystals increases the mineral content and decreases the permeability of dentin (Duke and Lindemuth, 1991).

Sidhu *et al.* (1991) found that the composition of the dentin substrate may affect the performance of bonding agents. Some bonding agents might bond more readily to a hypermineralized

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Received May 14, 1993; Accepted September 29, 1993

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Based on Dr. Perdigao's thesis, which was submitted to the Graduate College, The University of Iowa, in partial fullfillment of the requirements of the MS degree

Table 1. Batch numbers of materials used

All-Bond 2	
All-Etch	029072
Primer A	059282
Primer B	029092
Dentin/Enamel Bonding Resin	069262
Amalgambond Plus	
Activator	20402
Adhesive Agent	072792-397183
Base B	20401
Catalyst C	204031
Prisma Universal Bond 3	
Dentin Primer	920708
Adhesive	920723
Scotchbond Multi-Purpose	
Etchant	P920319
Primer	P920319
Adhesive	P920319

tissue and others to a more organic substrate. Duke and Lindemuth (1990) stated that increases in peritubular dentin and obliteration of tubular orifices may preclude the development of adequate micromechanical retention. For example, Scotchbond 2 primer (maleic acid and HEMA) does not condition sclerotic dentin effectively (Duke and Lindemuth, 1991).

The purpose of this study was to evaluate the shear bond strengths of four adhesive systems to dentin substrates with different levels of mineralization. This information could help to clarify the roles of the organic and inorganic components of dentin in resin bonding.

Materials and methods

Specimen preparation

One hundred twenty unrestored caries-free human molar teeth were selected for this study. Tissue remnants and debris were removed from the teeth, and the teeth were refrigerated for up to one year in a thymol disinfectant solution. The occlusal surface of each tooth was ground flat with a water-cooled orthodontic model-trimmer (Whip-Mix, Louisville, KY). Half of the crown height was removed to expose mid-coronal dentin (Gwinnett, 1992). Dentin was polished with wet 240-, 400-, and 600-grit silicon carbide abrasive paper on an Ecomet grinder (Buehler, Ltd., Lake Bluff, IL). The polished surfaces were inspected with a dissecting microscope (American Optical Company, Buffalo, NY). If any enamel remained, the surface was ground again until all enamel was removed. The apices were sealed with sticky wax (Whip-Mix), and the teeth were covered with two coats of acid-resistant varnish. A 5-mm circular area was left uncovered as a bonding site in the center of the occlusal surface.

The teeth were randomly assigned to three equal groups (n = 40). The exposed occlusal dentin surfaces of the first group were etched for 5 sec with 32% phosphoric acid (Uni-Etch, Bisco, Itasca, IL) to remove the smear layer (Brännström *et al.*, 1979). The teeth were suspended in 600 mL of a mineralizing solution (pH = 7) which contained 1.5 mM calcium (from $CaCl_2 \cdot 2H_2O$), 0.9 mM phosphate (from K_2PO_4), and 0.15 M potassium chloride (Heilman and Wefel, 1989). The solution was maintained at room T, and was continuously stirred. The speci-

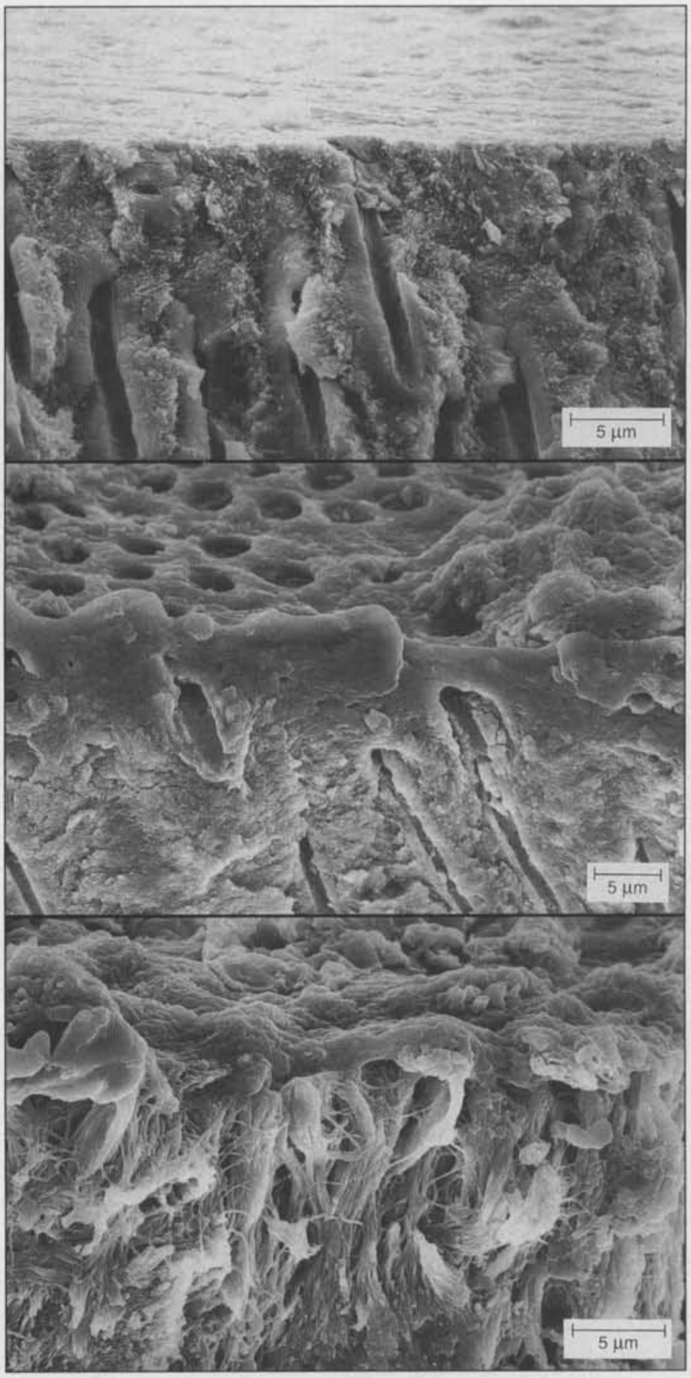

Figure 1. Scanning electron micrographs of (a) normal dentin; (b) artificially hypermineralized dentin; and (c) artificially demineralized dentin.

Table 2. Shear bond strengths (MPa)

	n	Mean	S.D.	Max	Min
All-Bond 2	30	10.62	6.60	23.86	0.00
Normal Dentin	10	16.84	4.06	23.86	10.60
Hypermineralized Dentin	10	12.07	3.11	15.24	7.02
Demineralized Dentin	10	2.93	1.67	5.56	0.00
Amalgambond Plus	30	11.16	7.47	21.47	0.00
Normal Dentin	10	17.47	4.59	21.47	5.03
Hypermineralized Dentin	10	14.05	2.33	16.96	9.81
Demineralized Dentin	10	1.96	2.41	7.42	0.00
Prisma Universal Bond 3	30	7.70	4.65	18.69	0.00
Normal Dentin	10	11.40	5.45	18.69	0.00
Hypermineralized Dentin	10	7.11	2.29	10.60	3.59
Demineralized Dentin	10	4.58	2.92	11.53	1.59
Scotchbond Multi-Purpose	30	9.63	6.93	19.09	0.00
Normal Dentin	10	16.14	2.21	19.09	11.40
Hypermineralized Dentin	10	11.23	5.06	17.32	2.38
Demineralized Dentin	10	1.51	1.04	2.91	0.00

mens were placed in fresh mineralizing solution every 24 h for 14 d, a duration that was determined by a pilot study. The second group of teeth (n = 40) was placed in a demineralizing solution of 0.1 mol/L acetic acid, which removes minerals but does not dissolve collagen (van Strijp *et al.*, 1992). The specimens were suspended in 400 mL of the solution (pH = 4.5) and stored in a refrigerator for 7 d (van Strijp *et al.*, 1992). The solution was changed after the second and fourth days. The remaining 40 teeth were stored in distilled water at room T.

Bonding procedures

All specimens were mounted in phenolic rings (Buehler) with self-curing acrylic resin (Shur-Tray, Miles Inc. Dental Products, South Bend, IN). The surface of the acrylic was coated with varnish to prevent any residual acrylic monomer from contaminating the dentin surfaces. Bonding procedures were carried out 7 d after the teeth were removed from the treatment solutions. The specimens were mounted on a custom apparatus for bonding with the adhesive systems All-Bond 2 (Bisco), Amalgambond Plus (Parkell, Farmingdale, NY), Prisma Universal Bond 3 (Caulk/Dentsply, Milford, DE), or Scotchbond Multi-Purpose (3M Dental Products Division, St. Paul, MN) (Table 1). For each adhesive system except Universal Bond 3, dentin was kept moist after being etched and rinsed by removal of excess water with a damp cotton pellet instead of by drying with compressed air. Moist dentin is a more appropriate substrate than dry dentin for some etched-dentin adhesives (Gwinnett, 1992; Kanca, 1992a; Swift and Triolo, 1992). For Prisma Universal Bond 3, which contains no dentin etchant, the surface was completely dried as directed by the manufacturer.

Gelatin capsules (Eli Lilly and Company, Indianapolis, IN) with an internal diameter of 4.85 mm were used as matrices. The gelatin capsules were filled to two-thirds of their length with resin composite (Command Ultrafine, Kerr Manufacturing Company, Romulus, MI) (Barkmeier *et al.*, 1991a) which was polymerized for 120 s. Restorative Z100 resin composite (A2 shade, 3M Dental Products Division) was inserted into the final one-third of each gelatin capsule, slightly overfilling the capsule. The capsule was applied to the dentin surface, excess material was removed, and the composite was cured for 160 s (40 s from each of four perpendicular directions) by use of a Demetron 401 visible-light-curing unit (Demetron Research Corporation, Danbury, CT). The intensity of the curing light was monitored periodically with a Curing Radiometer (Demetron Research Corporation), and its intensity was consistently in the range of 450-500 mW/cm^2. The specimens were set aside for 20 min and were then immersed in water (Leung *et al.*, 1983).

Bond strength testing

After storage in distilled water at room temperature for 7 d (Feilzer *et al.*, 1990), the specimens were subjected to 2000 thermal cycles (Brown *et al.*, 1972; Lloyd *et al.*, 1978). Each cycle consisted of 34 s in water baths of 10° and 50° ± 5°C, with an exchange time of 13 s between baths.

The specimens were then stored in distilled water for 48 h at room temperature. Bond strengths were measured in the shear mode by a universal testing machine (Instron Corporation, Canton, MA) with a 500-kg compression load-cell. A knife-edge shearing rod was attached to the crosshead, and the crosshead speed was set at 0.5 cm/min. The distance from the probe to the dentin was monitored by a spacer of two celluloid matrix strips (Hawe-Neos Dental, Gentilino, Switzerland). The force at which composite dislodged from the dentin surfaces was recorded on a strip chart, and shear bond strengths (MPa) were calculated from the cross-sectional area of the composite posts.

Table 3. Two-way analysis of variance

Source of Variation	Sum of Squares	DF	Mean Square	F Ratio	F Prob
Main Effects	3551.965	5	710.393	62.388	0.0001
Bonding System	208.741	3	69.580	6.111	0.001
Mineralization	3343.224	2	1671.612	146.805	0.0001
Interactions Bonding System vs. Mineralization	331.572	6	55.262	4.853	0.0001
Explained	3883.537	11	353.049	31.006	0.0001
Residual	1229.758	108	11.387		
Total	5113.295	119	42.969		

Table 4. Duncan's multiple-range test of shear bond strength

Group	Mean SBS (MPa)^a	Duncan Group^b				
ND/AM	17.47	A				
ND/AB	16.84	A	B			
ND/SB	16.14	A	B			
MD/AM	14.05		B	C		
MD/AB	12.07			C		
ND/PUB	11.40			C		
MD/SB	11.23			C		
MD/PUB	7.11				D	
DD/PUB	4.58				D	E
DD/AB	2.93					E
DD/AM	1.96					E
DD/SB	1.51					E

ND = normal dentin.
MD = hypermineralized dentin.
DD = demineralized dentin.
AB = All-Bond 2.
AM = Amalgambond Plus.
PUB = Prisma Universal Bond 3.
SB = Scotchbond Multi-Purpose.

[a] n = 10 for all groups.

[b] Values with the same letter are not significantly different at $p < 0.05$.

Microscopic evaluation

Each fractured specimen was examined with a dissecting microscope (American Optical Microscopy) so that the type of failure could be evaluated. In addition, two specimens from each bonding group were processed for SEM observation. Longitudinal 300 μm bucco-lingual sections were taken by means of a Silverstone-Taylor hard-tissue microtome (Sci-Fab, Littleton, CO). Sections of the restored specimens were immersed in 6 mol/L HCl for 30 s and 1% NaOCl for 12 h for partial demineralization and deproteinization of dentin (Nakabayashi and Takarada, 1992). The specimens were mounted on aluminum stubs with colloidal silver paint, and vacuum-desiccated for 24 h. They were sputter-coated with gold-palladium at 15 mA for 2 min and were examined in a Hitachi S-4000 Field Emission Scanning Electron Microscope (Hitachi, Tokyo, Japan) at a 12-kV accelerating voltage and 10-mm working distance. Also, specimens with cohesive failures were not demineralized, but were prepared for SEM observation by removing the coronal portion of each tooth and mounting it on an aluminum stub. Finally, six additional specimens were made and immersed in remineralizing solution, demineralizing solution, or water so that the surface morphology of the various dentin substrates could be examined with SEM.

Statistical analysis

Bond strength data were subjected to two-way and one-way analyses of variance (ANOVA). When the F-ratios were significant, Duncan's multiple-range test was used to compare specific mean values at $p < 0.05$. The statistical analysis was processed with the SPSS/PC+ software system (SPSS, Inc., Chicago, IL).

Results

The mean shear bond strengths and standard deviations for each group are listed in Table 2. All of the dentin bonding systems had higher shear bond strengths to normal dentin than to hypermineralized or demineralized dentin. Two-way ANOVA revealed significant differences in bond strengths based on dentin substrate and bonding system and their interaction (Table 3). Duncan's multiple-range test was used to test the significance of differences between specific means (Table 4). Amalgambond Plus, All-Bond 2, and Scotchbond Multi-Purpose on normal dentin had the highest mean bond strengths, and these were statistically similar. Bond strengths to demineralized dentin were the lowest of the three substrates for each adhesive. Prisma Universal Bond 3 was the only system that had statistically similar mean bond strengths regardless of whether the dentin was hypermineralized or demineralized. However, its bond strength to normal dentin was significantly greater than that to either of the other substrates.

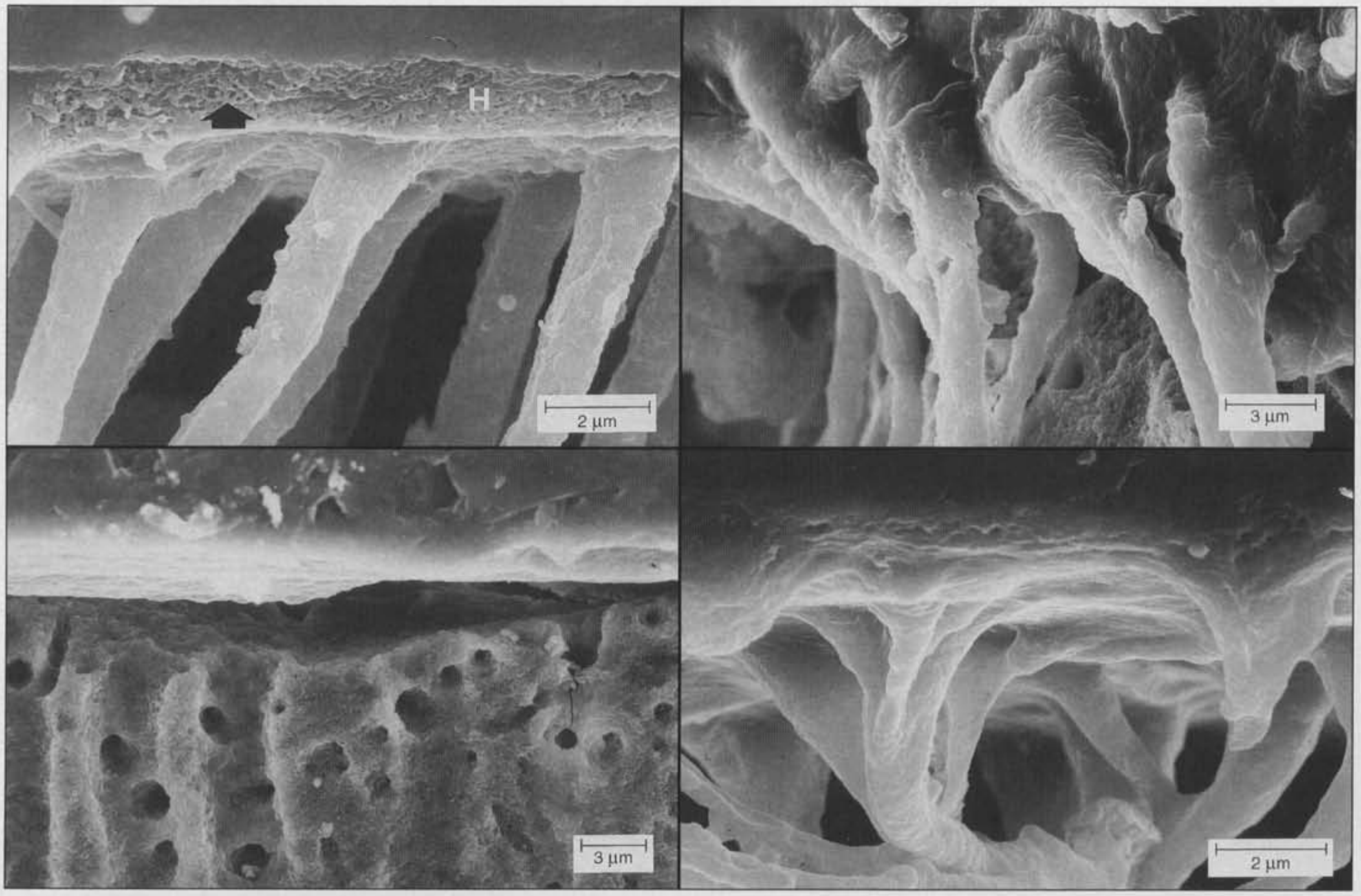

Figure 2. Bonding interfaces of (a) Amalgambond, (b) All-Bond 2, (c) Prisma Universal Bond 3, and (d) Scotchbond Multi-Purpose with normal dentin. The Amalgambond specimen shows a particularly well-defined hybrid layer (H). Arrow indicates one area of hybrid layer in which collagen fibers are evident.

The means were also examined for each of the two independent variables. When data were pooled by the degree of dentin mineralization, one-way ANOVA revealed that mean bond strengths to the three substrates were significantly different ($p < 0.0001$). The results of the *post hoc* test are shown in Table 5. When the data were pooled by dentin bonding system, ANOVA showed no statistically significant differences in mean bond strengths ($p = 0.18$).

The effects of the demineralizing and remineralizing solutions on the dentin substrates are shown in the scanning electron micrographs in Fig. 1. The dentin-resin interfaces of All-Bond 2, Amalgambond Plus, Prisma Universal Bond 3, and Scotchbond Multi-Purpose with normal dentin are shown in scanning electron micrographs in Fig. 2. The funnel-shaped configuration of the resin tags is evident mainly in Amalgambond Plus and All-Bond 2 specimens (Nakabayashi and Takarada, 1992). The necks of the resin tags are connected with resin-infiltrated dentin surface (Fig. 2b). All-Bond 2 and Amalgambond Plus also had a rough pattern on the superficial areas of the resin tags, whereas Scotchbond Multi-Purpose produced a smoother morphology. Prisma Universal Bond 3 generally did not penetrate the dentinal tubules, because this agent does not remove the smear layer (Barkmeier *et al*., 1990). However, scattered resin tags were present in some areas.

The dentin-resin interfaces of All-Bond 2, Amalgambond Plus, Prisma Universal Bond 3, and Scotchbond Multi-Purpose on mineralized and demineralized dentin are shown in Figs. 3-6.

Almost all failures were adhesive, with fractures occurring at the interface between dentin and resin (Fig. 7) (Table 6). As used here, the term "adhesive" means simply that no cohesive failure of dentin or resin was observed; it does not refer to the nature of the bond between resin and dentin. Sixteen of the 120 specimens had cohesive failures of dentin, with most of these occurring in normal dentin specimens. Another 16 specimens had mixed adhesive/cohesive failures, in which composite was still partially bonded to the dentin, but without dentin fracture. Eleven specimens had deep cracks into the tooth structure.

Discussion

Some investigators have proposed that chemical adhesion is the

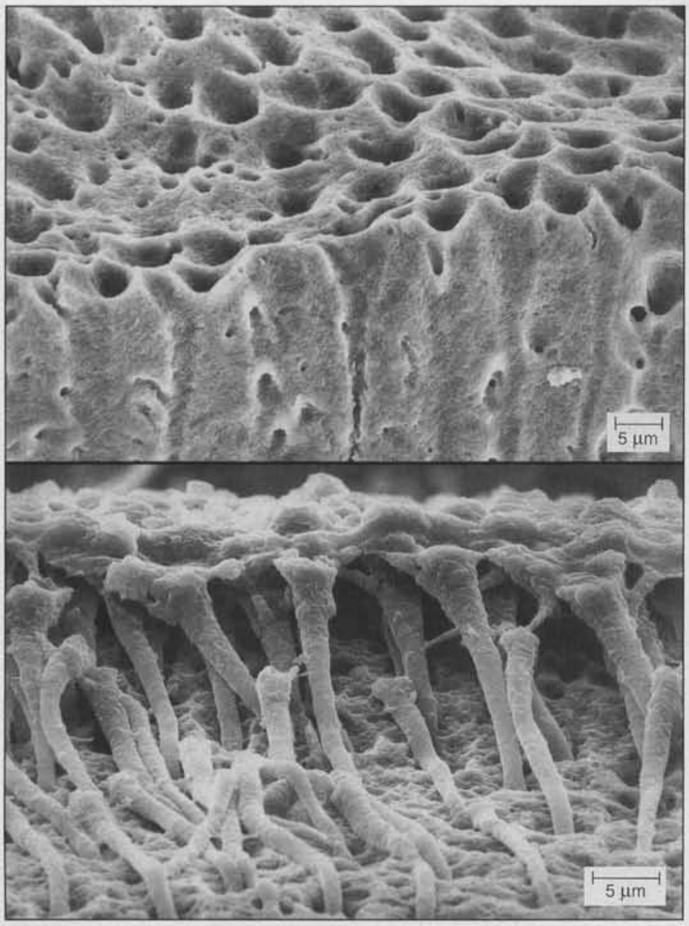

Figure 3. Resin dentin/interfaces of All-Bond with (a) demineralized and (b) hypermineralized dentin. No resin is evident on the demineralized dentin surface.

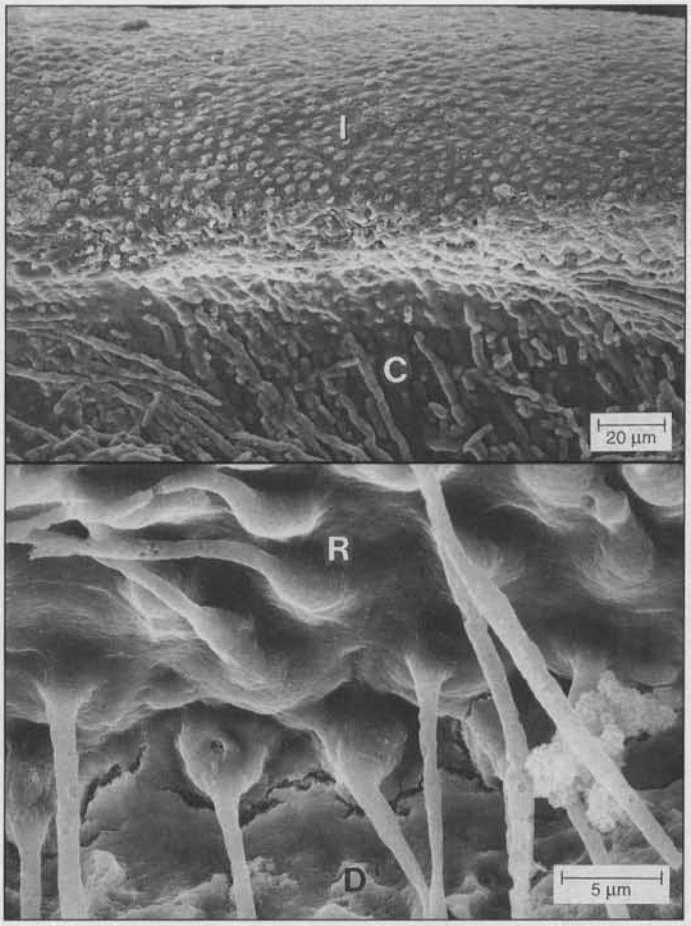

Figure 4. Resin dentin/interfaces of Amalgambond with (a) demineralized and (b) hypermineralized dentin. I = interface; C = dentin cross-section; R = bottom surface of resin; D = dentin.

principal mechanism of bonding to dentin (Buonocore *et al.*, 1956; Munksgaard and Asmussen, 1985; Ruse and Smith, 1991). Asmussen and Uno (1992) noted the presence of chemical groups in the collagen molecule which might be available for bonding, including hydroxyl, carboxyl, amino, and amido groups. However, micromechanical retention is now thought to be the most likely mechanism of resin/dentin bonding (Erickson, 1992; Spencer *et al.*, 1992). Micromechanical adhesion to dentin may depend on the hydrophilicity of the adhesive system (Erickson, 1992), and bonding of hydrophobic resins to etched dentin has proved unsuccessful (Torney, 1978). Acid-etching opens microporosities on the dentin surfaces and exposes collagen that collapses on itself due to the loss of inorganic support (Pashley, 1992; Van Meerbeek *et al.*, 1992). Primer application raises the collapsed collagen, keeping the porosities open (Erickson, 1992; Pashley, 1992; Van Meerbeek *et al.*, 1992). Resin penetrates the collagen network, resulting in a mechanical interlocking with dentin to form a "hybrid layer" or "resin-infiltrated layer" (Nakabayashi *et al.*, 1982; Erickson, 1989; Van Meerbeek *et al.*, 1992). All-Bond 2, Amalgambond Plus, and Scotchbond Multi-Purpose all rely on etched dentin, even though their specific mechanisms of action may differ. In this study, the mean shear bond strengths of these agents to normal dentin were not significantly different ($p < 0.05$).

All-Bond 2 uses a 10% phosphoric acid semi-gel to etch dentin for 15 s (Suh, 1991). Phosphoric acid decalcifies the superficial dentin, removes calcium and phosphate ions, and exposes collagen (Chiba *et al.*, 1989; Thompson *et al.*, 1989; Ruse and Smith, 1991). The depth of demineralization has been reported to be 7.5 µm with 10% phosphoric acid (Van Meerbeek *et al.*, 1992), although the decalcified dentin probably contains residual mineral particles (Van Meerbeek *et al.*, 1993). Adhesion is compromised if the depth of demineralization exceeds the depth of monomer penetration (Erickson, 1992; Pashley *et al.*, 1992). According to Van Meerbeek *et al.* (1992), the depth of the resin-reinforced layer for All-Bond 2 is 2.5 µm, meaning that the primers did not infiltrate through the entire depth of the decalcified dentin.

Bonding of the All-Bond 2 dentin adhesive system depends on the interaction of its primers with dentin and with each other (Bowen *et al.*, 1982; Bowen and Marjenhoff, 1991). The primers contain acetone, which acts as a "water-chaser" to carry resins into the etched dentin, resulting in a good adaptation to the surface (Bowen, 1985; Suh, 1991; Kanca, 1992b; Gwinnett,

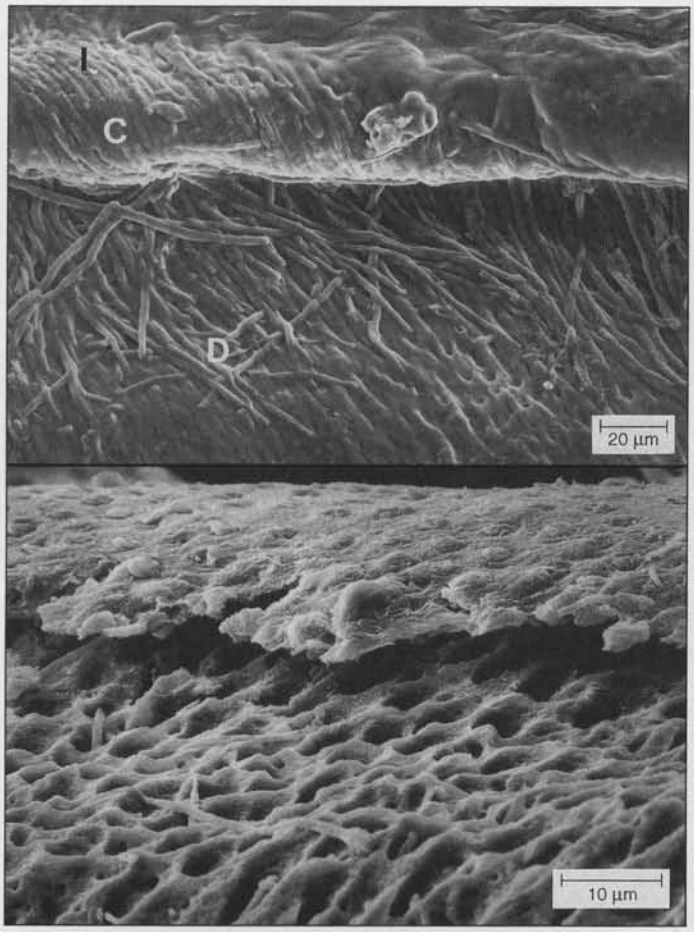

Figure 5. Resin dentin/interfaces of Prisma Universal Bond 3 with (a) demineralized and (b) hypermineralized dentin. I = interface; C = dentin cross-section; D = another plane of dentin.

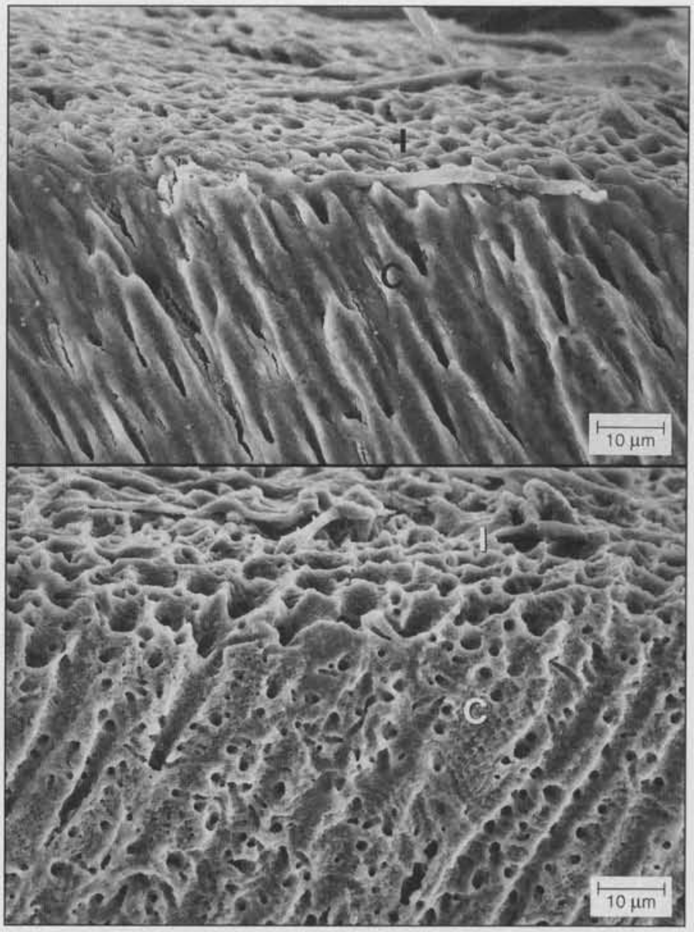

Figure 6. Resin dentin/interfaces of Scotchbond Multi-Purpose with (a) demineralized and (b) hypermineralized dentin. I = interface; C = dentin cross-section.

1992). All-Bond 2 is a modification of a system originally introduced by Bowen *et al.* (1982), and contains 2% NTG-GMA and 16% BPDM (Van Meerbeek *et al.*, 1992). The BPDM molecule has an extra benzene ring, but otherwise is similar to PMDM (Barkmeier *et al.*, 1991a). The PMDM molecule contains two methacrylate groups that polymerize to form insoluble polymers, while the carboxylic acid groups can bind to calcium and other components of enamel and dentin (Johnston and Bowen, 1991). Bowen *et al.* (1982) reported that the aromatic ring of NTG-GMA is electron-rich, while that of PMDM is electron-poor. The two molecules therefore have a mutual affinity, and NTG-GMA initiates polymerization of PMDM. Schumacher *et al.* (1992) hypothesized a synergistic reaction between surface-active monomers containing carboxylic groups, such as PMDM, and surface-active amine activators such as NPG. The chemical complexes formed by these two materials could decompose by an electron transfer mechanism, forming radicals that initiate copolymerization of the monomers.

The bond strengths of All-Bond 2 with normal dentin obtained in this experiment were lower than those reported in several recent studies (Kanca, 1992a,b; Barkmeier *et al.*, 1991a; Gwinnett, 1992), but were similar to those obtained by Triolo and Swift (1992), Barkmeier *et al.* (1991b), Chappell *et al.* (1992), and Kerr Manufacturing Company (1992). Differences in test methods account for some of the differences in bond strengths reported by various laboratories. For example, some researchers condition and prime entire dentin surfaces, while others treat only smaller, circumscribed areas. Researchers may use different crosshead speeds, with faster speeds resulting in lower bond strengths.

Amalgambond Plus etches dentin with a solution of 10% citric acid and 3% ferric chloride (10/3). Citric acid demineralizes the dentin surface, and ferric chloride reportedly prevents collagen denaturation and collapse (Nakabayashi, 1985a,b; Nakabayashi *et al.*, 1992). Etching with the citric acid/ferric chloride solution exposes 1-2 µm of the superficial dentin collagen (Fukushima and Horibe, 1990). Amalgambond primer is a 35% HEMA solution that increases the diffusion of monomer into dentin (Nakabayashi and Takarada, 1992; Nakabayashi *et al.*, 1992), and may be essential both for hybrid zone formation and for a gap-free dentin-resin interface (Nakabayashi *et al.*, 1992). The chemically cured Amalgambond Plus bonding resin contains 5% 4-methacryloyloxyethyl trimellitate anhydride (4-META), HEMA, and methylmethacrylate (MMA), and a tri-

Table 5. Duncan's multiple-range test, shear bond strengths by degree of dentin mineralization

Type of Dentin	n	Mean Shear Bond Strength (MPa)	Duncan Group*
Normal	40	15.46	A
Hypermineralized	40	11.12	B
Demineralized	40	2.74	C

* Values with different letters are significantly different at $p < 0.05$.

Table 6. Modes of failure

Group	n	Cohesive	Adhesive	Mixed
All-Bond 2	30	4 (2)*	23	3 (2)
Normal Dentin	10	4 (2)	4	2 (2)
Hypermineralized Dentin	10	0	9	1
Demineralized Dentin	10	0	10	0
Amalgambond Plus	30	5	19	6 (1)
Normal Dentin	10	3	5	2 (1)
Hypermineralized Dentin	10	2	4	4
Demineralized Dentin	10	0	10	0
Prisma Universal Bond 3	30	1	27	2 (2)
Normal Dentin	10	1	7	2 (2)
Hypermineralized Dentin	10	0	10	0
Demineralized Dentin	10	0	10	0
Scotchbond Multi-Purpose	30	6 (3)	19	5 (1)
Normal Dentin	10	5 (3)	2	3 (1)
Hypermineralized Dentin	10	1	7	2
Demineralized Dentin	10	0	10	0

* Numbers in parentheses denote specimens with visible cracks in the dentin surface.

n-butylborane (TBB) initiator. 4-META is a coupling agent similar to PMDM (Bowen *et al.*, 1982), and is hydrolyzed into 4-MET, which contains both hydrophilic and hydrophobic groups (Ozaki *et al.*, 1991; Hotta *et al.*, 1992).

4-META adhesives have had consistent results in many other experiments, with bond strengths ranging from 15 to 26 MPa (Pashley, 1991; Tao *et al.*, 1991; Gwinnett, 1992; Nakabayashi and Takarada, 1992; Triolo and Swift, 1992). Two factors may be responsible for the relatively high bond strength of Amalgambond Plus. First, the 4-MET molecule may bond both chemically and mechanically to hydroxyapatite. Two different forms of 4-MET occur at the adhesive interface (Ozaki *et al.*, 1991). One of these reacts with calcium ions, and the other forms a copolymer with MMA. Also, the slow chemical polymerization (interfacial initiation of polymerization) and high viscosity of Amalgambond Plus unfilled resin may reduce internal stresses and contribute to its effectiveness as an adhesive (Imai *et al.*, 1991; Pashley, 1991; Van Meerbeek *et al.*, 1992).

Prisma Universal Bond 3 includes a primer and an unfilled resin. The former is 6% PENTA and 30% HEMA in an ethanol solution. PENTA is an adhesion-promoting, weakly acidic, self-etching primer. It may or may not remove the smear layer, depending on the thickness of the smear layer and plugs (Barkmeier and Cooley, 1992; Erickson, 1992). PENTA may also facilitate penetration of resin monomers into dentin surfaces for micromechanical bonding (Van Meerbeek *et al.*, 1992). The adhesive resin contains 50% UDMA, 25% TEG-DMA (a diluent resin), 4.5% PENTA, 0.5% to 1% glutaraldehyde, and a photoinitiator (Albers, 1990a; Johnson *et al.*, 1991; Van Meerbeek *et al.*, 1992; Barkmeier and Cooley, 1992). The presence of a urethane group in the unfilled resin may result in bonding to surface-bound hydroxyl groups (Eliades *et al.*, 1985).

Reported bond strengths for Prisma Universal Bond 2/3 to normal dentin range from 11 to 19 MPa (Barkmeier and Cooley, 1992; Chappell *et al.*, 1992; Gwinnett and Kanca, 1992). In this experiment, the mean bond strengths of Prisma Universal Bond 3 to demineralized dentin and hypermineralized dentin were not statistically different. This finding suggests that collagen and calcium may both be involved in bonding of Prisma Universal Bond 3. On demineralized dentin, Prisma Universal Bond 3 had the highest mean bond strength of the adhesives tested, although the difference was not statistically significant. This may be the result of an interaction of its glutaraldehyde component with dentinal collagen (Albers, 1990a; Barkmeier and

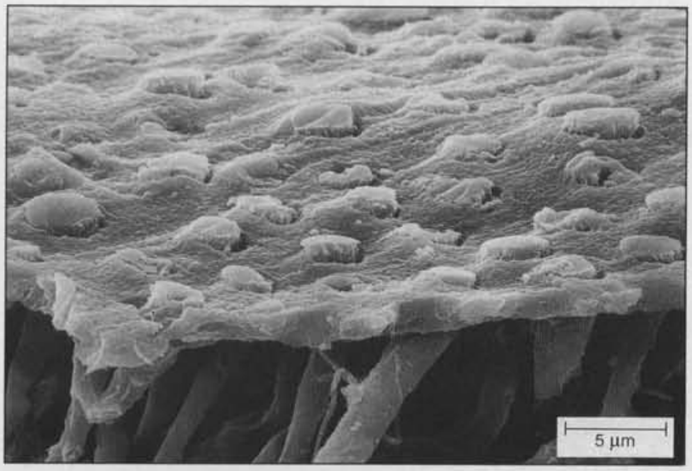

Figure 7. Adhesive failure of Amalgambond to normal dentin.

Cooley, 1992). Bonding to collagen has been associated with glutaraldehyde-containing dentin bonding systems (Munksgaard and Asmussen, 1985). Glutaraldehyde is a very effective protein cross-linking agent (Richard and Knowles, 1968) that bonds to the -NH_2 groups of amino acids such as lysine and hydroxylysine (Eliades *et al.*, 1985). Ionic bonding is evidently another major component of the bonding mechanism of Universal Bond 3, since Barkmeier and Jefferies (1992) reported that dentin conditioning with EDTA, 37% phosphoric acid, or 2.5% nitric acid/NPG decreased the mean bond strengths of Prisma Universal Bond 2 to dentin. Like second-generation phosphonate esters, the bonding mechanism of Prisma Universal Bond 3 could involve ionic bonds between the phosphate groups of PENTA and the calcium ions of the smear layer or the dentin surface (Albers, 1990a). However, its behavior on mineralized dentin in this study did not confirm this hypothesis. Some non-collagenous proteins removed by etching may be necessary for subsequent remineralization (Pashley *et al.*, 1992).

Scotchbond Multi-Purpose is the most recently developed dentin bonding system used in this experiment. It uses an aqueous solution of 10% maleic acid with a polyvinyl alcohol thickener (pH = 1.2) to etch dentin and enamel (3M Dental Products Division, 1992). The primer is an aqueous solution of HEMA and a polyalkenoic acid copolymer similar to that incorporated into Vitrebond glass-ionomer cement (3M Dental Products Division, 1992). The Vitrebond copolymer is a modified polyacrylic acid with polymerizable methacrylate groups. When powder and liquid are mixed, calcium aluminum polyacrylate gel forms as a result of the conventional acid/base glass-ionomer setting reaction. When light-cured, the methacrylate groups form inter-chain covalent bonds. The carboxylic groups of the polyacrylic acid form ionic bonds either with the dentin calcium or with the aluminum ions from the powder (Albers, 1990b; Mitra, 1991; Prati *et al.*, 1992; Smith, 1992). Prati *et al.* (1992) used calcium oxalate to increase the bond strengths of Vitrebond to dentin, verifying this hypothetical bonding mechanism. Scotchbond adhesive is a mixture of Bis-GMA and HEMA, with less than 1.0% hexafluorophosphate.

Other factors may be involved in the bonding of Scotchbond Multi-Purpose to dentin. Scotchbond Multi-Purpose showed a fairly consistent behavior on normal dentin (coefficient of variation = 13.7%), but its bonding to mineralized dentin was unpredictable (coefficient of variation = 45.0%). If ionic interaction with calcium was the main component of bonding, Scotchbond Multi-Purpose should have displayed higher bond strengths to mineralized dentin. Studies with Scotchbond Multi-Purpose are not abundant, because it is a new product. Swift and Triolo (1992) obtained 17.8 MPa on dry dentin and 21.0 MPa on moist dentin, after thermocycling—values that are slightly higher than the ones obtained in this study.

All of the dentin bonding systems used in this study had lower bond strengths to mineralized dentin than to normal dentin. This finding may be related to the partial or complete obliteration of tubules and intertubular dentin by mineral deposition. Occlusion of the tubules by minerals and the increased area occupied by peritubular dentin may preclude the development of a good interpenetration of the bonding systems in dentin (Duke and Lindemuth, 1990). Less resin tag formation is frequently associated with sclerotic substrates (Gwinnett and Jendresen, 1978; Duke and Lindemuth, 1991), and clinical and laboratory evidence shows that bonding to sclerotic dentin is a difficult task (Duke, 1992). Generally, dentin bonding systems do not bond as well to dentin surfaces with increasing age (Heymann *et al.*, 1991). Our results on mineralized dentin confirm that either the patency of the tubule orifices and the mineral content of intertubular dentin are important factors in bonding.

SEM observations failed to demonstrate a consistent resin-dentin interface morphology. Resin detached from the dentin substrate in some areas. Desiccation of dentin during SEM processing may have contributed to this detachment (Suzuki and Gwinnett, 1991) as specimens were vacuum-desiccated for 24 h (Nakabayashi and Takarada, 1992). However, in a pilot study, we treated several specimens with a regimen that included fixation with glutaraldehyde, rinsing with a sodium cacodylate buffer, post-fixation with osmium tetroxide, and dehydration in a graded series of ethanols. The specimens were then either dried in a hexamethyldisilazane (HMDS) solution or were critical-point-dried. These procedures did not improve the quality of the SEM specimens over simple vacuum desiccation. In our experience, fixation and critical-point or HMDS drying are essential only for evaluating the effects of conditioners on the dentin surface.

Discrepancies in the dentin-resin interface morphology may be related to variations in the dentin substrate (Pashley, 1989; Duke and Lindemuth, 1990, 1991). The morphology of the Amalgambond Plus resin-impregnated layer obtained in our study confirms the reports by Nakabayashi (1985a,b) and Nakabayashi and Takarada (1992). The reticular pattern suggests that collagen fibers are present (Fig. 2b). The interface morphology with mineralized dentin is somewhat peculiar.

Even though hybrid or "hybrid-like" layers are evident with All-Bond 2 and Amalgambond Plus (Figs. 3b, 4b), as well as tag necks (Wang and Nakabayashi, 1991; Nakabayashi and Takarada, 1992), the tags are thinner than in normal dentin. Resin penetration with Prisma Universal Bond 3 and Scotchbond Multi-Purpose in mineralized dentin was not consistent (Figs. 5b, 6b). No hybrid or "hybrid-type" layer was evident, which may explain their significantly lower bond strengths to mineralized dentin.

The interface morphology with demineralized dentin was similar for all bonding systems. The bonding area showed a depression, probably related with the collapse of collagen after demineralization. Surprisingly, one of the Prisma Universal Bond 3 specimens apparently showed a "hybrid-type" layer (Fig. 5a) that resisted the acid used for scanning electron microscopy processing. Nakabayashi (1985b) hypothesized that glutaraldehyde may play a role similar to that played by ferric ions as a collagen stabilizing agent, which may explain the presence of this hybrid-type layer as well as the relatively high mean shear bond strength of Prisma Universal Bond 3 to demineralized dentin.

The shear bond strengths of adhesives to normal dentin in this study are similar to those reported in recent studies (Pashley, 1991; Nakabayashi and Takarada, 1992; Swift and Triolo, 1992; Triolo and Swift, 1992). The relatively large standard deviations are a reflection of the differences in dentin substrate which have been described by several authors (Pashley *et al.*, 1978, 1984, 1987; Pashley, 1989; McGuckin *et al.*, 1991; Fowler *et al.*, 1992). The low bond strengths to demineralized dentin suggest that a micromechanical infiltration into etched dentin is a more important factor in adhesion than chemical bonding to collagen (Erickson, 1989; Misra, 1989).

The results with hypermineralized dentin suggest that the partial or total obliteration of the tubules and intertubular dentin with mineral deposits may prevent reliable bonding of resins. The mineral deposits probably prevent adequate etching and resin penetration. Clinically, hypermineralized substrates occur with sclerotic dentin (Duke and Lindemuth, 1991) and beneath carious lesions (Kurosaki *et al.*, 1990). Thus, appropriate mechanical retention should be used in these clinical situations.

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