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Reactive changes in dental tissues in response to curing light exposure: An experimental study

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ABSTRACT

BACKGROUND: The use of curing lamps has considerably improved the quality of dental treatment. Curing light exposure protocols are primarily based on the effectiveness (completeness) of cure. The risk of heat impact on the dental pulp and surrounding tissues is a downside of curing light radiation. There is a need for experimental confirmation of differentiated selection of curing light exposure protocols (intensity and time of exposure) in terms of minimum impact on tissues. **AIM:** To assess the effect of curing light on dental tissues.

MATERIALS AND METHODS: An experimental simulation of the use of curing light in dental practice during dental restoration was performed. The experiment was performed in 50 rats randomized into four groups: Group 1 (control, n=5) and Groups 2–4 (treatment, n=15 each). The control group was not exposed to curing light. In the treatment groups, mandibular incisors of the experimental animals were exposed to curing light in three modes. The animals were sacrificed by decapitation after 7 days in Group 1 and after 1, 3, and 7 days in Groups 2–4. A total of 100 teeth were extracted for pathomorphological examination. A total of 320 histological sections (enamel, dentin, and pulp slides) were prepared, stained, and examined. The descriptive method was used to assess the findings.

RESULTS: Changes in the pulp showed pathomorphological signs of acute inflammation, most prominent on Day 3, which was considered a defense response of the pulp to irritation. The microvasculature showed the most significant changes, with increased, inhomogeneous blood filling, plasmorrhagia of capillary walls, and stasis in capillary lumen. There were no morphological changes in hard dental tissues.

The initial response of the dental pulp to curing light exposure in experimental animals was uniform and unaffected by the technical specifications of curing lamps. On Day 7, there was no morphological response in the pulp when exposed to diode light at 1,000 and 1,400 mW/cm². At this point, the microscopic appearance of the pulp exposed to diode curing light at 3,200 mW/cm² was generally comparable to that at baseline, with residual perivascular cellular infiltration of the stroma and capillary congestion.

CONCLUSION: The experimental findings indicate a risk of negative photochemical reactions in the pulp following curing light exposure during treatment of patients with hard dental tissue pathologies. The data on the effect of curing light on the dental pulp can be used in real-world dental practice when selecting a composite curing algorithm during dental restoration to reduce the risk of unexpected reactions to curing light.

Keywords: diode curing lamp; tooth microstructure; odontoblasts; dental pulp; dental pulp inflammation; semiquantitative morphometric analysis.

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Реактивная динамика состояния тканей зуба в ответ на действие фотополимеризаторов (экспериментальное исследование)

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АННОТАЦИЯ

Обоснование. Использование фотополимеризационных устройств стало новой ступенью повышения качества стоматологической помощи. Протоколы применения фотополимеризаторов разработаны в первую очередь с учётом эффективности (полноты) полимеризации. Недостатком излучения фотополимеризаторов является возможность теплового воздействия на пульпу зуба и окружающие ткани. Существует потребность в экспериментальном обосновании дифференцированного выбора протокола фотополимеризации (интенсивности, времени облучения) с позиции минимального воздействия на ткани. Цель исследования — изучение влияния светового излучения стоматологических фотополимеризаторов на ткани зуба. Материалы и методы. Проведено экспериментальное моделирование применения света фотополимеризационных устройств в стоматологической практике для облучения зубов при их реставрации.

Эксперимент выполнен на 50 крысах, разделённых случайным образом на 4 группы: первая, контрольная (*n*=5), вторая—четвёртая, основные (*n*=15 в каждой). Животных контрольной группы воздействию светового излучения стоматологических фотополимеризаторов не подвергали. На резцы нижней челюсти экспериментальных животных основных групп оказывали воздействие светом стоматологического фотополимеризационного устройства в трёх режимах. Спустя 1, 3 и 7 сут животных второй—четвёртой групп и спустя 7 сут — животных контрольной группы выводили из эксперимента путём декапитации. Для патоморфологических исследований выделили 100 зубов. Подготовлены, окрашены, изучены 320 гистологических срезов (микропрепараты эмали, дентина, пульпы зуба). Для оценки полученных результатов использован описательный метод.

Результаты. Изменения в пульпе имели патоморфологические признаки острого воспаления, наиболее выраженные на 3-и сутки, что расценивается как защитная реакция пульпы на раздражение. Наиболее выраженные изменения отмечены со стороны микроциркуляторного русла в виде повышенного и неравномерного кровенаполнения сосудов, плазматического пропитывания стенок капилляров, стазов в их просвете. Морфологических изменений в твёрдых тканях зубов не выявлено.

Первичная реакция пульпы зубов экспериментальных животных на воздействие светового излучения фотополимеризаторов однотипна и не зависит от их технических характеристик. На 7-е сутки наблюдали полное купирование ответной морфологической реакции в пульпе при диодном облучении мощностью 1000 и 1400 мВт/см². В этот срок микроскопическая картина пульпы животных после воздействия диодным фотополимеризатором мощностью излучения 3200 мВт/см² приближалась к исходной при остаточном сохранении клеточной периваскулярной инфильтрации стромы и полнокровия капилляров.

Заключение. Полученные экспериментальные данные свидетельствуют о возможном развитии негативных фотохимических реакций в пульпе зуба после воздействия света стоматологических фотополимеризаторов в процессе лечения больных с патологией твёрдых тканей зубов. Данные о влиянии излучения стоматологических фотополимеризаторов на пульпу зуба имеют прикладное значение при выборе алгоритма фотополимеризации композита в процессе реставрации зубов для снижения риска непредвиденных реакций от действия излучения.

Ключевые слова: диодный фотополимеризатор; микроструктура зуба; одонтобласты; пульпа зуба; воспаление пульпы зуба; полуколичественный морфометрический анализ.

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BACKGROUND

Modern dentistry routinely employs light-curable restorative materials. The introduction of light-curing devices has marked a significant advancement in the quality of dental care. As with all medical devices, their safety must be ensured [1]. Light-curing protocols are primarily developed with regard to polymerization efficacy (completeness of cure). However, a notable drawback of curing light exposure is the potential for thermal injury to the dental pulp and surrounding tissues. There is a clear need for experimental validation of differentiated light-curing protocols, specifically intensity and exposure time, to minimize tissue impact [2]. No similar studies were identified in the available databases.

AIM: To assess the effect of curing light on dental tissues.

METHODS

Study Design

An experimental simulation of curing light application in dental restorative procedures was conducted. The study included 50 rats randomized into four groups: Group 1 (control, n=5) and Groups 2–4 (treatment, n=15 each). Animals in the control group were not exposed to curing light. In treatment groups, the mandibular incisors were irradiated twice with a dental curing device emitting light at 395-480 nm. Three light intensity modes were applied: Group 2 was exposed to 1,000 mW/cm², Group 3 to 1,400 mW/cm², and Group 4 to 3,200 mW/cm². The animals were sacrificed by decapitation after 7 days in Group 1 and after 1, 3, and 7 days in Groups 2, 3, and 4, respectively. A total of 100 teeth (two mandibular incisors per animal) were extracted for pathomorphological examination. From these specimens, 320 histological sections (including enamel, dentin, and pulp) were prepared, stained, and analyzed.

Study Setting

The experiment was carried out at Smolensk State Medical University. Animals were housed in a vivarium under standard sanitary conditions in labeled enclosures¹. For procedural sedation and decapitation, animals received intraperitoneal premedication with 2% xylazine hydrochloride solution (Xyla, Interchemie Werken "De Adelaar" B.V., Netherlands) at a dose of 0.15 mL/kg, followed by anesthesia with Zoletil (Virbac Santé Animale, France) at a dose of 0.01 mg/kg [3].

Study Duration

Dental tissue responses to curing light were assessed dynamically 1, 3, and 7 days post-exposure. The experiment was conducted in 2023.

Measurement Protocol

The extracted teeth were decalcified for 18 days in trichloroacetic acid. Following initial fixation, the specimens were trimmed, re-fixed above the liquid level in trichloroacetic acid for 24 hours, and embedded in paraffin. Histological sections 5-8 µm thick were prepared using a microtome. Deparaffinization was performed by treating the sections with three changes of xylene for 5 minutes each. Dehydration was achieved by sequential immersion in four ethanol concentrations (absolute, 96%, 70%, and 56%) for 3 minutes each. The material was fixed in 15% aqueous neutral buffered formalin and paraffin-embedded. Histological staining included hematoxylin and eosin, Van Gieson, Alcian blue. and periodic acid-Schiff (PAS) reaction. The vascular bed and its components were examined using the Gab-Dyban technique. Standardized comparative microscopic analysis of dental tissues (enamel, dentin, and pulp) was performed using the Axio Imager A2 imaging system (Carl Zeiss, Germany) at ×100 and ×200 magnification. Digital images were acquired using the Axiocam 506 color camera for light microscopy (Carl Zeiss, Germany).

Main Study Outcome

The primary objective was to test the hypothesis that curing light exposure, under manufacturer-recommended settings for composite restorative materials, induces reactive changes in dental tissues. The study also sought to characterize the underlying pathomorphological features of these responses.

Additional Study Outcomes

An additional outcome was to assess the reversibility of the observed tissue changes and their resolution.

Subgroup Analysis

The study included 8-month-old male rats. Group 1 served as the control, while Groups 2 through 4 received curing light exposure. Differences between the control and treatment groups were assessed by analyzing intragroup changes following exposure to diode curing light.

Outcomes Registration

Outcomes were assessed using descriptive statistics and semiquantitative evaluation, with ranked scoring of histopathologic features.

Given the inherent subjectivity of microscopic evaluation, determined by the study subject and observer, both intraobserver and interobserver validation methods were employed to minimize bias.

Ethics Approval

The study design was justified by weighing the potential harm to animals against the prospective benefit to human health. The use of animals was limited to the minimum necessary to achieve scientific validity. All procedures

¹ GOST 33215-2014. Guidelines for accommodation and care of animals. Environment, housing, and management. Available at: https://docs.cntd.ru/document/1200127789

adhered to current legal and ethical standards². The study was approved by the Ethics Committee of Smolensk State Medical University (Minutes No. 2 of September 23, 2022).

Statistical Analysis

Descriptive statistics were used to assess the findings.

RESULTS

Primary Results

Histologic examination of dental tissues in the control group (Group 1) revealed preserved structural components: enamel, dentin, and pulp. Enamel, physiologically thin in this species, covered the crown portion of the tooth. Dentin, a connective tissue, consisted of fibroblasts and odontoblasts within an extracellular matrix composed of procollagen, collagen, and reticular fibers. At the boundary with predentin, odontoblasts were arranged in rows of 6-8 cells within the coronal pulp. These cells varied in shape from prismatic or pear-shaped to cuboidal. Beneath the odontoblast layer in the coronal portion of the tooth, the subodontoblastic layer consists of the cell-free (zone of Weil) and cell-rich zones containing fibroblasts, lymphocytes, and poorly differentiated cells capable of differentiating into odontoblasts. The pulp exhibited rich innervation and vascularization. Concentric radial dentin layers followed the odontoblast layer, with the dentin layer approximately 1.5 times thicker than the pulp. Overall, the dental microstructure resembled that of a human tooth [4], with two distinguishing features: thinner enamel and more pronounced pulp vascularity.

In Groups 2–4, teeth were exposed to curing light (395–480 nm). In Group 2 (1,000 mW/cm²), no signs of pulp inflammation were observed on Day 1. By Day 3, individual lymphocytes and polymorphonuclear leukocytes appeared in

the loose connective tissue stroma of the pulp. Microvascular blood filling was heterogeneous; some vessels exhibited dilated lumens.

In Group 3 (1,400 mW/cm², 3-second exposure), histologic architecture was preserved on Day 1 (Fig. 1), and findings were largely within normal limits. However, heterogeneous blood filling of the pulp microvasculature was noted. The odontoblast layer appeared thinned and sparsely populated. Capillary walls exhibited plasmorrhagia, with occasional stasis in the lumen; endothelial cells remained intact. On Day 3, mild perivascular lymphocytic infiltration was observed in the pulp stroma (Fig. 2).

In Group 4 (3,200 mW/cm²), Day 1 findings showed preserved overall dental structure with scattered cellular infiltration of the pulp and good vascular filling (Fig. 3). Lymphocytes were noted in the loose connective tissue stroma. On Day 3, dental hard tissues remained structurally intact. Pulp microvasculature exhibited uneven blood filling, and sparse lymphocytes were present in the stroma. PAS staining revealed weak glycogen positivity in the pulp, with higher glycogen content in circumpulpal dentin compared to mantle dentin (Fig. 4). Pulp vessels remained engorged, with areas of vascular stasis.

Histochemical staining showed preserved structure of hard dental tissues in all groups at all time points. The dentin exhibited a radial structural pattern (see Fig. 1).

Secondary Results

The findings suggest that reactive changes in the dental pulp induced by curing light exposure are reversible.

On Day 7, histologic features of the pulp in Groups 2 and 3 approximated normal. The odontoblast layer maintained its histoarchitectonic integrity, and microvasculature showed vascular engorgement without evidence of inflammatory infiltrate.

In Group 4, Day 7 histology remained consistent with that observed on Days 1 and 3. The edematous stroma contained scattered lymphocytes, plasma cells, and



Fig. 1. Transverse histological section of a tooth (third experimental group, day 1). Hematoxylin and eosin staining; ×100.

² GOST 33044-2014. International standart. Principles of good laboratory practice. Available at: https://docs.cntd.ru/document/1200115791



Fig. 2. Perivascular infiltration in the dental pulp (third experimental group, day 3). Hematoxylin and eosin staining; ×200.



Fig. 3. Transverse histological section of a tooth (fourth experimental group, day 3). Intensive presence of glycogen in the tooth tissues around the pulp. Staining using the PAS reaction; ×100.



Fig. 4. Transverse histological section of a tooth (fourth experimental group, day 1). The degree of blood filling of the dental pulp is shown. Staining according to Gabu-Dyban; ×200.

polymorphonuclear leukocytes. PAS staining continued to demonstrate increased glycogen content in the dentin adjacent to the pulp. The pulp vasculature remained markedly engorged.

Adverse Events

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No adverse events were observed during the study.

DISCUSSION

The experimental findings indicate a risk of negative photochemical reactions in the pulp following curing light exposure during treatment of patients with hard dental tissue pathologies. Histopathologic changes in the pulp exhibited features of acute inflammation, most pronounced on Day 3, and were interpreted as a protective response to irritation [5,6]. The most significant alterations involved the microvasculature, including increased and heterogeneous blood engorgement, plasmorrhagia of capillary walls, and stasis within their lumens. Although this condition typically does not require specific treatment, it may cause transient postoperative discomfort in some patients following light curing.

There were no morphological changes in hard dental tissues.

The initial pulp response to curing light exposure in experimental animals was uniform and appeared independent of the technical specifications of the curing devices. By Day 7, reactive changes in the pulp resolved completely in groups

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exposed to diode light at 1,000 and 1,400 mW/cm². At this point, the pulp morphology in animals exposed to 3,200 mW/ $\rm cm^2$ light approximated baseline findings, with residual perivascular cellular infiltration and persistent capillary congestion.

CONCLUSION

These data regarding the effects of curing light on the dental pulp have practical implications for selecting curing protocols during composite restorations, aiming to minimize the risk of unexpected tissue reactions.

ADDITIONAL INFORMATION

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