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ORIGINAL ARTICLE

Potential of dental adhesives to induce mucosal irritation evaluated by the HET–CAM method

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Abstract

Objective. This study was undertaken to determine the potential of dental adhesive products to induce mucosal irritation based on their ability to damage the blood vessels of the chorioallantoic membrane (CAM) in fertilized hen's egg. ***Material and methods.*** Twenty-seven dental adhesive products (total 36 solutions) covering the four adhesive concepts, etch and rinse with two or three steps procedure and self-etch with one or two steps procedure, were evaluated using the hen's egg test–CAM method (HET–CAM). The blood vessels on the CAM of a fertilized hen's egg were used as the test system, and severity of the irritation was based on an assessment of the reaction of the blood and the blood vessels to the test chemical during 5 min of exposure. Three specific end-points – coagulation of blood, lyses of blood and rupture of blood vessel – were evaluated and their time-points for appearance noted. ***Results.*** Coagulation of the blood was the most frequent injury, and was observed within less than a minute's exposure in 25 of the 36 tested solutions. Seventeen of the solutions were rated as moderate irritants and 16 as strong irritants. The type and severity of reaction could not be linked to the type of solvent (water, ethanol, acetone) nor to the presence of 2-hydroxylethyl methacrylate (2-HEMA) in the products. ***Conclusions***. Most dental adhesives damage the blood vessels of the CAM, indicating irritant effects on mucous membranes.

Key Words: *Bonding, coagulation, in vitro, monomers*

Introduction

Dental adhesives were initially introduced to improve the adhesion between resin filling and enamel [1], and have since been modified to include binding to dentin and increased bond strength [2–4]. These modifications have led to the so-called seventh generation of dental adhesives [5]. They now have a wide range of applications comprising proper bonding-luting procedures for direct and indirect esthetic restorations [6,7], coupling pit and fissure sealants to the enamel in young patients [8], and desensitization of hypersensitive root surfaces [9,10].

Dental adhesives can induce inflammation in exposed tissue. Experiments in animals have shown that implanted light-cured dental adhesives induce inflammation and mast cell migration in surrounding tissue shortly after implantation [11,12]. Subsequently, the persistent inflammation induces formation of fibrous tissue surrounding the implants. Placement of dental adhesives on intentionally exposed human pulps has resulted in inflammation of varying severity [13–16]. One review of the current status of pulp capping with dentin adhesives concluded that such treatment was contraindicated [17]. Inflammation has also been observed in the pulp after placement of dental adhesives in deep cavities where only a thin dentin wall separates the pulp and the adhesives [11,18]. Elutes from dental adhesives are toxic to human primary pulp cells *in vitro* [19], and a number of constituents included in the adhesives have been found cytotoxic in other cell systems [20–24].

One test for mucous membrane irritation revealed that most dental adhesives elicited irritant reactions [25]. This so-called HET–CAM method has been used in the evaluation of other dental materials, e.g. denture relining materials [26], denture adhesives [27], dental polymer products [28], and alloys [29,30]. With this test method, the occurrence of adverse vascular changes in the vascularized chorioallantoic membrane (CAM) of embryonated

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chicken eggs after exposure to a test chemical indicates the potential of the chemical to cause irritation [31,32]. This is a relevant test for evaluation of the biocompatibility of dental adhesives with regard both to pulpal and mucous membrane exposure. The techniques employed for placement of dental adhesive agents may result in accidental exposure to adjacent oral soft tissue of the patient, and, due to extensive use, there is a possibility that this may occur in the clinic.

The objective of the present study was to determine the potential of dental adhesive agents to evoke irritation. Since many new products have been introduced to the market since the last survey [4,25], it was felt relevant to extend the former study and include new products on the market.

Material and methods

The materials used are listed in Table I. There are four different concepts for obtaining bond between the restoration and the tooth [33]: *Etch and rinse, three step*, involving an etchant and separate prime and bond constituents (ER, 3 step); *etch and rinse, two step*, using an etchant and a combined prime and bond solution (ER, 2 step); *self-etch, two step*, the etchant and the primer combined and a separate bond solution (SE, 2 step); and *self-etch, one step*, where etchant, prime and bond are combined (SE, 1 step). Four of the tested products were classified as SE 1 step, two as SE 2 step, 14 as ER 2 step and 7 as ER 3 step (Table I). The etching solutions of the "etch and rinse" products were not tested. The compositions of the tested products are given in Table II based on information from the manufacturers taken from their instructions for use or material safety data sheets (MSDS).

The HET–CAM procedure was slightly modified from a previously published method [31]. Fertilized eggs were purchased (Samvirkekylling, Valer, Norway) and placed in an automatically rotating incubator at 37°C in a humidified atmosphere until testing on day 9. Any defective eggs were discharged before testing. The CAM was accessed by removal of the shell above the air cell and inner egg membrane using a dental drill saw blade and forceps. The chemicals (300 μl) were applied directly onto the CAM. The membrane was examined for 5 min using a photo macroscope (Wild M400, Wild, Heerbrugg, Switzerland) with illumination. The illuminating light was filtered through an eye protection shield for light curing unit (MAX eye protection system; DeTrey Dentsply, USA; cut-off at 550 nm) to prevent light-cured materials from curing during the testing. Each product was tested on three eggs, and the experiment repeated once. The CAM was evaluated for the appearance of hemorrhage, vascular lysis, or coagulation [31]. The reader is referred to Dahl & Polyzois [34] for details of the reaction patterns. The irritation score was calculated from the recorded time for each reaction to occur from the following algorithm [31]:

$$\text{irritation score} = [5 \times (301 - \text{H})/300] + [7 \times (301 - \text{L})/300] + [9 \times (301 - \text{C})/300]$$

where H = time in seconds from start of exposure until hemorrhage occurs, L = correspondingly for lysis, and C = for coagulation. The products were classified, based on the mean irritation score of both experiments, as non-irritant (score 0–0.9), slight irritant (score 1–4.9), moderate irritant (score 5–8.9), and strong irritant (score 9–21) [31]. The mean detection time (in seconds) for appearance of coagulation using 100% solution (mtc_{100}) was determined [35]. Differences between groups were tested using the F-test, two-sample for variances of Microsoft Office Excel 2003.

Results

The results are summarized in Tables III and IV. Coagulation was the most frequently observed reaction, and usually it occurred shortly after exposure. The mean time to coagulation (mtc_{100}) was less than a minute for 25 of the 36 tested solutions. Seventeen of the solutions were classified as moderate irritants (score 5–8.9) and 16 as strong irritants (score 9–21) (Table III). The group mean irritation score was statistically significantly lower for the self-etch 1 step group compared to etch and rinse 2 step. The prime solutions of etch and rinse 3 step products were significantly more irritating than the bond solutions of this adhesion concept. No difference in irritation score or mtc_{100} value was observed among products based on different solvents (water, ethanol, acetone) and among those containing 2-HEMA or not (Table IV).

Discussion

The HET–CAM procedure was invented to replace the Draize eye test for ocular irritation [31], but has also been used for assessing the irritant potential in other mucous membranes. In comparing different methods for irritation, the results of the HET–CAM procedure have been found to correlate well with the results of dermal tests for irritation [36]. The results obtained here are therefore applicable in the clinical setting, including both accidental spilling on to the patient's oral mucosa and/or skin of the operator.

Two end-points are used in the HET-CAM method, i.e. the mean time to coagulation and the irritation score based on the observation of coagulation, bleeding, and lysis within the CAM. Coagulation is the end-point that is the best predictor of a chemical inducing clinical irritation and has the

Table I. The dental adhesives included in the study classified according to adhesion concept (SE 1 = self-etch, 1 step; SE 2 = self-etch, 2 steps; ER 2 = etch and rinse, 2 steps; ER 3 = etch and rinse, 3 steps) and function and batch numbers of the tested components

Manufacturer/product	Adhesion concept	Function of tested component	Batch no.
ANA			
Single Bond	ER 2	Prime/bond	00280138
Bisco			
One step	ER 2	Prime/bond	0100012481
All-Bond 2	ER 3	Prime	0100012169
		Bond	0100012169
Coltene			
One Coat Bond	ER 2	Prime/bond	LC306
Detrey Dentsply			
Prime and Bond	ER 2	Prime/bond	0012000207
Xeno III	SE 1	Etch/prime/bond	0207002219
DMG			
Contax	SE 2	Etch/prime	511316
		Bond	509161
ESPE*			
EBS-Multi	ER 3	Prime	102180
		Bond	FW0058261
GC Corporation			
Unifil Bond	SE 2	Etch/prime	306041
		Bond	307021
Heraeus Kulzer			
Gluma Comfort Bond	ER 2	Prime/bond	020023
Gluma One bond	ER 2	Prime/bond	135610
Gluma Solid Bond	ER 3	Prime	010025
		Bond	020031
I bond	SE 1	Etch/prime/bond	10021
Ivoclar Vivadent			
Excite	ER 2	Prime/bond	C35862
Syntac	ER 3	Prime	F65563
		Bond	C15085
Syntac Sprint	ER 2	Prime/bond	B24723
Syntac Single Component	ER 2	Prime/bond	B28533
Kerr			
OptiBond Solo Plus	ER 2	Prime/bond	3-1148
Kuraray			
Clerafil Se Bond	ER 3	Prime	00190A 00185A
		Bond	
3M ESPE			
Promt L-Pop	SE 1	Etch/prime/bond	114345
Adper Promt L-Pop	SE 1	Etch/prime/bond	133199
3M*			
Scotchbond Multi-Purpose	ER 3	Prime	20000212
		Bond	20000208
Scotchbond 1	ER 2	Prime/bond	19991208
Sun Medical & Co			
AQ Bond	ER 2	Prime/bond	FVI2005-09
Ultradent Products			
PermaQuick	ER 3	Prime	32PF
		Bond	32PF
PQ 1	ER 2	Prime/bond	3PHY
Voco			
Solobond M	ER 2	Prime/bond	21837

*The 3M and ESPE merger occurred after manufacture of the tested products, and thus the manufacturers stated in the table were according to the packaging of the tested product.

Table II. Composition of the tested products is according to the manufacturer's information in MSDS or instructions for use. The most used constituents (with CAS number) are tabulated in separate columns. In the column "Other", constituents are named as stated by the manufacturer. CAS number in parentheses, where available

Product	Water 7732-18-5	Acetone 67-64-1	Ethanol 64-17-5	HEMA[1] 868-77-9	TEGDMA[2] 109-16-0	UEDMA[3] 72869-86-4	Bis-GMA[4] 1565-94-2	Polyacrylic acid 9003-01-4	Maleic acid1 10-16-7	Gluther-aldehyde 111-30-8	Other
Self-etch 1 step											
Adper Promt L-Pop				X[5]			X				Methacrylated phosphoric ester, champhorquione, polyalkenoic acid
I Bond	X	X				X				X	4-metacryloxyethyltrimellititanhydrid (4-META) [70293-55-9]
Promt L-Pop											Methacrylated phosphoric ester 98%, dl-kamferkinon 1–2%
Xeno III	X		<35%	<50%		X					Butylated hydroxy toluene (BHT), highly dispersed silicon dioxide, phosphoric acid modified polymethacrylate resin, mono fluoro phosphazene modified methacrylate resin, camphorquinone, ethyl-4-dimethylaminobenzoate (Liquid A and B mixed)
Self-etch 2 steps											
Contax (etch/prime)	X								X		Sodium fluoride
Contax (bond)											Hydrophilic and acidic BIS-GMA based resin matrix
Unifil Bond (etch/prime)			42%	3%							
Unifil Bond (bond)				20%							
Etch and rinse 2 steps											
ANA Single Bond		75%		5%	4%						Methacrylated phosphoric acid 10%, elastomer 5%, catalyst/stabilizer 1%
AQ Bond	<45%	<45%		X		X					Methacrylate monomers (methyl methacrylate [80-62-6], 4-META [70293-55-9] urethane dimethacrylate [72869-86-4], 2-hydroxyethyl methacrylate [868-77-9]) 30%
Excite			<26%	<21%							"Phosphonic acid acrylate" <12%, dimetacrylates <45%
Gluma Comfort Bond			25–50%	10–25%							Poly(metacrylic-oligo-acrylic acid) 5–10%,4-metacryloxyethyltrimellitanhydrid 0–5%
Gluma One Bond		25–50%		10–25%							4-metacryloxyethyltrimellitanhydrid 2.5–10%

Table II (*Continued*)

Product	Water 7732-18-5	Acetone 67-64-1	Ethanol 64-17-5	HEMA[1] 868-77-9	TEGDMA[2] 109-16-0	UEDMA[3] 72869-86-4	Bis-GMA[4] 1565-94-2	Polyacrylic acid 9003-01-4	Maleic acid1 10-16-7	Gluther-aldehyde 111-30-8	Other
One step		40–70%		15–40%							Bipenyldimethacrylate [125086-9] 15–40%, dental glass [65997-18-4] 1–10%
One Coat Bond				35%							Methacrylates 10–40%
OptiBond Solo Plus			20–25%								Alkyl dimethacrylate resins 55–60%, barium aluminoborosilicate glass 5–10%, fumed silica (silicon dioxide) 5–10%, sodium hexafluorosilicate 0.5–1%
Prime and Bond		65%									Urethane dimethacrylate resin [105883-40-7] <20%, dipentaerythritol pentaacrylate phosphate [87699-25-0] <10%, polymerizable dimethacrylate resins [2358-84-1] <10%, polymerizable trimethacrylate resins [3290-92-4] <10%
PQ 1			5–10%	15%							Metacrylat 40–60%, inert filler 35–55%
Scotchbond 1	2–8%		20–50%	5–25%	5–20%		10–30%				Polyacrylic acid co-polymer 5–15%, Methacrylates, organic acids, organic fluoride compound
Solobond M (Voco)		X									
Syntac Sprint	X	<23%		<40%				<11%*	3%		*Methacrylate modified polyacrylic acid <11%
Syntac Single Component	X			<50%				6%*	3%		*Methacrylate modified polyacrylic acid 6%
Etch and rinse 3 steps											
All-bond 2 Primer A+B (prime)		40–70%	10–30%								Bipheny dimethacrylate [125086-31-9] 8–30%, Na-tolylglycine glycidylmethacrylate[133736-31-9]
All-bond 2 D/E resin (bond)				15–40%			15–40%				Methacrylates
Clerafil Se Bond (prime)	X			X							10-methacryloyldihydrogenphosfate, hydrophilic dimethacrylate, N,N-diethanol-p-toluidine, champhorkinone
Clerafil Se Bond (bond)				X			X				10-methacryloyldihydrogenphosfate, hydrophobic dimethacrylate, N, N-diethanol-p-toluidine, champhorkinone, silanated colloidal silica
EBS-Multi (prime)	30–40%			40–50%							2,2′-[[3,5-bis(1,1-dimethylethyl) phenyl]imino]bis-ethanol [64153-50-0] 5–10%, magnesium HEMA ester 5–10%

Table II (*Continued*)

Product	Water 7732-18-5	Acetone 67-64-1	Ethanol 64-17-5	HEMA[1] 868-77-9	TEGDMA[2] 109-16-0	UEDMA[3] 72869-86-4	Bis-GMA[4] 1565-94-2	Polyacrylic acid 9003-01-4	Maleic acid1 10-16-7	Gluther-aldehyde 111-30-8	Other
EBS-Multi (bond)				5–10%	5–10%		20–30%				Bisphenol A bis(3-methacryloyloxy-propyl) ether substituted dimethacrylate [27689-12-9] 30–40%, malonic acid ester 10–20%, 2-propenoic acid, 2-methyl-, 2-[(2-hydroxyethyl)(3-methoxypropyl) amino]ethyl ester [93962-70-0] 1–5%
Gluma Solid Bond P (prime)				25–50%	<2.5%						Maleic acid-mono-2-metacryloyl-oxyethylester [51978-15-5] 0–2.5%, metalacrylic polycarboxyl acid 2.5–10%, maleic acid [110-16-7] 0–5%
Gluma Solid Bond S (bond)					25–50%		25–50%				Maleic acid-mono-2-metacryloyl-oxyethylester [51978-15-5] 0–5%, 2,2-dimethoxy-1,2-diphenylethanone [24650-42-8] <1%
PermaQuick (prime)			20%								Methacrylic acid 7%
PermaQuick (bond)				15%							
Scotchbond Multi-Purpose Primer, (prime)	43–47%			35–40%							Light-cured polymer 10–15%, polyalkenoic acid
Scotchbond Multi-Purpose Adhesive (bond)				30–40%			60–70%				DL-champhorkinon <1%, N,N-dimethylbenzocain <1%, difenyliodoniumhexafluorphosphat <1%
Syntac Primer (prime)	X	<42%							4%		Dimethacrylate
Syntac Adhesive (bond)	X								<0.1%	<5%	Polyethyleneglycoldimethacrylate

[1]HEMA = 2-hydroxyethyl methacrylate.
[2]TEGDMA = 1,8-bis(methacryloyloxy)-3,6-dioxaoctane.
[3]UEDMA = 1,6-bis(methacryloyloxy-2ethoxy-carbonylamino)-2,4,4-trimethyl-hexane.
[4]Bis-GMA = 2,2 bis[4-(2-hydroxy-3-methacryloyloxy-propyl)-N-tolyglycine.
[5]X = The chemical is present, but the amount (concentration) is not stated by the manufacturer.

Table III. Results of the HET–CAM studies describing type of reaction(s), mean time to coagulation for a 100% solution (mtc_{100}), the calculated irritation score (IS) and classification of the products according to severity of the reaction. SD = standard deviation

Product	Reaction	mtc_{100} (in sec) (SD)	IS (SD)	Classification
Self-etch 1 step				
Adper Promt L-Pop	Coagulation	30 (0)	8.1 (0)	Moderate
I Bond	Coagulation /bleeding (1/6)*	35 (11)	8.5 (1.2)	Moderate
Promt L-Pop	Coagulation	30 (0)	8.1 (0)	Moderate
Xeno	Coagulation	30 (0)	8.1 (0)	Moderate
Self-etch 2 steps				
Contax (etch/prime)	Coagulation	30 (0)	8.1 (0)	Moderate
Contax (bond)	Coagulation /bleeding	45 (15)	12.3 (0.6)	Strong
Unifil Bond (etch/prime)	Coagulation	35 (11)	8.9 (2.2)	Moderate
Unifil Bond (bond)	Coagulation /bleeding	40 (14)	14.7 (1.8)	Strong
Etch and rinse 2 steps				
ANA Single Bond	Coagulation/bleeding	30 (0)	11.9 (0.6)	Strong
AQ Bond	Coagulation/bleeding (2/6)	30 (0)	9.9 (0.5)	Strong
Excite	Coagulation/bleeding	30 (0)	8.1 (0)	Moderate
Gluma Comfort Bond	Coagulation/bleeding	30 (0)	8.1 (0)	Moderate
Gluma One Bond	Coagulation/bleeding /lysis	35 (11)	18.1 (0.8)	Strong
One-step	Coagulation/bleeding	40 (14)	9.5 (1.7)	Strong
One Coat Bond	Coagulation/bleeding	85 (29)	9.8 (2.3)	Strong
OptiBond Solo Plus	Coagulation/bleeding (2/6)	65 (11)	8.2 (1.3)	Moderate
Prime and Bond	Coagulation/bleeding (3/6)	30 (0)	9.3 (1.4)	Strong
PQ 1	Coagulation	95 (70)	6.2 (1.2)	Moderate
Scotchbond 1 (bond)	Coagulation/bleeding /lysis (4/6)	60 (30)	12.5 (2.0)	Strong
Solobond M	Coagulation/bleeding	45 (22)	12.0 (1.0)	Strong
Syntac Sprint	Coagulation	30 (0)	8.1 (0)	Moderate
Syntac Single Component	Coagulation/lysis	30 (0)	13.7 (1.5)	Strong
Etch and rinse 3 steps				
All-bond 2 Primer A+B (prime)	Coagulation/bleeding	190 (80)	4.8 (3.4)	Slight
All-bond 2 D/E resin (bond)	Coagulation/bleeding	150 (67)	8.6 (4.9)	Moderate
Clerafil Se Bond (prime)	Coagulation/bleeding (2/6)	30 (0)	8.5 (0.5)	Moderate
Clerafil Se Bond (bond)	Coagulation	65 (27)	7.1 (1.8)	Moderate
EBS-Multi (prime)	Coagulation/bleeding	30 (0)	12.6 (0)	Strong
EBS-Multi (bond)	Coagulation/bleeding	65 (27)	7.1 (1.8)	Moderate
Gluma Solid Bond P (prime)	Coagulation/lysis (3/6)	30 (0)	10.2 (2.1)	Strong
Gluma Solid Bond S (bond)	Coagulation	288 (30)	0.4 (0.9)	Non-irritant
PermaQuick (prime)	Coagulation	35 (11)	8.0 (0.3)	Moderate
PermaQuick (bond)	None	–	0.0	Non-irritant
Scotchbond Multi-Purpose Primer (prime)	Coagulation/bleeding	70 (37)	10.9 (1.1)	Strong
Scotchbond Multi-Purpose Adhesive (bond)	Coagulation/bleeding /lysis (4/6)	55 (32)	14.1 (2.3)	Strong
Syntac Primer (prime)	Coagulation	30 (0)	8.1 (0)	Moderate
Syntac Adhesive (bond)	Coagulation/bleeding /lysis	80 (90)	13.6 (4.5)	Strong

*In cases where the reaction was not observed in all eggs, the number of responding eggs is stated: bleeding (1/6) = Bleeding was observed in 1 out of 6 eggs.

lowest variation and highest reproducibility [35]. Coagulation of the blood vessels was a common finding for all tested solutions. Intravascular coagulation may be initiated by damage to capillaries, the so-called extrinsic pathway, or after trauma to the blood (intrinsic pathway) [37]. It has not been documented which pathway dominates the HET–CAM method, but it is likely that the extrinsic pathway is important in the case of many of the tested products. This is supported by the finding that bleeding was the most commonly observed reaction in combination with coagulation (Table III). Related to the HET–CAM method, bleeding must be regarded as a result of toxic damage to the capillaries.

Sixteen solutions were classified as strong irritants and were found among all adhesive concepts except the newest, namely self-etch 1 step. In this group, all products obtained an irritation score (IS) of 8.1–8.5, which is close to the limit for strong irritant (IS = 9.0). In addition, the self-etch 1 step products had a low mtc_{100} value (<35 s), indicating that they are highly irritating to mucous membranes. It has also been found that products classified as carrying a risk of serious damage to the eye based on other test methods had mtc_{100} values <100 s in the HET–CAM [35]. Most of the tested solutions (32 out of 36) were from this category. In general, dental adhesives were found to have a potential for causing irritation when skin and oral mucosa are exposed,

Table IV. Calculated mean value and standard deviation (SD) of irritation score (IS) and mean time to coagulation (mtc_{100}) for groups related to adhesion concept, solvent used and the stated presence of HEMA

Group	Group mean IS (SD)	Group mean mtc_{100} (SD)
Adhesion concept[a]		
SE 1 ($n=4$)	8.2 (0.2)[d]	31 (2.5) sec[d]
SE 2 ($n=2$)		
Etch/prime	8.5	32 sec
Bond	13.4	42 sec
ER 2 ($n=14$)	10.4 (3.0)[d]	45 (22) sec[d]
ER 3 ($n=7$)		
Prime	9.0 (2.5)[e]	60 (15) sec[f]
Bond	7.3 (5.6)[e]	117 (83) sec[f]
Solvent[b,c]		
Water-based ($n=5$)	11.0 (2.7)[g]	45 (21) sec[g]
Ethanol-based ($n=7$)	8.5 (1.9)[g]	49 (23) sec[g]
Acetone-based ($n=10$)	10.6 (3.2)[g]	33 (5) sec[g]
Use of HEMA[b,c]		
Present ($n=21$)	9.6 (3.6)[h]	51 (31) sec[h]
Not present ($n=14$)	9.2 (3.6)[h]	51 (71) sec[h]

[a]See Material and methods section for definition.
[b]Products from all adhesion concepts merged.
[c]Based on information from manufacturer.
[d]Statistically significant difference between adhesion concepts (SE 1 vs ER 2) ($p<0.05$).
[e]Statistically significant difference between prime and bond ($p<0.05$).
[f]No statistically significant difference between prime and bond ($p>0.05$).
[g]No statistically significant difference among solvent groups ($p>0.05$).
[h]No statistically significant difference among HEMA and non-HEMA groups ($p>0.05$).

and the adhesion concept was not a predictor for the irritant potential of the products. Similar potential for causing irritation has been found for other uncured dental materials [28]. Chemicals to be polymerized are usually chemically reactive, and thus a biological reactivity is also to be expected.

Another explanation for the irritant reaction could be the composition of the product. The composition of dental adhesives is complex and variable (Table II) [38]. Most dental adhesives, except the bond solution of ER 3 step and SE 2 step products, contain solvent that evaporates from the cavity by air blasting during the bonding procedure. Water, ethanol, and acetone are used as solvent, and it has been suggested that hydrophilic solvents like ethanol and acetone play a major role in development of the irritation reaction in the CAM [25]. The present study, more comprehensive than the study of Dahl [25], did not confirm this finding: There was no significant difference between products based on various solvents. This was unexpected, as both ethanol and acetone are more toxic than water. However, this was a very crude evaluation based on the presence or non-presence of a chemical. Since a toxicological reaction is concentration-dependent and the concentration of the solvent in the final products varies, the result of the comparisons may have been different had the concentrations of the chemicals been taken into the evaluation. Unfortunately, this was not possible as the information provided by the manufacturer does not state the exact content.

2-hydroxyethylmethacrylate (2-HEMA) is a common constituent of dental adhesives found in 5 out of 7 dental adhesives [38] and, according to the manufacturer's information, in 60% of the tested products in the present study. 2-HEMA is a small molecule that readily penetrates skin and mucous membranes, and has been reported as a frequent course of contact dermatitis of either irritant or allergenic etiology [39]. In elucidating the mechanism of 2-HEMA-toxicity, 2-HEMA was found to be an inflammatogenic substance in mice [40]. It was therefore expected in the present study that the irritant reactions of products containing HEMA would exceed those of non-HEMA products, but this was not confirmed experimentally. The lack of exact product composition made proper comparison difficult for this aspect, too.

Dental adhesives have the potential to cause an irritant reaction if oral mucosa is exposed in the patient. Care should therefore be taken to avoid accidentally spilling dental adhesives.

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