

Colposcopy: The Direct Oral Microscopy

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ABSTRACT

Oral squamous cell carcinoma (OSCC) is the commonest cancer of the oral cavity. Oral cancer is usually first diagnosed when it becomes symptomatic and by this stage, approximately two-thirds of patients would already have advanced disease leading to poor prognosis. The early diagnosis of oral premalignant lesions prevents their progression to an invasive carcinoma, thus requiring minimal intervention and improving both the survival rate and quality of life. But, diagnosis of a dysplastic premalignant lesion of the oral mucosa cannot be based solely on clinical findings and supplementary histopathological analysis of a representative biopsy specimen is necessary. Therefore, the selection of a biopsy site is highly significant. However, the site for the biopsy is a subjective choice that sometimes raises doubts about its representativeness. Hence, a need for a non-invasive yet accurate technique for early diagnosis of premalignant and malignant oral lesions is long been recognized. Colposcopy is one such examination procedure that seems to offer advantages in selecting more representative sites for biopsy and shows more advanced histologic signs than those selected by routine clinical examination alone. This article presents a current review on colposcope, assessment and interpretation of colposcopic findings and oral applications of the colposcopy technique as an adjunct in the early diagnosis of premalignant and malignant lesions of the oral mucosa.

KEYWORDS: Biopsy, Colposcopy, Dysplasia, Squamous cell carcinoma

INTRODUCTION

Oral squamous cell carcinoma is the commonest cancer, accounting for more than 90% of all the cancers affecting the oral cavity.¹ It is one of the predominant causes of death worldwide. The survival rates for oral cancer patients will significantly be improved provided lesions are detected and treated at the stage of infancy. Early diagnosis is therefore of paramount importance.² However, the diagnosis of dysplastic pre-malignant lesions of oral mucosa cannot be based solely on clinical findings.³ For this reason, numerous diagnostic techniques such as vital staining, exfoliative cytology, oral brush biopsy and light-based detection systems have been developed to complement the clinical examination and to facilitate the identification of initial carcinomas. However, the authenticity of these methods are questionable and hence, are not accurate to establish a final diagnosis.³⁻⁷ Though, biopsy with histopathologic examination is still considered as the gold standard for the diagnosis of premalignant and malignant oral lesions, but the selection of the site for biopsy is still critical.⁸ Since, biopsy from a non-representative area may be misleading, there is a need for a non-invasive, more accurate diagnostic tool that will aid in selecting the most representative site for biopsy. Colposcopy is a novel diagnostic technique that was thought of to serve the purpose. The principle objective of colposcopy is to prevent malignant progression of the premalignant lesions through early detection and treatment. Here, a review on colposcope, assessment and interpretation of

colposcopic findings and oral applications of the colposcopy technique as a useful chair-side adjunct in the early diagnosis of premalignant and malignant lesions of the oral mucosa is presented.

HISTORICAL BACKGROUND

Colposcopy is a well-established examination procedure in gynaecology where it is used to examine an illuminated magnified view of vagina or cervix with good resolution using an instrument known as colposcope.⁹ The Greek words 'kolpos' means 'fold or hollow' and 'skopos' means 'to examine'.¹⁰ The first colposcope has been reported to be constructed in 1925 by Professor Hans Hinselmann of Hamburg, Germany, who attached a light source to the Leitz binocular dissecting microscope specifically for the purpose of detecting early cervical cancer.¹¹ However, it is in the year 2000, Goran Gynther first used colposcope for examining the oral cavity for mucosal lesions.³

COLPOSCOPE: THE INSTRUMENT

A colposcope is defined as a stereoscopic binocular field microscope with a long focal length and powerful light source, which provides an illuminated magnification of the area of interest 4-40 times its normal size.

Types of Colposcope:

There are two types of colposcope:

- Those whose magnification can be altered by

How to cite this article:

Asha ML, Ghorai L, Rajarathnam BN, Lekshmy J, Mahesh KHM. Colposcopy: The Direct Oral Microscopy. *Int J Oral Health Med Res* 2016;3(2):81-86.

changing the power of eyepieces

- Those whose magnification can be altered by changing the setting of the knob or pressing a button¹²

Parts of Colposcope: Each colposcope comprises of:

- Colposcope Head
- Filters
- Light Source
- Height adjustment knob
- Head inclination knob
- Stand

Colposcope Head: It consists of eyepieces, objective lenses, and three step magnification knob :

- **Eyepieces:** The axes of the eyepieces may be straight or usually inclined at 45° to the optical axis of the apparatus. The eyepieces should be provided with features to adjust for the individual's interocular distance and should have focusing elements that can be adapted to the individual's eyesight for the ease of operation. Compensation for ametropia is provided from + 7 to - 7 dioptries.
- **Objective lens:** It has a specific focal distance and, in turn, influences the working distance. The lenses with a focal distance between 250 and 300 mm are considered ideal for effortless manipulation of the instrument.
- **Magnification knob:** The three step magnification knob provides low, medium and high powers for visualization of tissues. Low power (×2-6) may be used to study the surface architecture of the area of interest whereas medium (×8-15) and high (×15-25) powers are used to examine the deeper layers of the tissue and to identify certain vascular patterns indicative of premalignant and malignant lesions.

Filters: The colposcope is usually fitted with green or blue filters. Unlike the unfiltered white or yellow light, the green or blue light absorbs the red light from the colour and enhances the image of the blood vessels which appear black, thereby improving the contrast between the capillaries and surrounding tissue.

Light Source: The light source should have the minimum capacity of the illuminance of 30,000 lux, should be centered over the instrument and should be provided with a rheostat knob to alter the intensity of illumination. Usually, the colposcopes are provided with a halogen bulb with strength ranging from 6 to 12 V and 20 to 75 W. However, a state-of-the-art innovation is the light-emitting diode (LED), which generate light not by heating a wire filament, like in a halogen lamp but by semiconductor processes. Hence, though expensive, it is cooler for the patient and the colposcopist. Moreover, the LED lights have luminous efficiency five to seven times higher than a halogen lamp, provide improved contrast and the life span is approximately 10,000 hours more than a halogen lamp.

Height adjustment knob: It is used to adjust the height of the instrument and includes a gross height adjustment knob and a fine height adjustment handle.

Head inclination knob: It is used to adjust the angulation of the colposcope head

Stand: It mounts the colposcope head, filters and light source and those provided with wheels allow for portability of the instrument from one place to another within the room.

Colposcope provides three-dimensional representation of the scanned tissue surfaces. Sometimes, colposcope may be provided with portable video camera so that image can be viewed on a television monitor screen. A digital colposcope not only permits the real-time viewing of the colposcopic image on the display screen, but also enables future manipulation of stored images.^{2, 12-14}

COLPOSCOPY: THE PROCEDURE

Colposcopic examination is a painless non-invasive procedure without the requirement of anaesthesia. The procedure includes acetic acid wash followed by application of lugol's solution before the magnification.

Due to the similarity between the mucosal anatomy and the types of cancer found in the oral cavity and cervix, acetic acid seems to be an appropriate clinical marker for the detection of oral cancer as well.¹⁵ 3% acetic acid soaked in a cotton swab for a minute is applied to the oral mucosa for about 30 seconds.¹⁶ The areas that appear white after the acetic acid wash are called aceto-white lesions. The application of acetic acid causes coagulation of mucus which is then easily removed, and the aceto-white effect is due to the reversible coagulation of the nuclear-proteins and cytokeratins.^{17, 18} Another concept is given by Burke et al. which states that the reaction is a result of a reversible osmolar change leading to cytoplasmic dehydration and cytoplasmic membrane collapse. This cellular change produces more reflected light, and hence, the colposcopic image appears white.¹⁹ The dysplastic cells having larger nuclei with abnormally large amounts of chromatin are therefore affected mostly. Just as the aceto-whitening begins to fade, the vascular patterns will be clearly visible. The epithelium being colourless, it acts as a filter through which both the incident and the reflected light pass and the redness of the stroma is transmitted through the epithelium and is visible through the colposcope. The visibility depends on the number of layers of epithelium, morphology and the organization of the epithelial cells, vascularity and nature of the underlying stroma, amount of tissue chromophores and the amount of haemoglobin.¹⁹⁻²¹

The aceto-whitening develops in about 40-60 seconds and then fades over a similar time scale.² The speed of appearance and disappearance of the aceto-white effect depends on the nucleo-cytoplasmic ratio and the number of cells. The immature metaplastic epithelium turns shiny white in colour which expresses its self-renewal ability, but this whitening disappears fast, whereas an opaque and long-lasting aceto-whiteness is noted in the areas of high-grade malignancy.¹² However, studies have shown that chronic trauma, chemical and mechanical irritation, such as that related to the use of tobacco, also cause increased

aceto-white effect. Moreover, the age of the subject also influences acetic acid staining as age-related degenerative changes makes the epithelium less responsive to acetic acid.²

Following acetic acid application, if no lesions are visible, Lugol's solution or Schiller's solution is used for further examination. Lugol's solution is applied to the mucosal lesion using a cotton swab or given as a rinse when no definite lesions are visible. Staining of mucosa with Lugol's iodine is dependent on the glycogen content of the tissue. The normal epithelium, being rich in glycogen, stains brown while the inflammatory or carcinomatous epithelium having poor glycogen content, does not retain the stain.²² Thus, both the acetic acid solution and Lugol's solution are used to improve the visualization of abnormal areas.

COLPOSCOPY: THE CRITERIA FOR DIAGNOSIS

The assessment and interpretation of the colposcopic findings is based on the following factors:

- Vascular pattern
- Inter-capillary distance
- Surface contour
- Colour tone and opacity
- Clarity of demarcation of the mucosal lesions

Vascular pattern: The normal mucosa exhibits two types of capillary patterns, Network capillaries, and Hairpin capillaries, with fine and regular caliber. The normal vascularity can be altered in various inflammatory, benign and malignant lesions and conditions. Inflammatory epithelium exhibits terminal capillaries which being overloaded with blood, is seen through the epithelium as intense red colouration without co-existing clinical and subclinical signs of a dysplastic lesion. The abnormal epithelium demonstrates three different types of capillary networks: Punctate, Mosaic and Atypical capillaries.

Punctate capillaries: These are characterized by dilated, often twisted, irregular, hairpin-type of vessels which terminate on the surface and appear from the ends as a collection of dots. Hence, the name punctuation. Punctuation is the commonest type of abnormal capillary network that is seen. It is also known as ground appearance.

Mosaic capillaries: These are characterized by terminal capillaries extending only partially into the epithelium and forming a basket around the roughly circular or polygonal blocks of aceto-white epithelium crowded together, exhibiting an appearance similar to mosaic tiles.

Atypical capillaries: These are characterized by terminal capillaries which may be looped, branched or reticular having sharp turns, accompanied by luminal dilatations and narrowing, thereby exhibiting irregular shape and size and coarse pattern of arrangement.

Fine punctuation and mosaicism, formed by narrow vessels and uniform inter-capillary distances usually signify low-grade lesions. A coarse pattern resulting from

a variable vessel caliber and increased inter-capillary spacing indicates higher grade abnormalities. The mosaic pattern with central punctuation is predictive of carcinoma in situ. Loss of previously intact mosaic and punctate patterns with the production of predominantly waste thread like vessels is an early colposcopic sign of squamous micro invasion or cancer.

Inter-capillary distance: The normal inter-capillary distance ranges from 50 to 200 μm . Once the neoplastic cells develop, they expand to perhaps exert pressure on these capillaries displacing those outwards. Now the inter-capillary distance is increased and randomly distributed. This randomly wide inter-capillary distances greater than 200 μm indicate malignancy. The distance between the capillaries increases as the degree of dysplasia increases, so that the maximum distance may exceed 700 μm .

Surface contour: A healthy mucosa presents moist, glossy, smooth and unfolded surface. In the areas of neoplastic growth, the surface epithelium ulcerates as the cells lose intercellular cohesiveness secondary to the loss of desmosomes and due to the propensity of hemidesmosomes to lose their adhesive properties in the neoplastic process. As a result, the surface may appear irregular or pebbled exhibiting a slightly depressed relationship to the elevated surrounding squamous and metaplastic epithelium.

Colour tone and opacity: The intact squamous epithelium appears yellow-orange rather than the expected pink colour. The colour tone may change as a result of the increasing vascularity, surface epithelial necrosis, and in some cases, production of keratin. On application of 3-5% acetic acid, the high-grade lesions display a more persistent duller shade of white, whereas low-grade lesions appear to be translucent or bright white and fade quickly.

Clarity of demarcation of the mucosal lesions: Low-grade lesions have feathery margins and irregular borders. High-grade lesions exhibit well-defined borders with straighter and sharper outlines. A lesion with an internal border, signifying a lesion within a lesion is typically indicative of high grade variety. These are the result of vascular and tissue changes.

After a complete detailed examination, the areas with the highest degree of visible abnormality are isolated and further subjected to biopsy and histopathological analysis.^{2, 11, 23-27}

Apart from the above colposcopic criteria given by Goran N. Gynther, the other grading and scoring systems that are used in gynaecology, include:

- Combined colposcopic index by Reid and Scalz [Table 1]²⁸
- Grading system of Coppleson et al. [Table 2]¹⁷
- Grading system of Burke and co-workers [Table 3]²⁹

COLPOSCOPY: THE SUGGESTED ORAL APPLICATIONS

Applications related to diagnosis of mucosal lesions:-

Colposcopic sign	Zero point	One point	Two points
Margin	Condylomatous or micro-papillary contour indistinct Aceto-whitening flocculated or feathered margins. Angular, jagged lesions, satellite lesions and aceto-whitening that extend beyond the transformation zone	Regular lesions with smooth, straight outlines	Rolled, peeling edges internal demarcations between areas of differing appearance
Colour (after application of acetic acid)	Shiny, snow-white colour indistinct aceto-whitening	Intermediate shade (shiny grey)	Dull, oyster-white
Vessels	Fine-calibre vessels, poorly formed patterns; condylomatous or micro-papillary lesions	Absent vessels	Definite punctuation or mosaic
Iodine	Positive iodine staining	Partial iodine uptake	Negative staining of significant lesion

- Scores of 0-2 usually indicates minor lesion (HPV or CIN)
 - Scores 3-5 usually indicates a middle-grade lesion (CIN2)
 - Scores of 6-8 usually indicate significant CIN (CIN2 or 3)
- HPV: Human papillomavirus; CIN: Cervical intraepithelial neoplasia

Table 1: Combined colposcopic index by Reid and Scalz

Grades	Features	Predicted histology
Grade 1 (insignificant, not suspicious)	Ace to white epithelium, usually shiny or semi-transparent, borders not necessarily sharp, with or without fine calibre vessels, often with ill-defined patterns, absence of atypical vessels; small inter-capillary distance	Metaplastic epithelium (both immature and mature), acantholytic epithelium, subclinical papillomavirus infection (SPI) and low grade CIN (CIN1)
Grade 2 (significant, suspicious)	Aceto-white epithelium with greater opacity with sharp borders; with or without dilated calibre, regularly shaped vessels; absence of atypical vessels; usually increased inter-capillary distance	High-grade CIN (CIN2or 3)
Grade 3 (Highly significant, highly suspicious)	Very white or grey opaque epithelium; sharply bordered; dilated calibre, irregularly-shaped, often coiled, occasionally atypical vessels; increased but variable inter-capillary distance and sometimes irregular surface contour (micro-exophytic epithelium).	CIN3 or early invasive cancer

Table 2: Grading system of Copleson et al.

Grade	Surface	Margin	Colour	Time	Vessels	Pathology
1	Flat	Indistinct	Normal/ slightly white	Appears slowly, remains for short time, disappears rapidly	Fine with normal ICD	SPI, inflammation, immature metaplasia, pregnancy, regeneration, repair
2	Flat	Distinct	Whiter	Average time to appear, remains for several minute ,disappears with average speed	Punctuations, mosaics lightly increased ICD	SPI, CIN1 and CIN2
3	Raised	Sharp	Whitest	Appears rapidly, stays a long time, disappears slowly	Coarse punctuations and mosaic, increased ICD, atypical vessels	CIN3

Table 3: Grading system of Burke and co-workers, CIN: Cervical intraepithelial neoplasia; SPI: Subclinical papilloma virus infection; ICD: Inter-capillary distance

- Direct optical visualization of vascular and tissue changes clarifies the nature of clinically suspicious mucosal lesions, aids in the follow-up of such lesions and also determines the effects of the treatment provided²
- Patients with oral squamous cell carcinomas usually have a marked inflammatory infiltrate, and this inflammation interferes with the evaluation of the dysplastic changes. However, a biopsy sample selected by colposcopy rarely shows a severe inflammatory infiltrate, providing better representative histologic signs of the underlying disease³

Other Oral Applications:-

- Intra-oral anatomical structures such as the papillae of the tongue can be three-dimensionally visualized owing to good resolution and incredible depth of visualization
- Surfaces of the teeth can be examined by colposcopic method which reveals details such as wear facets and the margins of restorations
- The colposcope has a resolution of 2 µm and the

erythrocytes being 7 µm in diameter, the instrument allows ready and detailed observation of flow in the larger blood vessels. This feature could be advantageous in the monitoring of grafting procedures in the oral cavity.³⁰

COLPOSCOPY: MERITS AND DEMERITS

Colposcope offers advantage to see what a healthy naked eye cannot. The direct optical visualization of vascular pattern changes, surface pattern, colour tone and opacity, clarity of demarcation of mucosal lesions by colposcopy provides better scope of selecting the more representative biopsy site than routine clinical examination alone, thereby avoiding multiple biopsies and providing broader range of diagnoses.² The main advantages of colposcopy over the other existing chair-side methods include high resolution, good magnification, good illumination, good storage capacity, early detection of lesions, painless procedure, non-invasive nature and an accuracy rate of 80-90%.⁹

Although colposcopy is technique sensitive and expensive, these drawbacks seem to have minor importance compared to the advantage it provides in terms of precision in clinical diagnosis.¹²

CONCLUSION

‘Screening is the key to early diagnosis’- Colposcopy aptly serves as a new ray of hope in early detection of premalignant and malignant oral mucosal lesions owing to its high precision, versatility, user friendly and non-invasive properties. Although this method should be evaluated in further clinical studies in terms of its accuracy and efficacy and a comparative evaluation should be carried out with other existing methods, but colposcopy can definitely aid in better diagnosis and should be implemented in routine clinical practice to provide better patient care than what we, the oral diagnosticians can presently offer.

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Source of Support: Nil
Conflict of Interest: Nil