IJBPAS, July, 2021, 10(7): 2392-2396

ISSN: 2277–4998

International Journal of Biology, Pharmacy and Allied Sciences (IJBPAS)

'A Bridge Between Laboratory and Reader'

www.ijbpas.com

SENNA AURICULATA AS AN ALTERNATE TO EOSIN IN ROUTINE STAINING

MITHRA S[1*], RAMANI P[2], SHERLIN HJ[3], GHEENA S[4], RAMASUBRAMANIAM A[5], JAYARAJ G[6], DON KR[7] AND SANTHANAM A[8]

1: MDS, Post Graduate student, Department of Oral and Maxillofacial Pathology, Saveetha Dental College & Hospitals, SIMATS, Chennai

2: Professor and Head, Department of Oral and Maxillofacial Pathology, Saveetha Dental College & Hospitals, SIMATS, Chennai

3: Professor, Department of Oral and Maxillofacial Pathology, Saveetha Dental College & Hospitals, SIMATS, Chennai

4: Reader, Department of Oral and Maxillofacial Pathology, Saveetha Dental College & Hospitals, SIMATS, Chennai

5: Reader, Department of Oral and Maxillofacial Pathology, Saveetha Dental College & Hospitals, SIMATS, Chennai

6: Reader, Department of Oral and Maxillofacial Pathology, Saveetha Dental College & Hospitals, SIMATS, Chennai

7: Senior Lecturer, Department of Oral and Maxillofacial Pathology, Saveetha Dental College & Hospitals, SIMATS, Chennai

8: Senior Lecturer, Department of Oral and Maxillofacial Pathology, Saveetha Dental College & Hospitals, SIMATS, Chennai

***Corresponding Author: Dr. S Mithra: E Mail: mithraswornappan@gmail.com**

Received 20th Aug. 2020; Revised 19th Sept. 2020; Accepted 10th Oct. 2020; Available online 1st July 2021

https://doi.org/10.31032/IJBPAS/2021/10.7.5562

ABSTRACT

Application of natural dyes for staining numerous biological tissues from an alternate source willdecrease the expense for the artificial dyes and put back their effects on human and environment. Therefore, the main target of this study was to analyse the extraction of natural dye from *senna auriculata* in both aqueous and alcoholic form and was proceeded with tissue processed slides. The Results showed that the tissues took up the stain overall. Hence

it can be concluded that standardising and modifying the stain can give positive results inorder to prove it is equivalent to or better than eosin.

Keywords: *Senna auriculata*, Eosin, Routine Staining

INTRODUCTION

Stains in histopathology are substances or biological dyes that color tissues so as to aid optical differentiation of tissue components.In histology, there are two types of dyes,

synthetic dyes made through chemical reactions and natural dyes obtained from natural sources **[1-2]**. The synthetic dyes are very efficient, but the dyes are a hazard to human and animal health **[3]**. Thus, the finding of recent natural dyes for histologic staining that are eco-friendly and biodegradable materials has been studied.Plants yield several colours for staining a variety of materials; some trees and plenty of herbs including fruit produces dyestuffs **[4]**.

Staining is utilized to highlight important features of the tissue as well as to enhance the tissue contrast. Hematoxylin is a basic dye that is commonly used in staining process and stains the nuclei giving it a unremarkable bluish color while eosin the other stain employed in staining the cell nucleus pink. Eosin was a name derived from Eos, the Ancient Greek word for 'dawn' and the name of the Ancient Greek goddess of the dawn. However, there are other several staining technicques used for particular cells and components **(Black, 2012)**. Staining is a commonly used medical process in the medical diagnosis of tumors in which a dye is applied on the posterior and anterior border of the sample tissues to differentiatea diseased or tumor cells or any other pathological cells **(Musumeci, 2014)**. In biological studies staining is used to mark cells and to flag nucleic acids, proteins or the gel electrophoresis to aid in the microscopic examination **(Jackson & Blythe, 2013)**. In some cases, Numerous multiple staining methods are used such as differential staining, double staining or the multiple staining **(Iyiola & Avwioro, 2011)**.

Cassia auriculata Linn goes by the nameTanners Senna, that is also known as Avaram tree.

Other Names used widely are as follows, Tanner's Cassia,Tanner's Senna, Mature Tea Tree (English) Avartaki, Pitapuspa, Pitkalika, Manojyna, Pitkala, Charmaranga (Sanskrit), Tarwar, Awal, Tarval (Hindi), Tangedu, Merakatangeedu (Telugu), Arsual, Taravada, Tarwad (Marathi) .

Itis distributed throughout hot deciduous forests of India. Wildin dry regions of Madhya Pradesh, Tamil Nadu Rajasthan

and other parts of India.The flowersare also used in treating urinary discharges, nocturnal emissions, diabetes and throat irritation. The flower is bright yellow coloured used for various medicinal purposes also. Henceforth the study was designed to investigate the staining property of *senna auriculata* by extracting the natural stain.

MATERIALS & METHODS:

Senna auriculata flowers were collected and shade dried for a week. Once dried it was powdered to a fine mixture. Stain extraction was carried out in two ways one is aqueous and the other is alcoholic.

Aqueous Extract of Senna:

200gms of the dried senna powder was taken and it was soaked in 100ml of water overnight, then the extract was purified using Whatman filter paper and the extract was collected in an Eppendorf tube.

Alcoholic Extract of Senna:

Theflowers were shade dried and powdered. 500g of powdered material was extracted with 1500 ml of methanol for 72h. The solvent was evaporated under reduced pressure, After 72hrs the extract was purified by 2 step filtration process. The extract was filtered using cheese cloth and then filtered with Whatman No.1 filter paper and finally a thick yellow coloured stain was obtained.

Alteration of pH:

Both aqueous & alcoholic extracts basically had a PH of 7 and as it is substituted for eosin the Ph was altered to 4 to make it acidic.

Once the PH was altered, Paraffin embedded processed tissue slides were collected from the Department of Oral Maxillofacial Pathology laboratory for the research and scoring was given based on epithelial differentiation, connective tissue differentiation, nuclearstaining, cytoplasmic details, staining intensity and contrast.

The Slides were treated with plain Senna stain to notice if the tissue takes up any stain, Later one more slide was taken and the staining process was done with hematoxylin, then washed with water followed by ammonia and In place of eosin, the slide was substituted with yellow senna stain.

Result:

The aqueous extract had no much staining property and moreover the aqueous extract gets washed away when processing.

The alcoholic extract stained the tissue overall when treated alone without any other dyes **(Figure 1, 2)**. When the slides were treated with hematoxylin, ammonia and the yellow stain of senna the tissues took up the stain overall but not all features can be differentiated **(Figure 3)**.

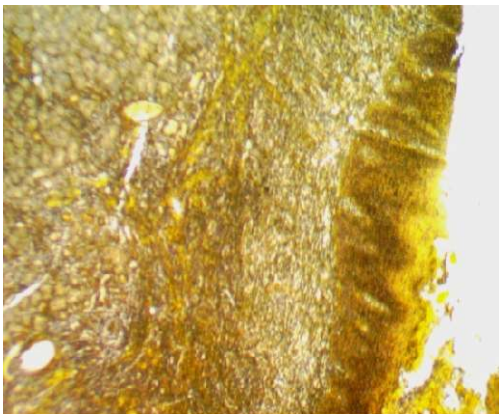

Figure 1

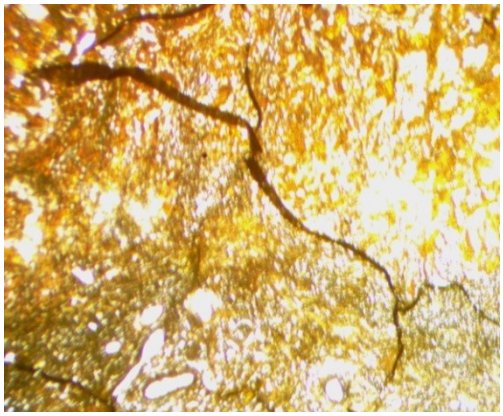

Figure 2

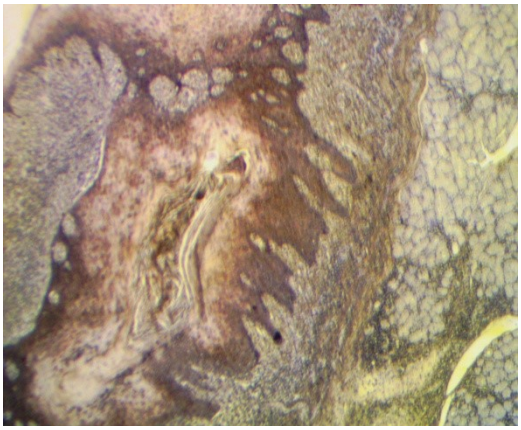

Figure 3

DISCUSSION:

Natural dyes has a tendency in producing extraordinary diversity of rich and complex colours along with unexpected results, making them exciting to use. Certain factors like electrostatic attraction determines the ability of a dye to stain specific tissue structures.

Acidic structures (e.g. nucleus) would be stained by basic dyes (e.ghematoxylin) while basic structures (e.g. cytoplasm) would be stained with acidic dyes (e.g. eosin) **[1, 5]**. Chemical exposures in these processes cause various health hazards to the laboratory technicians, pathologists, and scientists working in the laboratory. Hence, it is mandatory to introduce healthy and bio-friendly alternatives in the field.

This literature review explores the natural products and their efficiency to be used as alternatives for chemicals in the histopathology lab. Principle of staining suggest that there is an ionic bond between the tissue components and the dye, which is associated with the electrostatic attraction between dissimilar ions. A number of factors are responsible for better staining such as dye concentration, time of action on the solvent, its aqueous or alcoholic nature and most importantly pH of the solvent **[6]**.

CONCLUSION:

To go organic is the theme of the present study in order to combat the global warming. Implementing bio-compatible substitutes in routine histopathology is necessary. Though natural products are cost effective and non-hazardous, the efficiency and commercial availability of chemical products makes them indispensable. Furthermore, studies should be made with an aim to explore more natural products. For this studystandardization or modification of the stain can be done in order to prove it is better or equivalent to eosin.

REFERENCES

[1] OG. Avwioro, PC. Aloamaka, NU. Ojianya, T. Oduola, EO. Ekpo, Extracts of Pterocarpusosunas a histological stain for collagen fibres, African Journal of Biotechnology Vol. 4 (5), 460-462 (2005).

[2] HM. Carleton, RAB. Drumy, EA. Wallington, R. Cameron, Histological technique, Oxford University Press. London 1976.

[3] R. Bhuyan, CN. Saikia, Isolation of colour components from native dye-bearing plants in Northeastern India, Am. J. Pathol. 164 (5), 873-77 (2004).

[4] A.J. Akinloye, H.C. Illoh, A.O. Olagoke, Screening of some indigenous herbal dyes for use in plant histological staining, EJEAFChe. 9 (4), 772-779 (2010).

[5] OG. Avwioro, Histochemistry and tissue pathology, 1st ed., Claverianun press, Nigeria, 2002.

[6] Tousson, E., and Al- Behbehani, B. 2011.Black mulberries (*Morus alba*) as a natural dye for animal tissue staining. Animal Biology. 61(1): 49-56.