Original Research Article

University J Dent Scie 2022; Vol. 8, Issue 3

A unique method to study different histological structures using Hematoxylin and Eosin staining in partially decalcified ground sections of teeth : A research article

Abstract:

Introduction: Examining the tooth is considered as foremost segment in research in the field of oral biology and oral pathology. For studying various structures like enamel, dentin and cementum, ground section of teeth is used, while decalcified sections of tooth are used for studying the soft tissues.

Objective: The present study was aimed at exploring the normal structures of hard tissue parts of the teeth where partially decalcified ground sections were stained them with routine hematoxylin and eosin stain.

Methodology: For the present study, a total of 30 extracted teeth were collected. The freshly prepared ground sections were partly decalcified by putting ground sections of teeth in 10% nitric acid for a time period of 20- 25 seconds. After this, the ground sections of teeth are washed in tap water. The partly decalcified ground sections of teeth were stained with routine hematoxylin and eosin staining procedure and were mounted on clean glass slides using DPX mounting media. The sections were then analyzed for various histological structures under the microscope.

Results: When hematoxylin and eosin stained partially decalcified ground sections of teeth were viewed under light microscope, variety of histological structures appeared as stained, while some structures did not take stain and remain unstained.

Conclusion: After preparing and staining the ground section of teeth by our method, we hope that it would be of great help for undergraduate students and to some extent the research scholars to understand the various structures of the teeth in a better and easy way.

Key words: Decalcification, ground section, enamel, dentin, cementum

Introduction:

Human teeth consist of various hard and soft tissues. Enamel is the mineralised tissue which have most calcium content in human body in the range of 96-98%,[1,2] which is followed by dentin with approx 70% of mineralized content.[3,4] However in case of cementum, mineralization ratio is about 50 %.[5]

Examining the tooth is considered as foremost segment in research in the field of oral biology and oral pathology. For studying various structures like enamel, dentin and cementum, ground section of teeth is used, while demineralized or decalcified sections of tooth are used for studying the periodontal ligament, cells, pulp etc.

The process of decalcification was illustrated many years ago when researchers noticed bone which is immersed in acid underneath the microscope. After that, different decalcifying agents have been reported which was used for decalcifying bone and teeth. Such commonly used decalcifying agents includes nitric acid and formic acid etc.[6]

Access this article online

Website: www.ujds.in

Quick Response Code

DOI: https://doi.org/10.21276//ujds.2022.8.3.2

[1]HIMANSHU SINGH, [2]VEDANT PATEL, [3]KUNDENDU ARYA BISHEN, [4]URVASHI TOMAR, [5]MAGESH KUMAR

[1,4]Department of Oral Pathology & Microbiology, Index Institute of Dental Sciences, Indore

[2]Department of Prosthodontics and Crown and Bridge, Index Institute of Dental Sciences, Indore

[3]Department of Oral Pathology & Microbiology, Government College of Dentistry, Indore

[5]Department of Periodontology, Index Institute of Dental Sciences, Indore

Address for Correspondence: Dr. Himanshu Singh
MDS, Senior Lecturer
Department of Oral Pathology and Microbiology
Index Institute of Dental Sciences, Indore
Email : himanshustar3g@gmail.com

Received : 28 Dec., 2021, **Published :** 30 Sep., 2022

How to cite this article: Himanshu Singh, & Vedant Patel. (2022). A unique method to study different histological structures using Hematoxylin and Eosin staining in partially decalcified ground sections of teeth : A research article.UNIVERSITY JOURNAL OF DENTAL SCIENCES, 8(3). 6-9

The present study was aimed at exploring the normal structures of hard tissue parts of the teeth by adopting a novel technique where we used partially decalcified ground sections and stained them with routine hematoxylin and eosin stain.

Aims and objective:

The main aim of the present study was to introduce a novel technique for identification of various histologic structures in partially decalcified ground sections of teeth which would further aid in academic/teaching as well as for research purposes. Other aim was to check whether these histologic structures can be stained by routine hematoxylin and eosin stain or not.

Materials and methods:

For the present study, a total of 30 extracted teeth were collected. All the teeth which were extracted were caries free and without any morphological anomalies. All the teeth were put in hydrogen peroxide solution for a period of 24 hours which was followed by thorough rinsing in running tap water. After that, the ground section of all the tooth were prepared. For preparing ground sections, initially tooth were grossly reduced mesio-distally on a lathe machine till the tooth reached to a thickness of 1-2 mm. After achieving suitable thickness, the sections were then grounded on arkansas stone until a suitable thickness of 25-35 microns was achieved.

The freshly prepared ground sections was then partly decalcified by putting ground sections of teeth in 10% nitric acid for a time period of 20- 25 seconds. After this, the ground sections of teeth are washed in tap water. The partly decalcified ground sections of teeth were stained with routine hematoxylin and eosin staining procedure and were mounted on clean glass slides using DPX mounting media. Ground sections were then analyzed for various histological structures under the microscope.

Results:

When hematoxyl in and eosin stained partially decalcified ground sections of teeth were viewed under light microscope, variety of histological structures appeared as stained, while some structures did not take stain and remain unstained.

The histologic structures clearly seen were incremental lines of salter along with dentinal tubules (Figure 1), interglobular dentin, primary dentinal tubules, enamel tufts, enamel lamellae and dentino-enamel junction (Figure 2), sharpey's fibers (Figure 3), enamel tufts, enamel lamellae and dentino-enamel junction (Figure 4), hunter schreger bands (Figure 5), striae of retzius and enamel lamellae (Figure 6), enamel rods along with their directions (Figure 7) and keyhole pattern of enamel rods (Figure 8).

Fig 1 : Incremental lines of Salter

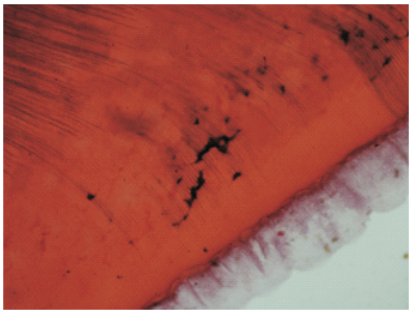
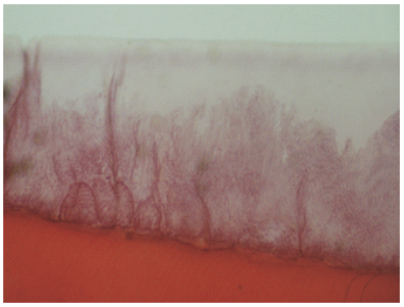


Fig 2 : Interglobular dentin, enamel tufts, enamel lamellaeFig



3 : Sharpey's Fibers

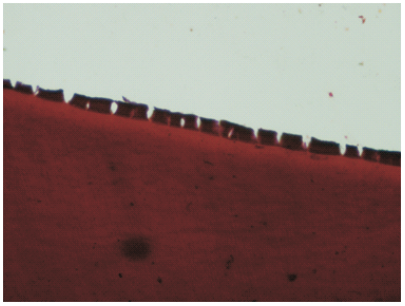


Fig 4 : Enamel Lamellae and tufts

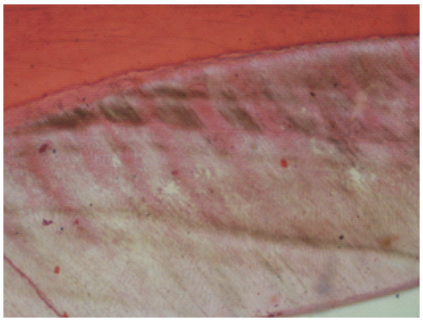


Fig 5 : Hunter-Schreger bands

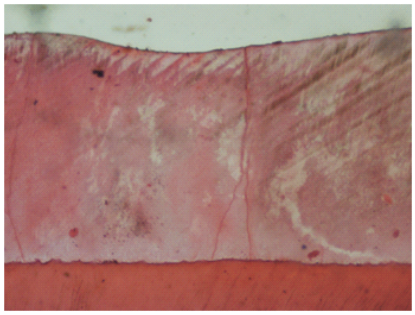


Fig 6 : Striae of Retzius

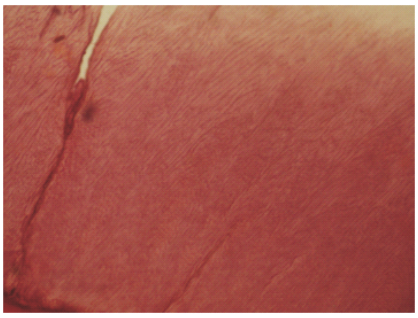


Fig 7 : Enamel rods

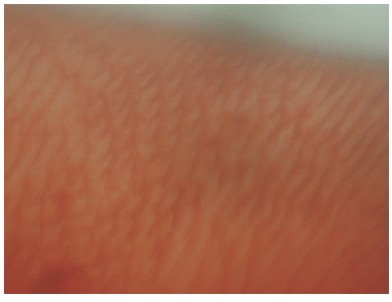


Fig 8 : Lock and Key Structure

Discussion:

The main aim of this study was to introduce a novel and unique technique which helps in demonstration of various histological structures in partially decalcified ground section of teeth. In the present study, we used nitric acid as a decalcifying agent. For the purpose of staining, hematoxylin and eosin was used which is more commonly used stain for routine staining.

Investigation of teeth is considered as an essential and fundamental part of oral pathology as well as oral biology research. The well prepared ground sections of tooth can be used to examine the various histologic structures of enamel, dentin as well as cementum[6].

Staining of tooth or tissue is important as it helps in studying the distinctive components of tooth and tissues. It is observed that tissues and teeth possess different affinity for different stains. The most commonly used histopathological stain is hematoxylin and eosin stain. The hematoxylin and eosin staining procedures contains number of steps, but still this staining is considered as gold standard in routine staining procedure[7,8].

The present study demonstrated the presence of various structures like enamel tuft and lamellae, interglobular dentin, incremental lines of salter etc. Some structures remained unstained.

Conclusion:

In the present study, we have tried to introduce a new technique to study various histological features which is seen in well prepared partially decalcified ground sections of teeth. After preparing and staining the ground section of teeth by our method, we hope that it would be of great help for undergraduate students and to some extent the research scholars to understand the various structures of the teeth in a better and easy way.

References:

1. Eimar H, Ghadimi E, Marelli B et al. Regulation of enamel hardness by its crystallographic dimensions. Acta biomater. 2012;8:3400–3410.
2. Feuerstein O, Mayer I, Deutsch D. Physico-chemical changes of human enamel irradiated with arf excimer laser. Lasers surg med. 2005;37:245–251.
3. Piesco NP. Histology of dentin. In: Avery JK (ed.). Oral development and histology. 3rd ed. New york, thieme 2002:172–189.
4. Nanci A. Dentin-pulp complex. In: Nanci a (ed.). Ten cate's oral histology development, structure, and

function. 7th ed. Louis, mosby-elsevier 2008:191–238.

5. Nanci A, Bosshardt DD. Structure of periodontal tissues in health and disease. Periodontol 2000. 2006;40:11–28
6. Bankroft JD. Theory and practice of histological techniques. 5th ed, philadelphia: Churchill living stone; 2003. P.274-9
7. Alturkistani HA, Tashkandi FM, Mohammedsaleh ZM. Histological stains: A literature review and case study. Glob j health sci. 2016;8(3):72.
8. Insira sarbeen J, Jayaraj G. Light microscopic study of cementum under different histological stains. J pharm sci & res. 2015;7(9):720–23