



## The Museum Maze in Oral Pathology Demystified—Part I

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### ABSTRACT

Museum technologies provide a wide array of choice of museums to those who wish to exploit technology to attract, excite and ensure an unrivalled visitor experience, as well as capture and sustain share of mind and heart. Museum being a combination of both art and science requires skilled workmanship, meticulous planning and execution to exhibit a specimen to its optimal elegance due to its relatively smaller size and fragile nature. A well established oral pathology museum is rarely seen due to negligence of oral specimens, dearth of knowledge in this field and also available data on it. An insight on oral pathology museum, including its establishment, importance and advanced technologies to make it more simple and accessible are discussed in two parts. Part I emphasizes on basics in oral pathology museum, whereas part II highlights the specialized techniques and recent advances in museum technology. Our effort is to present this article as hands on experience for the pathologists, student population and the technicians.

**Keywords:** Kaiserling's solution, Museum technique, Oral pathological specimens, Specimen mounting, Tissue preservation.

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### INTRODUCTION

A museum is defined as 'a nonprofit, permanent institution in the service of society and its development, open to the public, which acquires, conserves, researches, communicates and exhibits the tangible and intangible heritage of humanity and its environment for the purposes of education, study and enjoyment—Statutes of International Council of Museums.<sup>1</sup> The objective of the museum is to provide a running visual revision of high-end teaching quality which has the advantage of always being open.<sup>2</sup>

The demonstrations and exhibits of the museum should have an esthetic appeal as well as pedagogical purpose and they should be designed to make things clearer.<sup>3</sup>

The successful museum specimen, like the good histological preparation is dependent upon many minor technical points, and calls for an artistic presentation.<sup>4</sup> Here arise different methods of collection, preservation and display of museum specimen. All these together constitute 'museum technology'.

### EXHIBITS IN ORAL PATHOLOGY MUSEUM

Any museum should present a student with the full picture of human disease.<sup>5</sup> In particular, Oral pathology museum should contain head and neck specimens displayed systematically, along with its photomicrographs of the histopathology slides. Also, relevant clinical photographs, radiographs make the display complete. Various kinds of unique microscopes, casts or models of deciduous, mixed and permanent dentition with their anomalies, natural/carved tooth specimens, informative posters evoke interest in students and laymen. Not the least, there should be photographs and inspiring words of contributors in the field of dentistry.

### GUIDELINES FOR SETTING UP OF A MUSEUM

- Plan/layout of the museum be made and presented at the entrance along with specified timings for the visitors.
- Motto to be put up
- Guidelines to the visitors should be displayed for proper use of the museum
- A data on the total number and type of specimens on display should be put up at the entrance and has to be updated periodically.
- A catalog or brochure should be provided for the visitor to guide in navigation of the museum
- Specimens need to be arranged system-wise

- Museum jars to be labelled
- A book should be kept for feedback and suggestions.

## IMPORTANCE OF A MUSEUM

A well organized museum should serve the following purpose:

- Useful as teaching aid for students: it is said by Sir William Osler that, ‘We expect too much of the students and we try to teach them too much. Give them good methods and a proper point of view, and all other things will be added, as their experience grows.’ A museum is one such method of teaching
- Educative tools for public
- Preservation of uncommon specimens, e.g. syphilitic oral lesions, tuberculous ulcers<sup>6</sup>
- Archival collection of specimens that depict history
- Preservation of tissue as evidence in forensic field.

## REQUISITES OF A MUSEUM

The skeletal outline for setting up of the museum is as follows:

- Large, well-illuminated room. Fluorescence lighting is preferred<sup>5,7</sup>
- The museum room should be spacious enough to display the specimens systematically
- The entire area of the museum should be smartly utilized with convenient space for visitor navigation
- Shelves for systematic display of specimens
- Tables, chairs and white boards for teaching purposes
- A curator’s room with exhaust fan attached to it
- Museum should be preferably near the entrance of the institution so as to be the center of attraction to the visitors.

## METHOD OF MOUNTING THE SPECIMEN

Various methods of mounting the specimens have been developed for more than a century. Irrespective of the method used, mounting of the specimen includes 6 basic steps:

1. Collection of the specimen
2. Preparation
3. Fixation and color restoration
4. Treatment before mounting
5. Storage of specimens before mounting (if required)
6. Mounting.

Different methods vary in the solutions adopted for fixation, color restoration and mounting whereas the first two steps are common in all. Of all, Pulvertaft’s modification (1936) of the Kaiserling method (1900) is still the most widely used method for mounting of museum specimens.<sup>8</sup> Three solutions are employed in this method (Table 1).

The whole procedure starting from collection of the specimen to mounting using Pulvertaft’s modification of Kaiserling method is discussed hereafter.

**Table 1:** Solutions required for Pulvertaft’s modification of Kaiserling method<sup>8</sup>

| Fluid used               | Purpose           |
|--------------------------|-------------------|
| Kaiserling fluid no. I   | Fixation          |
| Kaiserling fluid no. II  | Color restoration |
| Kaiserling fluid no. III | Mounting          |

## COLLECTION OF THE SPECIMEN

It is most desirable that a centralized clearing station for specimens should be established.<sup>5</sup> Two major sources of specimen may be postmortem/autopsy specimen and surgical material from operation theater.<sup>5,8</sup> Specimens should be received with complete details of the patient/lesion. Every specimen has to be given due importance as Sir Arthur Keith said, ‘Never treat as junk anything that has been brought to you’<sup>9</sup> as we may unearth some hidden treasure. Each specimen should be assigned a museum reference number so as to avoid any confusion.

## PREPARATION OF THE SPECIMEN

Preparation of the specimen is carried out in the curator’s room. It should be well illuminated, ventilated and should be equipped with all the apparatus summarized in Table 2.

### Grossing

- Grossing of the specimen can be done either before fixation or following it. It is ideal to gross the larger specimen in a fresh state and smaller following fixation.<sup>7</sup>
- The details of the lesion: number, size, shape, color, consistency, weight, any cystic changes, necrosis, hemorrhage in the specimen should be recorded.<sup>13</sup>
- Cut surfaces should be smooth and even. This is achieved by using a continuous stroke with a long bladed sharp knife.<sup>8</sup>

### Special Considerations during Grossing

- Saline is most preferred over tap water to wash the specimens as contact with tap water causes hemolysis.<sup>5</sup>
- In case of a cystic specimen, capillary tubes might be used to keep the thin lining from folding in/collapsing into the lumen. Smaller cysts should be filled with a clear acrylate to maintain the patency of the cystic lumen.<sup>12</sup>
- Photographs of fresh or fixed specimens are captured to aid in documentation of pathologic lesions.<sup>7</sup>
- The radiographs are preferred for radiologic-pathologic correlation. Specimens suitable for radiography include bone lesions, calcified soft tissue masses, lesions with embedded tooth, radiopaque foreign bodies, ducts/vessels after injection of radiopaque material and for locating lymph nodes in radical neck dissection specimens.<sup>10,14-16</sup>

## FIXATION AND COLOR RESTORATION

Specimens should be put into a primary fixative immediately after the surgical procedure and the volume of the fixative should be in excess of 20 times the volume of the specimen.<sup>7,8</sup> Fixation time varies based on the size of the specimen. It requires 1 hour per mm of tissue thickness.<sup>17</sup> Ten percent neutral buffered formalin can be used for primary fixation and then the specimen is transferred to a Kaiserling fluid No. I or may be directly fixed in Kaiserling fluid no. I. fluid.<sup>7,8</sup> Larger specimens need injection of the fixative for uniform penetration. Unopened cystic cavities should be injected with fixative; if opened they should be packed with cotton-wool.<sup>5</sup>

Composition of Kaiserling fluids and their importance is elaborated in Table 3.

Specimen is washed gently in running water.<sup>4</sup> No. II fluid may be used to restore color in an emergency (e.g. for photography). The time in this solution should be controlled; continued immersion in alcohol has a permanent bleaching effect.<sup>8</sup>

Specimen is washed again in running tap water and placed in solution no. III.<sup>4</sup> The pH of the mounting fluid (no. III) is important. Colors could be preserved well at pH 8.0, but tend to fade if the pH changes. Hence, pH of this solution is adjusted to 8.0 with N/1 sodium hydroxide. Sodium hydrosulfite is added immediately before sealing the jar.<sup>8</sup>

## TREATMENT OF SPECIMENS BEFORE MOUNTING

When specimens are retrieved from storage they usually require meticulous attention to details before being actually mounted. Slight irregularities may have developed on the surface of the specimen during fixation and it may need to be recontoured. Unopened cysts and cavities may, if thin walled, need to be supported by the injection of gelatine, after removal of injected fixative. Specimens which are particularly friable may be covered with a thin layer of arsenious acid- gelatine to support them. (As suggested by Wentworth, 1947).<sup>5,8</sup>

## STORAGE OF SPECIMENS BEFORE MOUNTING (IF REQUIRED)

The storage of potential display specimens is important because the supply of these specimens usually exceed the number actually mounted. The method of storage must permit easy and certain identification of each specimen. The container should be adequately labelled from on the outside. A reference book should be kept to record necessary details (Specimen reference number, date of storage, etc.) of the specimens.<sup>8</sup>

## PROCEDURE FOR MOUNTING OF SPECIMEN

The following requirements should be within reach while mounting a museum specimen: specimen to be mounted,

**Table 2:** Requirements of curator's room

| <i>Armamentarium</i>                                                                                                                                                                                                                                                       | <i>Purpose</i>                                                                                                                                                                                                                                                                                                                                                                                               |
|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <ul style="list-style-type: none"> <li>Goggles, nasal mask and hand gloves</li> <li>Cutting board, box of instruments with scissors, forceps, probe, scalpel handle, disposable blades, long-bladed sharp knife, ruler, dissecting microscope.</li> </ul>                  | As a safety measure for the curator <sup>7</sup><br>Soft tissue grossing <sup>10</sup>                                                                                                                                                                                                                                                                                                                       |
| <ul style="list-style-type: none"> <li>Table vacuum vise</li> <li>Hard pressed carton</li> <li>Hand saw, tooth extraction forceps</li> <li>Saline</li> <li>Capillary tube and clear acrylate</li> <li>Photographic facility, balances, X-ray unit with view box</li> </ul> | Device for immobilization of hard tissue during grossing <sup>11</sup><br>To stabilize the hard tissues during sectioning <sup>11</sup><br>Additional instruments for hard tissue grossing and tooth extraction <sup>11</sup><br>To wash the specimen <sup>5</sup><br>To maintain the patency of cystic lumen <sup>12</sup><br>For documentation and better understanding of pathologic lesions <sup>7</sup> |

**Table 3:** Composition of Kaiserling fluids and their importance

|                                             | <i>Reagents</i>                                                                                                | <i>Importance</i>                                                                               |
|---------------------------------------------|----------------------------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------|
| Kaiserling fluid no. I (Fixative)           | Formalin (40%) (400 ml)<br>Potassium nitrate (30 gm)<br>Potassium acetate (60 gm)<br>Tap water (Up to 2000 ml) | Basic fixative <sup>8</sup><br>Preservative <sup>18</sup><br>Preservative <sup>18</sup><br>—    |
| Kaiserling fluid no. II (Color restorative) | Ethyl alcohol 80%                                                                                              | Restores the color of the specimen <sup>8</sup>                                                 |
| Kaiserling fluid no. III (Mounting fluid)   | Glycerine (300 ml)<br>Sodium acetate (100 gm)<br>Formalin (5 ml)<br>Tap water (Up to 1000 ml)                  | Preservative <sup>18</sup><br>Antimicrobial, buffer <sup>19</sup><br>Fixative <sup>8</sup><br>— |

Perspex jars, nylon thread, Perspex cement, long needle, Kaiserling fluids III and specific label.

- Perspex jars are universally preferred over glass jars for mounting as they are break resistant.
- The specimens are laid on a flat waterproof bench in the position in which they are to be mounted. The specimen is then measured, allowing 1cm clearance at the top and sides and 2 cm at the bottom. The depth of the specimen is measured, and approximately 5 mm added for the center plate. A suitable Perspex jar is then chosen from stock.
- The specimen is arranged in the desired position, stitched onto the center plate (center plate of a contrasting color can be used for better demonstration of the specimen) using nylon thread.
- Mounting fluid is run in to within 1 cm of the top and then the specimen is placed in the jar. Air bubbles trapped between the specimen and center plate are released with a broad-bladed spatula and then the jar is filled with the mounting fluid.
- The top of the box is wiped dry and Perspex cement applied. After 30 seconds the lid is laid lightly in position, surplus Perspex cement being carefully removed.
- After a further 30 seconds, a lead weight is applied and left for at least 1 hour.
- The holes on the lid are sealed with Perspex cement after 48 hours after all the air bubbles escape out of the jar (Figs 1A to F).<sup>8</sup>

### Advantage of Pulvertaft's Modification of Kaiserling Method

The original specimens mounted by this method show remarkably little fading even after 35 years.<sup>8</sup>

### TROUBLESHOOTERS

A thorough review on the possible problems encountered during mounting procedure and remedial measures are enlisted in Table 4.

### STAINING OF THE GROSS SPECIMEN

Large gross specimens may be stained before mounting to enhance the macroscopic view of the specific components like amyloid, fat, etc. (Table 5).

### DISPLAY OF THE MOUNTED SPECIMEN

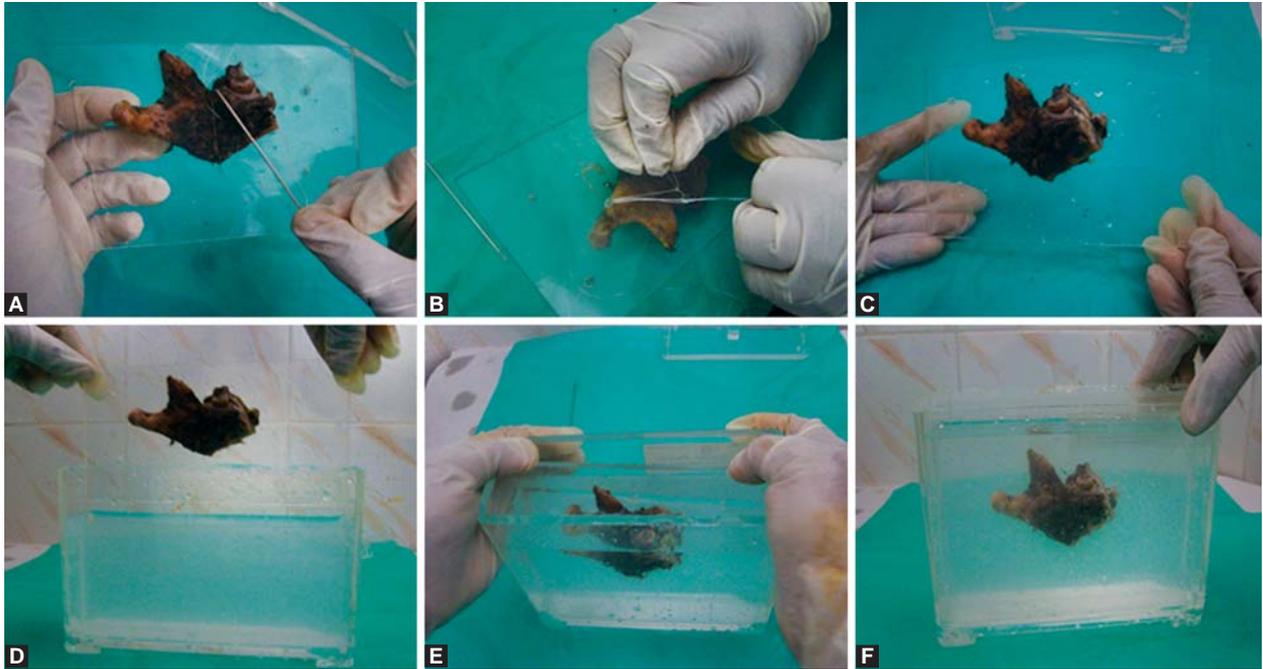
- Color coding the specimen-specimens may be grouped into different categories and each category assigned a specific color according to our convenience. (e.g. cysts-yellow, benign-blue or malignant lesions-red)
- Labeling of the museum jar-reference number, name of the lesion, color coding for the category of the specimen are the most desired details on the label of the museum jar (Fig. 2).
- Equal importance has to be given to the statistical significance of each disorder, and the changes which are taking place in the incidence of each.<sup>5</sup>

**Table 4:** Troubleshooters<sup>4</sup>

| <i>Problems/errors</i>                                         | <i>Cause</i>                                                      | <i>Preventive measures/corrections</i>                                                     |
|----------------------------------------------------------------|-------------------------------------------------------------------|--------------------------------------------------------------------------------------------|
| • Greyish areas                                                | Incomplete fixation due to specimen resting against the container | The specimen to be submerged in the fixative solution                                      |
| • 'Bacon-rind' patches                                         | Air-drying before fixation                                        | Immediate immersion of specimen in fixative postoperatively                                |
| • Specimen breakages                                           | Friable                                                           | Cover with gelatine                                                                        |
| • Adherence of droplets of fat to the walls of the museum jars | Fatty specimens                                                   | Complete fixation by providing sufficient time in accordance with the size of the specimen |
| • Growth of fungi                                              | Airborne fungal contamination                                     | Chilled mounting preferred.                                                                |
| • Discoloration of the mounting fluid                          | Hemoglobin from hemolysis caused by washing in water              | Thymol to be added to the mounting fluid.                                                  |
|                                                                |                                                                   | – Liquid paraffin to be used or surfaces to be painted with gelatine                       |
|                                                                |                                                                   | – Remounting of the specimen in liquid paraffin preferred                                  |

**Table 5:** Staining techniques for specific components of the specimen

| <i>Type of tissue</i>                         | <i>Staining technique</i>                  | <i>Result</i>                                                                                                                                                            |
|-----------------------------------------------|--------------------------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| • Amyloid                                     | Iodine technique <sup>5</sup>              | Amyloid typically stains mahogany-brown, and this color reaction changes to blue (A 'starch-like' reaction) after the application of dilute sulfuric acid. <sup>20</sup> |
|                                               | Congo red technique <sup>5</sup>           | Pink or orange <sup>20</sup>                                                                                                                                             |
| • Hemosiderin and free iron                   | Perls' Prussian blue reaction <sup>5</sup> | Blue/purple deposits <sup>21</sup>                                                                                                                                       |
| • Fat                                         | Sudan III or oil red O method <sup>5</sup> | Red <sup>22</sup>                                                                                                                                                        |
| • Fat necrosis                                | Copper acetate (Benda's test) <sup>5</sup> | Green <sup>23</sup>                                                                                                                                                      |
| • Cellular tumors                             | Hematoxylin <sup>5</sup>                   | Blue <sup>8</sup>                                                                                                                                                        |
| • Respiratory epithelium and mucous membranes | Alcian blue <sup>5</sup>                   | Blue <sup>8</sup>                                                                                                                                                        |



**Figs 1A to F:** Steps in mounting of oral specimen: (A) the specimen is held in desired position; (B) specimen is stitched onto the center plate; (C) the center plate with attached specimen, (D) specimen is placed in Perspex jar containing mounting fluid; (E) position of the specimen as seen from above, with required clearance around the specimen; (F) the jar is sealed with Perspex cement and the lid is laid in position



**Fig. 2:** Color coded, labelled oral specimen-ready to display

- Clinical, radiographical, surgical details of the specimen in brief can be displayed in a chart beside the specimen.

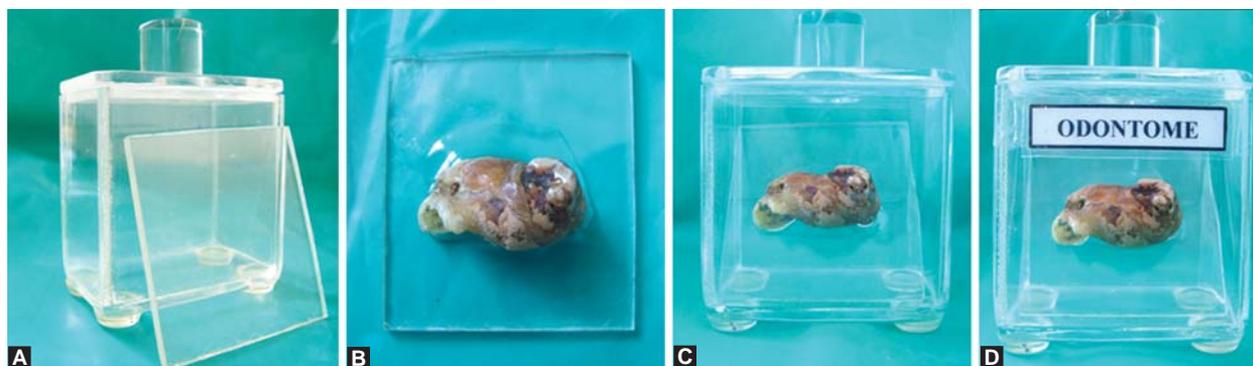
### SPECIAL METHOD FOR MOUNTING TOOTH SPECIMENS

The tooth specimens should be initially immersed in hydrogen peroxide for a day to dissolve out debris. Then they can be attached to the glass slide using acrylate glue/DPX, to facilitate handling and preservation. The glass slide with attached specimen can be inserted into a small Perspex jar, labelled and displayed as shown in Figures 3A to D.<sup>12</sup>

### HEALTH HAZARDS AND SAFETY MEASURES

Staff members working in the curator room encounter many possible risks including infections, chemicals which may be flammable, toxic, allergenic or carcinogenic, electrical and physical hazards as well as cuts and needle stick injuries.<sup>24</sup> Bone dust as well as bone fragments and crumbles, disseminated in the working environment are potentially biohazardous.<sup>25</sup> Formalin is highly toxic. The International Agency for Research on Cancer (IARC) classifies formaldehyde as a human carcinogen that can cause nasopharyngeal cancer.<sup>26</sup> Acute exposure to formaldehyde can, however, cause various health-related issues such as irritation on various body parts (eyes, nose, throat and skin). Moreover, sustained exposure can lead to certain types of cancers (e.g. nasopharyngeal) and asthma.<sup>27</sup>

These can be minimized by proper tissue handling and fixation of the specimen before grossing. All tissues must be considered potentially hazardous and universal precautions must be taken as per occupational safety and health administration guidelines. Adequate protective measures to protect from infection must be undertaken such as disposable gloves, facemasks and eye gear. Contact with chemicals should be minimized and the protective gear should be disposed off in correct manner. The laboratory personnel should clean the instruments and wash hands regularly to avoid spread of infection.<sup>7</sup>



**Figs 3A to D:** Tooth specimen mounting: (A) a small perspex jar with the lid selected based on the size of the specimen; (B) tooth specimen attached to the center plate in the desired position using sufficient DPX; (C) the center plate with the specimen is placed in the jar and the lid is sealed with Perspex cement; (D) labelled tooth specimen-ready to display

## CONCLUSION

The preservation of pathological material has never been of greater importance than at present, when the introduction of new, successful methods of therapy is changing the picture of disease out of all recognition.<sup>5</sup>

Every histologist must be able to prepare rare or important specimens for permanent preservation and display. The need for this must always be kept in mind when accepting a specimen for histology.<sup>5</sup> Also, creativity and innovations add lustre to the exhibits. 'A great museum is a laboratory where ideas get tested, not a mausoleum full of dead thoughts and bromides.'<sup>28</sup> It would be very much acceptable and noteworthy if oral specimens deserve a due importance in all medical museums. Museums in dental colleges have been ignored, awareness and training is the need of the hour. There's a definite need for awareness and training among all the students and staff regarding the importance of museum and its wide applications.

In addition to the old time honored methods of museum technology, newer methods are now available and are being developed to improve museums and their reach to a greater number of people. Specialized methods for different specimens and recent discoveries in the field of museum technology and its application in oral pathology are highlighted in part II of this article.

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