DIAGNOSTIC TEST FOR CANCER DETECTION IN DENTAL & ENT CLINICS: THE TOLUIDINE BLUE TEST.


Abstract
There are various important investigations which are required for the diagnosis and treatment plan of various disorders related to oral cavity. The use of toluidine Blue (toluidine chloride) dye as a mouthwash is currently receiving much attention as an aid to the diagnosis of oral cancer and potentially malignant lesions. The method has good sensitivity with a very low false negative rate.

Introduction:
Oral Cancer is a global health problem with increasing incidence and mortality rate. India accounts for 86% of the world's oral cancer cases, according to the study conducted by the National Institute of Public Health in February 2011. An early oral cancer can appear as innocuous red or white patches, as an ulcer, or a lump, mimicking many benign lesions that may develop in an area of previously healthy epithelium or from a precancerous lesion. If these lesions are undetected and untreated, they can invade and destroy adjacent structures and spread to lymph nodes in the neck and to distant sites.

Additionally, when the discomfort is minimal, professional consultation often is delayed, increasing the chance for local spread and regional metastases. As dental surgeons are health personnel specialized in dental and oral health, moreover dental surgeons are the first one to examine the patient before others. So it is the responsibility of the dentist to examine any suspicious lesions and detect cancerous and precancerous lesion as soon as possible in their clinics, so that early detection and appropriate referral can be made.

There are different methods of screening for oral cancer. Clinical Examination is the current most common method used to detect visible lesions. On the other hand, for clinically undetectable lesions, there is need for an adjunctive method to visual examination to detect precancerous lesions. The current adjunctive methods are toluidine blue, brush biopsy and fluorescence imaging.

An effort is made through this paper to review and educate Dental Surgeons, ENT Surgeons for early detection of Oral cancerous and precancerous lesion with the help of Toluidine blue dye.

Toluidine blue mouth rinse for screening precancerous and cancerous lesions in oral cavity:
There are various important investigations which are required for the diagnosis and treatment plan of various disorders related to oral cavity. The use of toluidine Blue (toluidine chloride) dye as a mouthwash is currently receiving much attention as an aid to the diagnosis of oral cancer and potentially malignant lesions. The method has good sensitivity with a very low false negative rate.

Indication:
It is effective in demonstrating dysplasia and early malignant lesion which is not clinically recognizable.

Mechanism:

Binding with DNA:
It is an acidophilic, metachromatic nuclear dye of Thiazine group that selectively stains acidic tissue components particularly nucleic acid such as DNA and RNA. Dysplastic areas contain more DNA.

Intracellular canal:
Malignant epithelium contains wider intracellular canal which facilitates the penetration of dye.

Effect on normal epithelium:
Most of the epithelial surfaces stain blue after application of 1% toluidine blue solution but the stain is lost after application of 1% acetic acid to normal epithelial surface.

Effect on benign ulceration:
Benign ulceration has well defined uptake of dye at the margins whereas diffuse marginal pattern is characteristic of dysplasia or malignancy.

Technique:
Step 1: Rinsing: - initially ask the patients to rinse the mouth twice with water (20 sec each). After rinsing ask the patient to rinse with 1% acetic acid.
Step 2: Drying of area: - gently dry suspicious mucosal areas with gauze. Care should be taken not to abrade the tissue while drying.
Step 3: application of toluidine blue solution: - Apply 1% solution to the lesion with the cotton swab.
Step 4: Rinsing: - Ask the patient to rinse again with acetic acid. After rinsing ask the patient to rinse with water.
Step 5:- Positive staining: - If the mucosa is stained positive, repeat the procedure in 1-2 weeks. Biopsy of all the sites is advised, which stains positive on successive 2nd visit.

Ear, nose and throat Surgeon is recommended:
In the monitoring of suspicious lesions over time. In the screening of pre cancerous lesions in high risk population.
In helping to determine an optimal site for biopsy, where a suspicious lesion is present.

Discussion:
Toluidine blue, a basic metachromatic dye, stains the nuclear material of malignant lesions, but not that of normal mucosa. The nuclei of cancer cells show increased DNA synthesis, resulting in increased pickup of toluidine blue. A Toluidine blue rinse may be utilized in screening to include all high-risk sites. Lesions not detected during a visual examination may therefore be revealed by the stain.

Toluidine blue was first used by Richart in 1963 to stain uterine cervical carcinoma in Situ. Unfortunately false positives have appeared high in early oral studies because of the conclusion of traumatic lesions and other oral inflammatory studies. Imtiaz et.al, Silverman S, Migliorati, Strong et al advocate use of toluidine blue to detect malignant lesions. These studies indicate that all or part of carcinomas or carcinomas in situ stain blue with topical application.

Screening with toluidine blue has its advantages and disadvantages. Screening may offer the opportunity to reduce the incidence of invasive lesions and help in decreasing the mortality rate associated with oral cancer. The disadvantage is that screening might be associates with significant problems related to false positives such as psychological trauma. In 1980 Masberg showed that false positives using applications for asymptomatic lesions could be reduced to 8.5% by a second evaluation of lesions in 10-14 days thereby allowing for the clinical healing of traumatic and non malignant inflammatory lesions. Richard et.al expressed concerns regarding the general use of toluidine blue without proper training may result in false positive and false negative results that will be detrimental to public health. Specific training is needed for correct application of the test and correct interpretation of the result. The gold standard for the diagnosis of oral cancer is biopsy and histological examination. The use of toluidine blue dye has been suggested as an adjunct to visual examination in the identification and management of oral cancer. It is not suggested that the failure to stain a visually observable lesion should rule out the need to biopsy such a lesion. It is a chair side test which can be performed easily in every Dental & ENT Clinics, cost effective for the patients. Patients who have fear for surgical procedures, this test can be an alternative.

In a country like India, betel nut, paan, tobacco chewing
is more prevalent along with smoking. These are high risk factors for developing cancerous lesions, dental surgeons and ear, nose and throat surgeons most commonly encounter such patients, so clinical examination and Toluidine blue screening are recommended for every patient with suspicious lesions. It is the responsibility of medical and dental institutions to provide necessary training to conduct such screening test at community level.

**Conclusion:**
Toluidine blue staining is a useful diagnostic adjunct, it may be used as a screening rinse in high risk patients for Dental Surgeons and ear, nose, throat surgeons who encounter pre cancerous lesions in everyday practice, so that they can refer to a specialist centres experienced in the diagnosis and treatment of oral cancer and potentially malignant lesions or conditions.

**References:**
1. Times of India Newspaper: May 31, 2011