

RESEARCH ARTICLE

Interfacial microscopic examination and chemical analysis of resin-dentin interface of self-adhering flowable resin composite [version 4; referees: 2 approved]

Tamer M. Hamdy 1,2

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https://doi.org/10.12688/f1000research.12306.4)

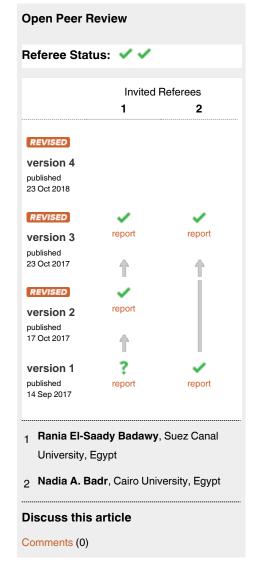
Abstract

Background: The newly introduced self-adhering flowable resin-composites decrease the required time for application by incorporation of an acidic adhesive monomer, thus reducing the number of steps, but its bonding is still uncertain. The aim of this study was to evaluate the interfacial microscopic examination and chemical analysis at the resin-dentin interface of a self-adhering flowable resin composite (Vertise™Flow Self-Adhering Flowable Composite, Kerr Dental, USA) versus a total-etch (Te-Econom Plus) resin composite, using an etching agent (Eco-Etch gel) and bonding agent (Single Bond Universal).

Methods: Sixteen freshly extracted sound human posterior teeth were used. The teeth were randomly divided into two groups: 8 specimens per type of composite. Standard-shaped class V cavities were prepared on the buccal surface. One group was restored by Te-Econom Plus resin composite by total-etch technique using Eco-Etch gel, which was applied to dentine for 15 seconds, followed by rinsing, drying and bonding agent application (Single Bond Universal). The other group restored directly with self-adhering resin composite (Vertise-Flow) without application of etch or bond. Curing was done for 20 seconds using a light emitting diode light curing unit. Evaluation of the resin-dentin interface was done microscopically by examination of marginal gap distance in µm using scanning electron microscope (SEM), and chemical analysis of silver particles was observed using SEM with energy-dispersive X-ray spectrometry after 24 hours of specimen storage in ammoniacal silver nitrate

Results: Regarding marginal gap distance (μm) and silver atomic % mean values, teeth restored with self-adhering resin composite (Vertise-Flow) showed significantly higher mean values than the multi-step etch and rinse resin composite group (5.2 *vs* 0; 12.2 *vs* 8.2, respectively).

Conclusions: Resin-dentin bonding using total-etch resin composite technique



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was more effective than self-adhering flowable resin composite (Vertise-Flow) regarding marginal gap formation and penetration of silver particles. Further studies for bond strength could be performed.

Keywords

Self-adhering, total-etch, bonding system, resin composite, gap distance, resin-dentin interface

Corresponding author: Tamer M. Hamdy (dr_tamer_hamdy@yahoo.com)

Author roles: Hamdy TM: Investigation, Methodology, Project Administration, Writing – Original Draft Preparation, Writing – Review & Editing **Competing interests:** No competing interests were disclosed.

Grant information: The author(s) declared that no grants were involved in supporting this work.

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REVISED Amendments from Version 3

I re-order my affiliation and update the new name of my department due to my institute's request.

See referee reports

Introduction

Adhesive dentistry has seen a paradigm shift from the invasive to be minimally invasive, due to a revolution in bonding systems. There are great demands for simplified restorative materials. A new self-adhering flowable resin composite (VertiseTM Flow Self-Adhering Flowable Composite, Kerr Dental, USA), was recently introduced onto the market. Bonding of flowable composites to tooth structure is achieved by incorporation of an acidic adhesive monomer into the material¹. It is still a big challenge to seal the resin-dentin interface^{2,3}.

The total-etch (etch and rinse) technique is a widely accepted technique to improve bonding of dental resins to tooth structure². The dentin bonding mechanism is based on the micro-mechanical interlocking of the infiltrated resin monomers into porosities created in demineralized inorganic part⁴. Debonding of restorations may arise from gap-formation at the resin-dentin interface and hence recurrent caries, discoloration and tooth pain may follow⁵. Thus sufficient marginal seal should be obtained. Recently, an innovative self-adhesive and flowable resin composite was developed. These materials are claimed to eliminate the need for a separate step of bond-application, finally simplifying the restorative procedure. Therefore, the aim of this study was to evaluate the sealing performance of this new material.

Methods

Preparation of specimens

After attaining written informed consent from each patient to use their extracted teeth in research, sixteen sound human molar teeth were extracted in a private dental clinic (Dr. Tamer Hamdy Dental Clinic), which were randomly divided into two groups (eight specimens per group). Standard-shaped class V cavities (3 mm width, 3 mm length, 2 mm of depth) were prepared in the teeth using a #169L carbide bur (KG Sorensen, Brazil) on the buccal surface. One group's (Group A) cavities were filled with Te-Econom Plus® (Ivoclar Vivadent, Africa) resin composite after etching and bond application. The etching agent, Eco-Etch gel (Ivoclar Vivadent), was applied to dentine for 15 seconds, followed by rinsing and drying. After rinsing, a bonding agent (Single Bond Universal, 3M ESPE, USA) was applied to teeth for 20 seconds, afterwards the teeth were air-dried for 5 seconds and light-cured for 10 seconds. Finally, the Te-Econom Plus resin composite was applied. The other group's (Group B) cavities were filled with self-adhering resin composite (VertiseTMFlow Self-Adhering Flowable Composite, Kerr Dental, USA), which was applied without etch or bond. Curing was done for 20 seconds using a light emitting diode (LED) light curing unit (Satelec, Acteon, France).

Interfacial microscopic examination

All teeth were stored in distilled water for 24 hours at 37°C. Subsequently, the specimens were vertically sectioned with a diamond

saw (Isomet, Buehler Ltd., USA) under water lubrication into approximately 1mm thick slab composed of tooth structure bonded to resin composite. These were examined for marginal gap distance in µm using scanning electron microscope (SEM; Model Quanta 250 FEG; FEI, Thermo Fisher Scientific, USA): accelerating voltage 30 K.V., magnification 14x up to 1000000 and resolution for Gun.1n, to ensure high brightness and resolution at low accelerating voltage.

Chemical analysis of the interface

Specimen slabs were placed in freshly prepared 50 weight % ammoniacal silver nitrate solution for an additional 24 hours at 37°C in the dark. Ammoniacal silver nitrate solution (pH=9.5) was prepared according to *Tay et al.* (2002)⁶. After 24 hours of storage in silver nitrate solution, the silver impregnated specimens were rinsed thoroughly in distilled water and placed in photo-developing solution for 8 hours under a fluorescent light (200 Watt)³.

The specimens were then observed under environmental SEM Model Quanta 250 FEG attached with energy-dispersive X-ray (EDX; Inspect S 50, FEI, Netherlands): accelerating voltage 30 K.V., magnification 4000x and resolution for Gun.1n. The Blackscattered backscattered electron mode was used for elemental analysis of the atomic silver %.

Statistical analysis

Numerical data were explored for normality using Kolmogorov-Smirnov and Shapiro-Wilk tests, followed by Student's t-test to compare between both groups. The significance level was set at $P \leq 0.05$. Statistical analysis was performed with IBM®SPSS® Version 20 for Windows (SPSS Inc., IBM Corporation, USA).

Results

Regarding marginal gap formation, Group A showed a significantly lower mean gap distance values than Group B (p<0.001), as shown in Table 1 and Figures 1 and 2.

The SEM with EDX analysis results revealed significantly lower mean silver atomic % values for Group A compared to Group B (p< 0.001). A selected SEM/EDX analysis is shown in Table 2, and Figures 3 and 4.

Table 1. Marginal gap formation (μm) between groups of teeth restored with different composites. Group A, treated with total-etch technique; Group B, treated with self-adhering resin composite. Mean and standard deviation (SD) values and results of Student's t-test for the comparison between gap distances are shown (n=8/group).

Group A		Group B		P-value
Mean	SD	Mean	SD	r-value
0.0	0.0	5.2	0.3	<0.001*

^{*:} Significant at P ≤ 0.05

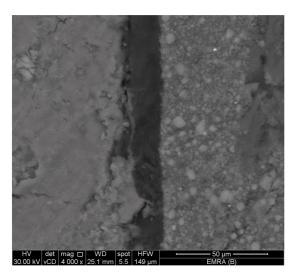


Figure 1. Selected scanning electron microscopy shows absence of gap formation in dentin for teeth treated with total-etch technique. Image representative of 8 teeth.

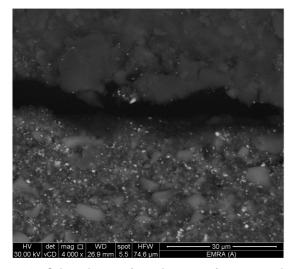


Figure 2. Selected scanning electron microscopy shows presence of gap formation in dentin for teeth treated with a self-adhering resin composite. Image representative of 8 teeth.

Table 2. Chemical analysis of the interface between groups of teeth restored with self-adhering resin composite. Group A, treated with totaletch technique; Group B, treated with self-adhering resin composite. Mean and standard deviation (SD) values and results of Student's t-test for the comparison between silver atomic % values are shown (n=8/group).

Group A		Group B		P-value	
	Mean	SD	Mean	SD	P-value
	8.2	0.4	12.2	0.7	<0.001*

^{*:} Significant at P ≤ 0.05

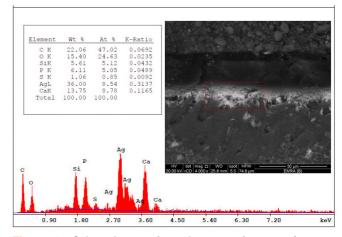


Figure 3. Selected scanning electron microscopy/energydispersive X-ray analysis at the resin-superficial dentin interfaces in teeth treated with total-etch technique. Image representative of 8 teeth.

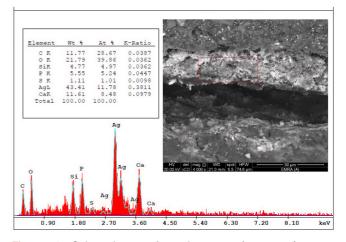


Figure 4. Selected scanning electron microscopy/energydispersive X-ray analysis at the resin-superficial dentin interface in teeth treated with self-adhering resin composite. Image representative of 8 teeth.

Dataset 1. Raw values for silver atomic % in teeth treated with total-etch technique (Group A) and self-adhering resin composite (Group B) (n=8/group/method)

http://dx.doi.org/10.5256/f1000research.12306.d177061

Dataset 2. Scanning electron microscopy (SEM) showing gap formation (raw values included on the images) and SEM/energy-dispersive X-ray analysis (EDX) at the resin-superficial dentin interface in teeth treated with total-etch technique (Group A) and self-adhering resin composite (Group B) (n=8/group/method)

http://dx.doi.org/10.5256/f1000research.12306.d177062

Discussion

A proper marginal seal of restoration is essential to improve the durability of resin composite/bonding systems⁷. Most of the clinical studies assessing the performance of an adhesive system use class V cavities⁸. EDX analysis permits identification of silver particles, thus giving an indication about the chemical analysis of the interface⁹.

Our results revealed better sealing ability of composites treated with multi-step etch and rinse technique, presenting lower marginal gap formation and lower penetration of silver particles compared to Vertise-Flow. The presence of exposed collagen fibers could increase micromechanical interlocking of the bonding agent within the dentin surface¹⁰. The poorer sealing of Vertise-Flow may be due to included adhesive monomer, the glycerol that etches instead of bonds to hydroxyapatite¹¹.

Conclusions

Total-etch resin composite technique was more effective regarding marginal gap formation and penetration of silver particles as

compared to a flowable resin composite (Vertise-Flow). Further studies on bond strength should be undertaken.

Data availability

Dataset 1: Raw values for silver atomic % in teeth treated with total-etch technique (Group A) and self-adhering resin composite (Group B) (n=8/group/method). doi, 10.5256/f1000research.12306. d177061¹²

Dataset 2: Scanning electron microscopy (SEM) showing gap formation (raw values included on the images) and SEM/energy-dispersive X-ray analysis (EDX) at the resin-superficial dentin interface in teeth treated with total-etch technique (Group A) and self-adhering resin composite (Group B) (n=8/group/method). doi, 10.5256/f1000research.12306.d177062¹³

Grant information

The author(s) declared that no grants were involved in supporting this work.

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 - http://www.doi.org/10.5256/f1000research.12306.d177062

Open Peer Review

Current Referee Status:





Version 3

Referee Report 26 October 2017

https://doi.org/10.5256/f1000research.14079.r27183



Rania El-Saady Badawy

Department of Dental Materials, Faculty of Dentistry, Suez Canal University, Ismailia, Egypt

The author has corrected all points required satisfactorily.

Competing Interests: No competing interests were disclosed.

I have read this submission. I believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Referee Report 25 October 2017

https://doi.org/10.5256/f1000research.14079.r27182



Nadia A. Badr

Department of Dental Biomaterials, Faculty of Oral and Dental Medicine, Cairo University, Cairo, Egypt

The article still lacks the name of company and country of production of the investigated materials (Vertise-Flow) in section of methodology.

Competing Interests: No competing interests were disclosed.

I have read this submission. I believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Version 2

Referee Report 18 October 2017

https://doi.org/10.5256/f1000research.13966.r27027



Rania El-Saady Badawy

Department of Dental Materials, Faculty of Dentistry, Suez Canal University, Ismailia, Egypt

The following points need to be corrected, otherwise it is approved



- is based on the micro-mechanical interlocking of the infiltrated resin monomers into porosities created <u>by</u> demineralized inorganic part of dentin 4.
 - Comment: created in demineralized inorganic part....
- Chemical analysis of the interface Specimens slabs were then placed in freshly prepared 50 weight/volume % ammoniacal silver nitrate solution for an additional 24 hours at 37°C in the dark.
 Comment: Specimen slabs were placed......
- lower mean gap distance values than Group B (p<0.001), as shown in Table 1 and Figure 1 and Figure 2.

Comment: and Figures 1 and 2

- The SEM with EDX analysis results revealed that Group A showed significantly lower mean silver atomic % values than Group B (p<0.001), as shown in Table 2 A. Selected SEM/EDX analysis is shown in Figure 3 and Figure 4.
 - <u>Comment:</u> The SEM with EDX analysis results revealed significantly lower mean silver atomic % values for Group A compared to Group B (p< 0.001). A selected SEM/EDX analysis is shown in Figures 3 and 4.
- A proper <u>marginal sealing</u> is essential to improve the durability of resin composite/bonding systems

Comment: A proper marginal seal is essential

This may be attributed to the fact that phosphoric acid included in acid-etch step demineralized the smear layer, exposing the collagen fibers of superficially demineralized dentin. These could increase micromechanical interlocking of the bonding agent within the dentin surface 10 Comment: The presence of exposed collagen fibers could increase....

Competing Interests: No competing interests were disclosed.

I have read this submission. I believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Author Response 18 Oct 2017

Tamer Hamdy, National Research Centre, Egypt, Egypt

Thanks for your concern, corrections done.

Competing Interests: No competing interests were disclosed.

Version 1

Referee Report 11 October 2017

https://doi.org/10.5256/f1000research.13321.r26333





Nadia A. Badr

Department of Dental Biomaterials, Faculty of Oral and Dental Medicine, Cairo University, Cairo, Egypt

The scientific idea of this article to evaluate the adaptability of resin/dentine interface of the most commonly used direct aesthetic restorative material is superb.

The article lacks the name of the company & country of the main investigated material (flowable composite, Vertise-Flow).

Despite the used method is well known for detection of nanoleakage of restorative materials, it is smartly employed for assessment of adaptability of composite at resin/dentine interface.

The suggestions is logical but it is preferable to be separate item not involved in context of conclusion section.

Summary:

- 1. The article is presented clearly & accurately
- 2. The study design is appropriate
- 3. Sufficient details were provided except the name of the company & country of the investigated material (Vertise-Flow)
- 4. The used method for detection of marginal gap is smartly employed
- 5. The data are reproducible
- 6. And the unique conclusion is obvious
- 7. The suggestion is promising for further researches

Is the work clearly and accurately presented and does it cite the current literature? Yes

Is the study design appropriate and is the work technically sound? Yes

Are sufficient details of methods and analysis provided to allow replication by others? Yes

If applicable, is the statistical analysis and its interpretation appropriate? Yes

Are all the source data underlying the results available to ensure full reproducibility? Yes

Are the conclusions drawn adequately supported by the results? Yes

Competing Interests: No competing interests were disclosed.

Referee Expertise: Dental biomaterials



I have read this submission. I believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Author Response 18 Oct 2017

Tamer Hamdy, National Research Centre, Egypt, Egypt

Many thanks for your valuable comments, I do the required corrections, belong to "the suggestions is logical but it is preferable to be separate item not involved in context of conclusion section" I do that according to editorial request according to the journal role

Competing Interests: No competing interests were disclosed.

Referee Report 06 October 2017

https://doi.org/10.5256/f1000research.13321.r26488

Rania El-Saady Badawy

Department of Dental Materials, Faculty of Dentistry, Suez Canal University, Ismailia, Egypt

After reviewing the entire manuscript, several modifications were added to different parts of the manuscript.

For the Introduction part, it shortly covered the idea of investigated resin composites describing the aim of the study.

Materials and Methods described steps and method of slab preparation (missing a few details).

Results were objectively presented.

The results were vaguely discussed.

This manuscript is approved for publication after elaborating the changes required.

Specific Comments:

Bonding is achieved by incorporation of an acidic adhesive monomer into the flowable composites

Comment: Bonding of flowable composites to tooth structure is achieved by incorporation of an acidic adhesive monomer into the material.

• It is still a big challenge to seal the resin-dentin interface, due to the heterogeneous nature of dentin, the wet tubular structure, composition and surface morphology and or improperly designed adhesives^{2,3}

Comment: Too long sentence



• The total-etch (etch and rinse) technique is a widely accepted technique to improve bonding of dental resins².

Comment: improve bonding of dental resins to tooth structure

• ...is based on the micro-mechanical interlocking of the infiltrated resin monomers into porosities created <u>by</u> demineralized inorganic part of dentin⁴.

Comment: created in demineralized organic part....

 Restoration debonding may arise from gap formation at the resin-dentin interface and hence recurrent caries, discoloration and tooth pain⁵

Comment: Debonding of restorations may arise from gap-formation, discoloration and tooth pain may follow

• These materials are claimed to eliminate the need for a separate bonding application step, thus simplifying the restorative procedure

Comment: to eliminate the need for a separte step of bond-application, finally simplifying the restorative procedure

- Thus, the aim of this study was to evaluate the sealing performance of this new material Comment: Therefore, the aim of....
 - Preparation of specimens. After attaining written informed consent from each patient to use their extracted teeth in research

Comment: Why should we get a patients approval to work on extracted teeth?? You should better start with: Sixteen sound human molar teeth......

- ...sixteen sound human posterior molar teeth were extracted in a private Comment: Only say human molar teeth- it is known that molars are posterior
 - (eight specimens per group). Standard-shaped class V cavities (3x3 mm, 2 mm of depth) were prepared

Comment: Dimensions of the cavity are unclear

• One group's (Group A) cavities were filled with Te-Econom Plus® (Ivoclar Vivadent, Africa) resin composite after etching and bonding application.

Comment: and bond application

- A bonding agent (Single Bond Universal, 3M ESPE, USA) was applied to the teeth for 20 Seconds Comment: After rinsing, a bonding agent was applied to the teeth for 20 sec
- ...teeth for 20 seconds, then air-dried for 5 seconds, then light-cured for 10 seconds.

 Comment: was applied to teeth for 20 seconds, afterwards the teeth were air-dried for 5 sec and light-cured for 10 sec
 - ...under water lubrication into approximately 1mm thick slabs.

Comment: Slab of what??? Please explain specific composition the slab. Please mention that it is a slab composed of tooth structure bonded to resin composite of any type!!!



• Chemical analysis of the interface. Specimens slab were then placed in freshly prepared 50 weight/volume % ammoniacal silver nitrate solution for an additional 24 hours at 37°C in the dark.

Comment: Specimen slabs were placed......weight or volume percent?????

• Ammoniacal silver nitrate solution (pH=9.5) was prepared according to the protocol of Tay et al. (2002)⁶.

Comment: prepared according to Tay et al

• After 24 hours of storage in ammoniacal silver nitrate, the silver impregnated specimens were then rinsed thoroughly in distilled water and placed in photo-developing

Comment: storage in silver nitrate solution, the silver impregnated specimens were rinsed. Thoroughly

• The specimens were then observed under environmental SEM Model Quanta 250 FEG attached with energy-dispersive X-ray (EDX; Inspect S 50, FEI, Netherlands): accelerating voltage 30 K.V., magnification 4000x and resolution for Gun.1n.

Comment: he treated specimens were observed.

Backscattered electron mode was used for elemental analysis of the atomic silver %.

Comment: The Blackscattered

...followed by Student's t-test to compare between the two groups.

Comment: to compare between both groups

• lower mean gap distance values than Group B (p<0.001), as shown in Table 1 and Figure 1 and Figure 2.

Comment: and Figures 1 and 2

• The SEM with EDX analysis results revealed that Group A showed significantly lower mean silver atomic % values than Group B (p<0.001), as shown in Table 2 A. Selected SEM/EDX analysis is shown in Figure 3 and Figure 4.

Comment: revealed significantly lower mean silver atomic % values for Group A compared to Group B (p< 0.001) A selected..... in Figures 3 and 4.

• Table 1. Marginal gap formation (μm) between groups of teeth treated with different composites

Comment: groups of teeth restored with different...

• Figure 1. Selected scanning electron microscopy shows absence of gap formation in dentin in teeth treated with total etch technique.

Comment: in dentin for teeth treated with....

• Figure 2. Selected scanning electron microscopy shows presence of gap formation in dentin in teeth treated with self-adhering resin composite.

Comment: in dentin for teeth...... with a self-adhering

 Table 2. Chemical analysis of the interface between groups of teeth treated with different composites.

Group A, treated with total-etch technique; Group B, treated with self-adhering resin composite.

Comment: restored with self-adhering resin composite



 Dataset 1. Raw values for silver atomic % in teeth treated with total-etch technique (Group A) and self-adhering resin composite (Group B) (n=8/group/method)

Comment: Group (A), ...

• Dataset 2. Scanning electron microscopy (SEM) showing gap formation (raw values included on the images) and SEM/energy dispersive X-ray analysis (EDX) at the resin-superficial dentin interface in teeth treated with total-etch technique (Group A) and self-adhering resin composite (Group B) (n=8/group/method)

Comment: Group (A), ...

A proper <u>marginal sealing</u> is essential to improve the durability of resin composite/bonding systems

Comment: sealing of what???

• From the present results, it was obvious that the multi-step etch and rinse technique provides better sealing regarding

Comment: Our results revealed better sealing ability of composites treated with multi-step etch and rinse technique, presenting lower marginal gap formation and lower penetration of silver particles compared to Vertise-Flow.

This may be attributed to the fact that phosphoric acid included in acid-etch step demineralized the smear layer, exposing the collagen fibers of superficially demineralized dentin. These could increase micromechanical interlocking of the bonding agent within the dentin surface 10

Comment: Presence of exposed collagen fibers could increase....

The poorer sealing of Vertise-Flow may be due to the included adhesive monomer, which is called glycerol phosphate dimethacrylate "GPDM", that etches instead of bonds to hydroxyapatite 11.

Comment: included adhesive monomer, the glycerol.......

• Further studies for bond strength could be performed. It is important to emphasize that this study ignored the effect of oral condition,

Comment: Further studies on bond strength should be undertaken.

Is the work clearly and accurately presented and does it cite the current literature? Partly

Is the study design appropriate and is the work technically sound?

Are sufficient details of methods and analysis provided to allow replication by others?

If applicable, is the statistical analysis and its interpretation appropriate? Yes

Are all the source data underlying the results available to ensure full reproducibility? Yes



Are the conclusions drawn adequately supported by the results? Yes

Competing Interests: No competing interests were disclosed.

I have read this submission. I believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard, however I have significant reservations, as outlined above.

Author Response 06 Oct 2017

Tamer Hamdy, National Research Centre, Egypt, Egypt

Thanks for your valuable revision. Corrections done

Belong to your comment

Comment: Why should we get a patients approval to work on extracted teeth?? You should better start with: Sixteen sound human molar teeth.....

It was done according to journal editor revision due to the rule of the journal.

Competing Interests: No competing interests were disclosed.

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