

Release of monomers in patient saliva following bonding of orthodontic brackets

Håkon Gulliksen¹, Hilde M. Kopperud², Marit Midtbø³, Hanne Wellendorf²,
Nils Roar Gjerdet^{4,*}

¹Private Orthodontic Practice, Stord Tannregulering AS, Hamnegata 2, NO 5411 Stord, Norway

²Nordic Institute of Dental Materials (NIOM), Sognsveien 70A, NO 0855 Oslo, Norway

³Private Orthodontic Practice, Midtbø Tannregulering, Bønneslien 49, NO 5155 Bønnes, Norway

⁴Faculty of Medicine, Department of Clinical Dentistry, University of Bergen, Årstadveien 19, NO 5009 Bergen, Norway

*Corresponding author. Faculty of Medicine, Department of Clinical Dentistry, University of Bergen, Årstadveien 19, Bergen NO 5009, Norway.

E-mail: gjerdet@uib.no

Abstract

Introduction: This study aimed to investigate whether orthodontic treatment with fixed appliances increased concentration of various monomers in patients' saliva.

Methods: The study included 30 patients aged 10–18. Saliva samples were collected at three time points: before the start of active treatment, immediately after the bonding of orthodontic brackets and during the first follow-up visit between 4 and 6 weeks after bonding. Gas chromatography/mass spectrometry (GC/MS) and liquid chromatography/mass spectrometry (LC/MS) were employed to analyse the samples.

Results: Measurable levels of poly-EGDMA, HEMA, and BMAEPH were detected in saliva samples collected immediately after bracket bonding. Concentrations varied among patients. However, these substances were not quantifiable in saliva 4–6 weeks after bracket placement.

Conclusions: The bonding of orthodontic brackets is associated with a transient increase in the concentration of bonding material constituents in saliva immediately after placement. The findings suggest that, using the current analytical methods, this release is temporary.

Keywords: orthodontic bonding; brackets; saliva; chemical analysis; monomer release

Introduction

Orthodontic treatment involving fixed appliances relies on the attachment of brackets to the teeth. Various types of brackets are affixed using bonding materials applied between the bracket base and the enamel. This bonding material can be manually applied, or the adhesive may come preapplied to the bracket base by manufacturers in the form of precoated brackets.

Orthodontic treatment is commonly administered to children and adolescents. In Norway, ~40% of children aged 13–14 undergo such treatment [1]. Similarly, in Finland, it has been reported that nearly one-third of children under 12 years old have received some form of orthodontic treatment [2]. These orthodontic appliances typically remain in the mouth for ~1.5 to 2 years. Thus, it is of interest to investigate whether the use of bonded brackets exposes individuals to chemical substances.

Polymer-based adhesives for attaching orthodontic brackets are akin to polymer-based restorative materials and usually consist of methacrylate monomers, inorganic fillers, and initiation systems to enable light curing [3]. Typically, a bonding promoter, often termed a 'primer', is applied to the enamel surface before attaching the bracket with the adhesive.

Questions about the biocompatibility of orthodontic adhesives arose shortly after their introduction [4, 5]. Studies have revealed cytotoxic effects [5, 6] as well as genotoxic effects of

dental monomers [7–9]. Methacrylate-based monomers can cause adverse reactions and contact allergy to their constituents has been documented in sensitized individuals, as evidenced in case reports related to orthodontic adhesives [10–13].

There is limited information available regarding the release of substances from orthodontic adhesives, whereas many have reported on elution from polymer-based dental materials in general [14, 15]. To our knowledge, only a few studies have been carried out on patient saliva with polymer-based filling materials [16, 17]. It has been recommended that the biological properties of acrylate-based composites should be further investigated in patients [18], particularly regarding exposure during treatment with fixed appliances [19, 20].

The aim of this study was to assess the concentration in saliva of substances associated with a standard bonding procedure for the attachment of orthodontic brackets. The hypotheses were (i) that monomers will be released to patient saliva related to bonding of brackets, and (ii) that the release will be detectable also at recall after 4–6 weeks.

Material and methods

Saliva was sampled from patients scheduled for orthodontic treatment with fixed appliances at the Orthodontic Clinic at the Department of Clinical Dentistry, University of Bergen,

Table 1. Products used for bonding of orthodontic brackets.

Product (Manufacturer)	Use	Composition (from manufacturers)
Transbond Plus Color Change Adhesive (3 M Unitek) Brackets pre-coated by manufacturer	Adhesive paste for bracket bonding	Polyethylene glycol dimethacrylate (5–15 wt%) Citric acid dimethacrylate oligomer (1–10 wt%) Bisphenol A diglycidyl ether dimethacrylate (<2 wt%) Butylated hydroxytoluene (BHT) < 0.5 wt% (+inorganic particles)
Transbond Plus Self-Etching Primer (3 M Unitek) Delivered as an integrated blister pack	Priming of tooth surface prior to bracket/paste application	2-propenoic acid, 2 methyl phosphinicobis(oxy-2,1 ethandiyl)ester (=BMAEPH) (25–40 wt%) Mono HEMA phosphate (10–25 wt%) Tris[2-(methacryloyloxy)phosphate (1–10 wt%) DL-camphoroquinone (<3 wt%) N,N-dimethylbenzocaine (<3 wt%) Dipotassium hexafluorotitanate (<3 wt%)

Information on components from safety data sheets for the products involved in the study, issued by the manufacturers.

Table 2. Reference materials and solvents used for analyses of patient saliva by GC/MS and LC/MS.

Chemicals	CAS number	Manufacturer
Poly(ethylene glycol)dimethacrylate (Poly-EGDMA) Mixture of ethyleneglycol dimethacrylates, average Mn: 550	25852-47-5	Sigma-Aldrich Co, St. Louis MO
Bis[2-(methacryloyloxy)-ethyl]phosphate (BMAEPH) Mixture of Bis- and Tris-[2-(methacryloyloxy)-ethyl] phosphate	32435-46-4	Sigma-Aldrich Co, St. Louis, MO
Bis-GMA	1565-94-2	Polysciences Inc, Warrington, PA
HEMA (puriss)	868-77-9	Fluka Chemie, Buchs, Switzerland
TEGDMA	109-16-0	Polysciences Inc, Warrington, PA
Amyl methacrylate (Internal Standard for HEMA)	7336-27-8	Polysciences Inc, Warrington, PA
1,6 Hexanedioldimethacrylate (Internal Standard for TEGDMA)	6606-59-3	Esschem Co, Essington, PA
Elix water (Grade 2, deionized)	7732-18-5	Merck Millipore, Darmstadt, Germany
Ethyl acetate for spectroscopy	141-78-6	VWR International, Leuven, Belgium
Methanol (gradient grade)	67-56-1	Merck, Darmstadt, Germany
Acetonitrile (pro analyze)	75-05-8	Merck, Darmstadt, Germany

Norway. The patients were included consecutively during a 2-month period. The sample size, based on 95% confidence level, was estimated to 30; however, there is lack of comparable data.

Patient inclusion

The inclusion criteria were patients' age between 10 and 18 years and with no prior completed orthodontic treatment. Patients were enrolled sequentially, according to the order in which they commenced treatment at a university specialist clinic. A total of 30 patients were included. The methods and aim of the research project were explained to the patients, and written informed consent was obtained. Each patient received 10–12 brackets. Four patients had received cemented molar bands and/or bite raising several months prior to bracket placement. In 9 patients, bite raising was performed after bracket bonding and saliva sampling. One patient was excluded due to earlier orthodontic treatment and missed appointments.

Bonding materials and procedure

The products for bonding are described in Table 1. Information on the chemicals used in the analytical part of

the study is given in Table 2. The brackets used had preapplied adhesive (precoated brackets) of the same product series from a single manufacturer (Victory Series Brackets, 3 M, Monrovia, CA, USA). The bonding procedures were done by one operator (HG) according to the manufacturers' recommendations. An adhesion primer (delivered as an integrated blister pack) was applied on the tooth before placement of the precoated bracket. Bite raising (in 9 patients) was done with Ultra Band Lok (Reliance Orthodontic Products, Itasca, IL, USA) after bracket bonding and saliva sampling.

Light curing was done from two directions, each for 20 seconds, with a curing light delivering 1400 mW/cm² (Acteon MiniLED, Mérégnac, France).

Saliva sampling

The timeline for sampling saliva is illustrated in Fig. 1. Before beginning of treatment, the patients were instructed to rinse the mouth for approximately 15 seconds with distilled deionized water (Millipore Milli-Q, Bedford, MA, USA) and spit it out. They were then given 2 ml of distilled deionised water as a starter volume and asked to swish it around the oral cavity for about 30 seconds and continued to collect unstimulated saliva into a graduated glass tube (Brand, Wertheim, Germany),

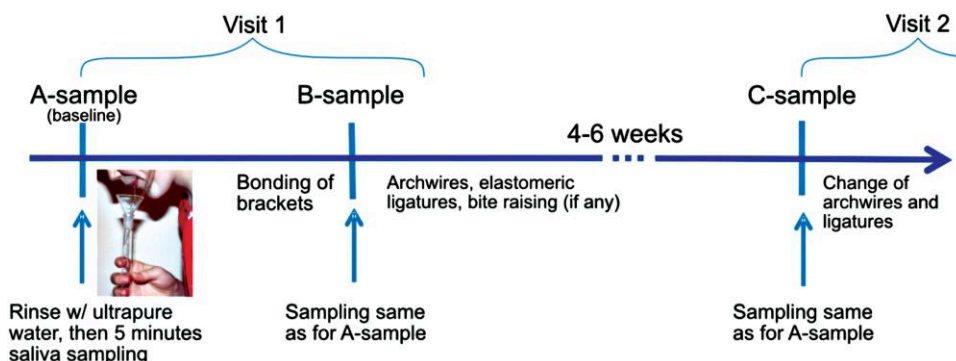


Figure 1. Timeline for saliva sampling for analyses of released monomers associated with bonding of orthodontic brackets.

with a glass funnel. Saliva was collected for a total of 5 minutes. This served as the reference sample for each individual patient (A-sample). The samples were frozen immediately and stored at -20°C .

Immediately after placement of the orthodontic bracket and removal of excess material the bonding was light-cured. After bonding of one jaw, the patients rinsed briefly with distilled deionized water. The 5 minutes saliva sampling process was then repeated (B-sample).

Upon returning for the first control after 4–6 weeks, the third and final sample (C-sample) was collected as described above, before further examination or treatment. The samples were frozen immediately at -20°C .

Saliva sample preparation

The samples were brought to NIOM in frozen condition and placed in a freezer at -20°C until preparation of solutions for analysis by Solid Phase Extraction (SPE). The samples were thawed and monomers extracted using separate SPE columns (HF Bond Elut C18, Agilent Technologies, Santa Clara, USA). The SPE columns were conditioned using 3 ml methanol and 3 ml deionized water. Subsequently 1 ml of saliva was placed on the SPE-column and the column was rinsed for impurities with 1 ml ethanol/water solution (ratio 1:9). Extraction from the individual SPE-columns was done with 1 ml ethyl acetate, then 1 ml methanol, and finally 1 ml acetonitrile, which were all collected. An amount of 500 μl of each sample was used for GC/MS (gas chromatography/mass spectrometry) analysis and another 500 μl for LC/MS (liquid chromatography/mass spectrometry) analysis.

Analysis

To determine and quantify the monomers in the saliva, both GC/MS and LC/MS were used for the investigation of volatile and nonvolatile substances. Initial screening analysis for the detection of monomers in the materials was performed. Analysis of each saliva sample was performed at least twice. GC/MS was used to analyse the amounts of HEMA (2-hydroxyethyl methacrylate) and TEGDMA (triethylene glycol dimethacrylate) using separate internal standards for each chemical, while LC/MS was applied to analyse Poly-EGDMA (poly (ethylene glycol) dimethacrylate), BMAEPH (bis[2-(methacryloyloxy)-ethyl]phosphate) and Bis-GMA (bisphenol A-glycidyl methacrylate). The presented quantitative results are of monomer concentrations in

patient saliva by backtracking the dilution effects and using recovery estimates from the SPE procedure.

GC/MS

The qualitative and quantitative analyses of volatile monomers were performed with an Agilent 7890A GC system, 5975C VL MSD with triple-axis detector, and a HP-5MS column (Agilent Technologies). For quantitative analyses, internal standards were added to each analysed solution. The following temperature program was used for quantification: 100°C (4.5 min)– $50^{\circ}\text{C}/\text{min}$ – $250^{\circ}\text{C}/\text{min}$ (4 min) combined with a selective ion monitoring (SIM). Limit of detection (LOD) calculated as per volume of saliva were for HEMA 250 ng/ml and for TEGDMA 2.2 ng/ml, whereas the limits of quantification (LOQ) were determined to be 750 ng/ml for HEMA and 6.6 ng/ml for TEGDMA.

LC/MS

The qualitative and quantitative analysis of the content of nonvolatile monomers in the samples was conducted with an Agilent 1100 series LC/MSD Ion Trap XCT liquid chromatograph—mass spectrometer (LC/MS), with a Zorbax XDB-C8 5 μm column (Agilent Technologies Santa Clara, CA USA). The LC-system was connected to the mass spectrometer with an electrospray ionization interface. The quantitative analyses were done with isocratic elution with methanol/water (80/20 vol/vol), with 10 mM ammonium acetate. LOD calculated as per volume of saliva were for Poly-EGDMA 150 ng/ml, BMAEPH 300 ng/ml, and Bis-GMA 120 ng/ml, whereas the LOQs were determined to be 450 ng/ml for Poly-EGDMA, 900 ng/ml for BMAEPH, and 360 ng/ml for Bis-GMA.

Statistics

The original data were presented in figures with box-plots and dot plots. As the data were not normally distributed, gamma distribution was used in the analyses. Variance estimates were computed based on bootstrapping using 1000 replicates. The glm module in Stata version 13.1 was used for the statistical analysis [21].

The sample size was calculated based on a precision of margin error of 5, assuming a standard deviation of 15 for a 90% confidence interval. This resulted in a sample size of 27 for the calculation of the confidence limits.

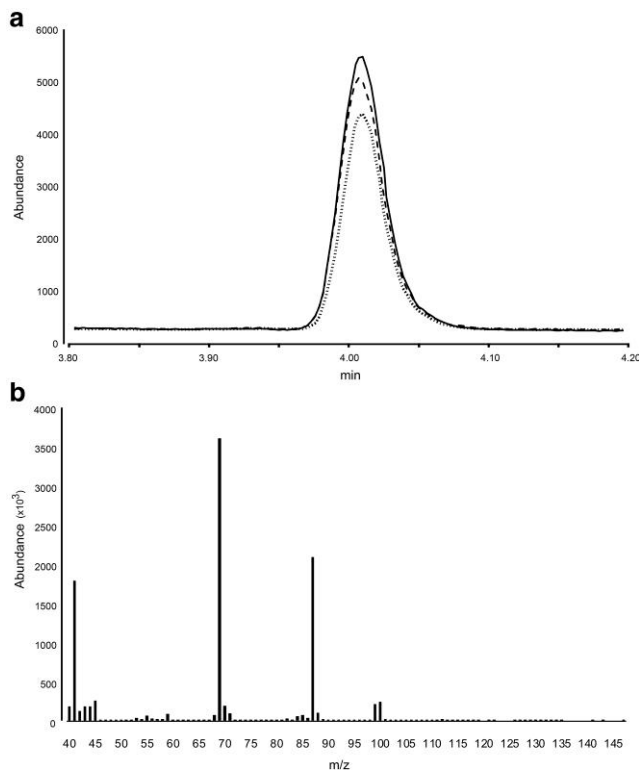


Figure 2. a) Example of chromatograms of HEMA (retention time 4.01 min) in saliva from three patients. b) Mass spectrum of HEMA reference material.

Results

The initial screening for monomer composition in the used bonding materials indicated the presence of additional substances to those listed in the materials safety data sheets (Table 1). In the uncured material Transbond Plus Color Change Adhesive we detected small amounts (not quantified) of the initiator camphorquinone, and the co-initiator ethyl-4-dimethyl aminobenzoate. In the uncured material Transbond Self Etching Primer we detected the undeclared substances HEMA (possibly from declared mono HEMA phosphate), and TEGDMA (triethylene glycol dimethacrylate) including small amounts of EGDMA (ethylene glycol dimethacrylate) and DEGDMA (diethylene glycol dimethacrylate).

Analyses of the B-samples (collected after bracket bonding) showed presence in saliva of the components poly-EGDMA, BMAEPH, and HEMA. Example chromatograms of HEMA (GC/MS) for three patients including mass spectrogram of the reference are presented in Fig. 2. The detected monomers were quantified and the amounts found in the saliva of each patient were calculated (Fig. 3). There were no detectable levels of other monomers found in the B-samples.

From the saliva samples taken before treatment (A-samples) and in the samples taken when returning for control (C-samples), no monomers could be detected. Substances from the bite raising material, applied after bonding and collection of B-samples, could not be detected in the saliva samples taken 4–6 weeks after bracket bonding (C-samples). Nor were any monomers from the pretreatment cementation of molar bands detected in the A-samples.

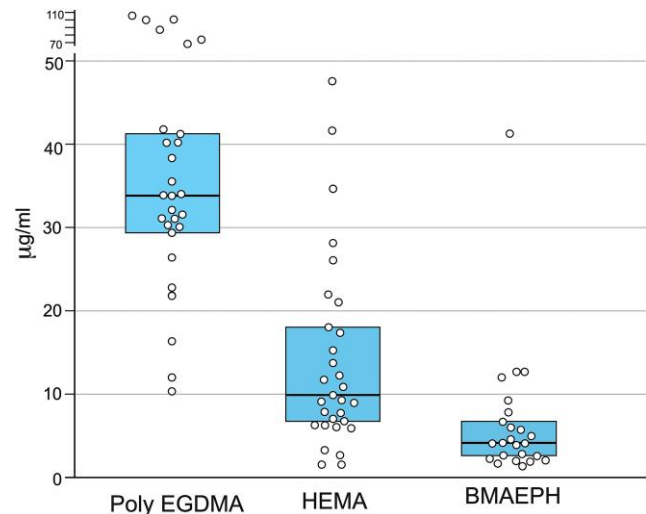


Figure 3. Boxplots and dotplots of quantifiable monomers in saliva after bracket bonding and initial clean-up (B-sample). Circles represent results from individual patients.

Discussion

Orthodontic appliances affixed with bonding materials have the potential to release components or degradation products that may be transferred into saliva. Safety data sheets indicate that the chemical composition of polymer-based orthodontic adhesive systems resembles that of restorative composites and related bonding products [3]. This study confirms that methacrylate-based monomers are used; however, the initial screening in material composition showed that not all monomers are listed in the safety data sheets.

Numerous *in vitro* elution tests employing a wide range of incubation media have been conducted to evaluate the leaching of substances from restorative materials [14, 15, 22]. However, relatively few studies have studied the release of substances from various polymer-based dental materials in actual patients [16, 17]. Even fewer data are available regarding the oral exposure associated with polymeric orthodontic materials in patients, these are often related to bisphenol-A [23, 24]. *In vivo* release into saliva appeared to be low and transient, with the highest concentrations occurring shortly after the initial insertion [23].

In the present study, a standardized clinical procedure was employed, utilizing self-adhesive brackets in which the polymer adhesive was preapplied to the bracket base by the manufacturer [25]. Before attaching the brackets to the enamel surface, an adhesive promoter (primer) was applied, followed by the removal of any excess material before light curing. This procedure likely results in a reduced excess of adhesive material when compared to adhesives applied manually by the operator.

The early release of monomers after the bonding process could be attributed to the oxygen inhibition of the bonding material [26], or to excess material remaining on the tooth surface, particularly from the low-viscosity primer. Based on the current data, it appears that orthodontic bracket bonding materials are not a prolonged source of leachable substances. These findings align with previous observations made with polymer-based filling materials [16, 17].

An extensive *in vitro* study addressed roughness alterations and compound release from two types of composite materials

used for attachments for thermoformed aligners [27]. Release of monomers (TEGDMA, Bis-GMA, UDMA) were detected after 1 week of simulated use in water. Also HEMA was detected, likely originating from the bonding material. Screening for untargeted substances identified hydrolysates as degradation products, highlighting the complexity and scatter of analyses of *in vivo* saliva from individual patients. Due to direct abrasion and greater exposed surface area compared to bracket bonding, these attachments are expected to exhibit higher compound release than in the present study. Recently, an artificial saliva model for analysis of leachable monomers was published that may allow for clinically relevant results without using patient saliva [28].

The present results revealed notable variations in substance concentrations among patients (Fig. 3). Even under well-controlled *in vitro* studies, there is analytical scatter particularly of degradation products [27]. In a clinical setting, this variability is most likely higher due to varying removal of excess material. Additionally, individual saliva properties and fluid dynamics during sample collection may contribute to the variations in concentrations observed among patients.

It is worth noting that the primer used in the bonding process may remain on the teeth and represent a source of substances like HEMA. The manufacturer's safety data sheet lists the primer's composition, which includes mono HEMA phosphate (10–25 wt%). It is possible that this substance could be cleaved down into HEMA, either *in vivo* or before analysis, as well as in the product container before use. It is possible that HEMA is present in the primer as a contaminant. HEMA is also listed as an ingredient in the material for biteraising and/or band cementation, which was performed for some patients months before the bracket placement. However, no HEMA was found in the A-samples indicating that these materials were not the source of the HEMA found in the saliva after bonding.

Low-level exposure to monomers and associated additives may potentially lead to biological effects, such as allergic reactions [10, 12, 13]. Exposure to substances such as HEMA, commonly used in bonding materials, could have adverse effects on the cellular-level [29]. The data from this study suggest that exposure remains low throughout the treatment period.

Given that the primer may act as a source of released monomers, it might be worthwhile to explore a no-primer bonding procedure [30], even though the exposure is transient.

Furthermore, undeclared substances in products were detected. This emphasizes the importance of accurate and comprehensive disclosure of material composition by manufacturers [31, 32].

Within the limitations of this study, the primary hypothesis—that monomers would be released into patient saliva following bracket bonding—was confirmed. However, since no monomers were detected after 4–6 weeks, the secondary hypothesis was rejected.

Conclusion

Considering the limitations of this *in vivo* study, the release of monomers into patient saliva associated with bracket bonding seems to be temporary as no exposure was detected at patient recall.

Acknowledgement

The authors thank Ms. Inger Kleven, a retired colleague, for her expert assistance with the LC-MS analyses.

Author contributions

Håkon Gulliksen (Conceptualization [equal], Data curation [equal], Investigation [equal], Project administration [equal], Writing—original draft [equal], Writing—review & editing [equal]), Hilde M. Kopperud (Data curation [equal], Formal analysis [equal], Methodology [equal], Resources [equal], Validation [equal], Writing—original draft [equal], Writing—review & editing [equal]), Marit Midtbø (Conceptualization [equal], Investigation [equal], Methodology [equal], Project administration [equal], Supervision [equal], Writing—review & editing [equal]), Hanne Wellendorf (Data curation [equal], Methodology [equal], Validation [equal], Writing—review & editing [equal]), and Nils Roar Gjerdet (Conceptualization [equal], Funding acquisition [equal], Investigation [equal], Methodology [equal], Project administration [equal], Supervision [equal], Writing—original draft [equal], Writing—review & editing [equal])

Conflict of interest

The authors declare no conflicts of interest. All authors agreed on publication.

Funding

Funding was provided by University of Bergen – Department of Clinical Dentistry, and Nordic Institute of Dental Materials.

Data availability

The datasets generated and analysed during the current study are not publicly available due to limitations related to ethics approvals, but parts could be available from the corresponding author on reasonable request.

Ethics

The study protocol, including requirements for age-adapted informed consent, was approved by the Regional Committee for Medical and Health Research Ethics of Western Norway, Ref: 2013/1752/REK vest.

References

- Ekornrud T, Skjøstad O, Texmon I. Orthodontic treatment among children and young adults. An analysis of the course of treatment and socioeconomic differences [Tannregulering blant barn og unge. En analyse av behandlingsforløp og sosioøkonomiske forskjeller]. Statistisk sentralbyrå/Statistics Norway 2019. https://www.ssb.no/helse/artikler-og-publikasjoner/_attachment/398789?_ts=16d7296cec8.
- Arpalahti A, Saarnio-Syrjalainen A, Laaksonen S *et al.* Early orthodontic treatment in a Finnish public health centre: a retrospective cross-sectional study. *Acta Odontol Scand* 2023;81:396–401. <https://doi.org/10.1080/00016357.2022.2161623>
- Peutzfeldt A. Resin composites in dentistry: the monomer systems. *Eur J Oral Sci* 1997;105:97–116. <https://doi.org/10.1111/j.1600-0722.1997.tb00188.x>
- Fredericks HE. Mutagenic potential of orthodontic bonding materials. *Am J Orthod* 1981;80:316–24. [https://doi.org/10.1016/0002-9416\(81\)90293-1](https://doi.org/10.1016/0002-9416(81)90293-1)
- Tell RT, Sydiskis RJ, Isaacs RD *et al.* Long-term cytotoxicity of orthodontic direct-bonding adhesives. *Am J Orthod*

- Dentofacial Orthop* 1988;93:419–22. [https://doi.org/10.1016/0889-5406\(88\)90101-1](https://doi.org/10.1016/0889-5406(88)90101-1)
6. Geurtsen W. Substances released from dental resin composites and glass ionomer cements. *Eur J Oral Sci* 1998;106:687–95. <https://doi.org/10.1046/j.0909-8836.1998.eos10602ii04.x>
 7. Arossi GA, Dihl RR, Lehmann M *et al.* Genetic toxicology of dental composite resin extracts in somatic cells *in vivo*. *Basic Clin Pharmacol Toxicol* 2010;107:625–9. <https://doi.org/10.1111/j.1742-7843.2010.00541.x>
 8. Arossi GA, Lehmann M, Dihl RR *et al.* Induced DNA damage by dental resin monomers in somatic cells. *Basic Clin Pharmacol Toxicowql* 2010;106:124–9. <https://doi.org/10.1111/j.1742-7843.2009.00479.x>
 9. Samuelsen JT, Holme JA, Becher R *et al.* HEMA reduces cell proliferation and induces apoptosis *in vitro*. *Dent Mater* 2008;24:134–40. <https://doi.org/10.1016/j.dental.2007.08.006>
 10. Connolly M, Shaw L, Hutchinson I *et al.* Allergic contact dermatitis from bisphenol-A-glycidylmethacrylate during application of orthodontic fixed appliance. *Contact Dermatitis* 2006;55:367–8. <https://doi.org/10.1111/j.1600-0536.2006.00932.x>
 11. Kiviat J, Fleming Y. Orthodontic appliance intolerance due to dental adhesive allergy. *Dermatitis* 2018;29:349–50. <https://doi.org/10.1097/DER.0000000000000423>
 12. Barber SK, Dhaliwal HK. Allergy to acrylate in composite in an orthodontic patient: a case report. *J Orthod* 2018;45:203–9. <https://doi.org/10.1080/14653125.2018.1476037>
 13. Peterson MR, Wong PH, Dickson SD *et al.* Allergic stomatitis from orthodontic adhesives. *Mil Med* 2017;182:e1883–5. <https://doi.org/10.7202/MILMED-D-16-00232>
 14. Van Landuyt KL, Nawrot T, Geebelen B *et al.* How much do resin-based dental materials release? A meta-analytical approach. *Dent Mater* 2011;27:723–47. <https://doi.org/10.1016/j.dental.2011.05.001>
 15. De Angelis F, Sarteur N, Buonvivero M *et al.* Meta-analytical analysis on components released from resin-based dental materials. *Clin Oral Investig* 2022;26:6015–41. <https://doi.org/10.1007/s00784-022-04625-4>
 16. Michelsen VB, Kopperud HB, Lygre GB *et al.* Detection and quantification of monomers in unstimulated whole saliva after treatment with resin-based composite fillings *in vivo*. *Eur J Oral Sci* 2012;120:89–95. <https://doi.org/10.1111/j.1600-0722.2011.00897.x>
 17. Vervliet P, De Nys S, Duca RC *et al.* Degradation products of resin-based materials detected in saliva *in vivo*. *Clin Oral Investig* 2023;27:7189–98. <https://doi.org/10.1007/s00784-023-05075-2>
 18. Goldberg M. *In vitro* and *in vivo* studies on the toxicity of dental resin components: a review. *Clin Oral Investig* 2008;12:1–8. <https://doi.org/10.1007/s00784-007-0162-8>
 19. Kloukos D, Pandis N, Eliades T. Bisphenol-A and residual monomer leaching from orthodontic adhesive resins and polycarbonate brackets: a systematic review. *Am J Orthod Dentofacial Orthop* 2013;143:S104–112.e2. <https://doi.org/10.1016/j.ajodo.2012.11.015>
 20. Gupta SK, Saxena P, Pant VA *et al.* Release and toxicity of dental resin composite. *Toxicol Int* 2012;19:225–34. <https://doi.org/10.4103/0971-6580.103652>
 21. McCullagh P, Nelder JA. *Generalized Linear Models*, 2nd ed. Boca Raton, FL: Chapman & Hall/CRC, 1989.
 22. Hampe T, Wiessner A, Frauendorf H *et al.* Monomer release from dental resins: the current status on study setup, detection and quantification for *in vitro* testing. *Polymers (Basel)* 2022;14:1790. <https://doi.org/10.3390/polym14091790>
 23. Kang YG, Kim JY, Kim J *et al.* Release of bisphenol A from resin composite used to bond orthodontic lingual retainers. *Am J Orthod Dentofacial Orthop* 2011;140:779–89. <https://doi.org/10.1016/j.ajodo.2011.04.022>
 24. Seifi S, Mirzakouchaki B, Rafighi A *et al.* Evaluation of the bisphenol released in the saliva after residual adhesive removal in orthodontic patients by using ultrasonic scaling and rotary system: a single-center randomized clinical trial. *Am J Orthod Dentofacial Orthop* 2023;163:148–53. <https://doi.org/10.1016/j.ajodo.2022.06.023>
 25. Alakttash AM, Fawzi M, Bearn D. Adhesive precoated bracket systems and operator coated bracket systems: is there any difference? A systematic review and meta-analysis. *Angle Orthod* 2019;89:495–504. <https://doi.org/10.2319/051818-373.1>
 26. Jagdish N, Padmanabhan S, Chitharanjan AB *et al.* Cytotoxicity and degree of conversion of orthodontic adhesives. *Angle Orthod* 2009;79:1133–8. <https://doi.org/10.2319/080808-418R.1>
 27. Iliadi A, Zervou SK, Koletsi D *et al.* Surface alterations and compound release from aligner attachments *in vitro*. *Eur J Orthod* 2024;46:cjae026. <https://doi.org/10.1093/ejo/cjae026>
 28. Grutle LA, Holm HV, Kopperud HBM *et al.* Validation of a human saliva model for the determination of leachable monomers and other chemicals from dental materials. *J Chromatogr B* 2024;1236:124073. <https://doi.org/10.1016/j.jchromb.2024.124073>
 29. Becher R, Valen H, Olderbo BP *et al.* The dental monomer 2-hydroxyethyl methacrylate (HEMA) causes transcriptionally regulated adaptation partially initiated by electrophilic stress. *Dent Mater* 2019;35:125–34. <https://doi.org/10.1016/j.dental.2018.11.008>
 30. Bazargani F, Magnuson A, Lothgren H *et al.* Orthodontic bonding with and without primer: a randomized controlled trial. *Eur J Orthod* 2016;38:503–7. <https://doi.org/10.1093/ejo/cjv075>
 31. Nicol AM, Hurrell AC, Wahyuni D *et al.* Accuracy, comprehensibility, and use of material safety data sheets: a review. *Am J Ind Med* 2008;51:861–76. <https://doi.org/10.1002/ajim.20613>
 32. Henriks-Eckerman ML, Kanerva L. Product analysis of acrylic resins compared to information given in material safety data sheets. *Contact Dermatitis* 1997;36:164–5. <https://doi.org/10.1111/j.1600-0536.1997.tb00405.x>