*International Journal of Pharmaceutical Science Invention*
*ISSN (Online): 2319 – 6718, ISSN (Print): 2319 – 670X*
*www.ijpsi.org Volume 3 Issue 2 ǁ February 2014 ǁ PP.07-12*

# Clinico-Pathologic Evaluation & Medical Treatment of Oral Leukoplakia

Dr. Nasir A Salati
*Assistant professor, Department of Oral pathology, Dr Z.A. Dental College, A.M.U., Aligarh, India*

***ABSTRACT:*** *Leukoplakia is defined as a predominantly white lesion of the oral mucosa that cannot be characterized as any other definable lesion. Histopathological diagnosis is needed to evaluate prognosis of this lesion. Chewing tobacco and smoking are distinct risk factors particularly among males in certain countries; however it has been noted that females or non-smokers may be at risk of malignant transformation. HPV has been detected in oral dysplasia lesions and cancer in non-smokers. Malignant transformation rates of oral leukoplakia (OL) range from 0.13 to 17.5%, while the rates of five year cumulative malignant transformation range from 1.2 to 14.5%. Although the administration of retinoic acid and beta-carotene has some efficacy to resolve OL, the role of photodynamic therapy (PDT) is believed to play an important therapeutic role in these patients. This article emphasizes need of uniform classification methods to evaluate the malignant potential of leukoplakia, and new treatment avenues in its resolution.*

***KEYWORDS:*** *Epithelial dysplasia, Histopathology, Oral leukoplakia, Malignant transformation, Photo-sensitisers, White lesions.*

## I. INTRODUCTION

According to WHO definition (1978)[1], premalignant lesion is defined as "a morphologically altered tissue in which cancer is more likely to occur than in its apparently normal counterpart. Examples: leukoplakia, erythroplakia, palatal keratosis associated with reverse smoking. WHO (1978) defined an oral precancerous condition as "a generalized pathological state of oral mucosa associated with a significantly increased risk of cancer." Examples: OSMF, lichen planus, DLE, epidermolysis bullosa. Warnakulasuriya[2] *et al* have recommended abandoning the distinction between the terms "potentially malignant lesions" and "potentially malignant conditions" and to use the term "potentially malignant disorders" instead. According to them, the term "potentially malignant disorders" means that not all abnormalities may transform into cancer, but some may have an increased potential for malignant transformation.

Leukoplakia is most common premalignant, potentially malignant or precancerous lesion of the oral mucosa. The word leukoplakia is derived from two words, *leuko* meaning white and *plakia* meaning patch. Leukoplakia is usually found in between fourth to seventh decade of life, but has been found to occur in initial decades of life also. Males have been found to be more affected than females. According to Petti[12] *et al* (2003), oral pre-malignant diseases affect males at least three times as often as females. Clinically leukoplakia exhibits varying features.[20, 45] The surface can be smooth or wrinkled, sometimes smooth surfaced lesions may be traversed by small cracks or fissures .The lesions may be white, whitish yellow or gray. The surface may be homogenous, ulcerated, or speckled. The homogenous varieties are uniformly flat and thin having shallow cracks of surface keratin giving an appearance likened to cracked mud. The non-homogenous varieties, like nodular and verrucous have irregular surfaces.

## II. FEATURES OF ORAL LEUKOPLAKIA

Leukoplakia is generally defined by exclusion of other diseases, particularly white lesions.[2-7] W.H.O. defines it as "A white patch or plaque that cannot be characterized clinically or pathologically as any other disease and is not associated with any other physical or chemical causative agent except use of tobacco." According to Warnakaulasuriya[8] *et al*., in 2007, "Leukoplakia should be used to recognize white plaques of questionable risk having excluded (other) known disease or disorders that carry no increased risk for cancer." Leukoplakia is caused by cumulative effect of various risk factors.[29, 30] Wynder[9] *et al* (1957) applied the terms 'intrinsic' and 'extrinsic' to group factors which act together to produce a premalignant state. Exogenous or "extrinsic" factors in leukoplakia include tobacco, alcohol and mechanical trauma. The endogenous or "intrinsic" factors include systemic or generalized states such as malnutrition or iron deficiency anemia and genetic factors. Tobacco has been considered as an important causative agent.[31, 34, 35, and 36] The role of human papilloma virus (HPV) in the etiology of premalignant and malignant lesions has been studied extensively. In a

study done by Nielsen[10] *et al*, HPV was mentioned as a likely causative agent. Meta analysis by Miller and Johnstone[11] (2001), reported that the likelihood of detecting HPV was 2-3 times higher in precancerous oral mucosa. HPV genotypes 16 and 18 have been detected in many cases of leukoplakia. Mehta[14] *et al* (1971) classified leukoplakia in three categories: 1) *Homogenous leukoplakia*: Characterized by a raised plaque formation consisting of a plaque or groups of plaques varying in size with irregular edges. The lesions are predominantly white but can be grayish white or yellow. 2) *Ulcerated leukoplakia:* Characterized by a red area at times with yellowish area of fibrin, giving the appearance of ulceration. 3) *Nodular (speckled) leukoplakia:* Characterized by white patches on an erythematous base. Banoczy[15] *et al* in 1982, categorised leukoplakia in three types: *Type I or Leukoplakia simplex:* Keratinized mucosa with flat or very slightly elevated white lesions. *Type II or Leukoplakia* verrucosa: Verrucous proliferation raised above the mucosal surface. *Type III or Leukoplakia erosive:* White lesion with erythematous area or erosion, and fissures. In an International seminar on Oral leukoplakia, Malmo, Axell[3] *et al* (1984) classified leukoplakia as: *Homogenous leukoplakia (simplex):* A uniform whitish lesion with a smooth or corrugated surface. *Erythro-leukoplakia (erosive leukoplakia):* A whitish lesion that includes red areas. *Nodular leukoplakia:* A lesion with slightly raised, rounded, red and whitish excrescences that may be described as granules or nodules. *Verrucous leukoplakia:* An exophytic lesion with irregular sharp or blunt projections.

In some prospective study on large samples from India, carried out in several geographic areas with various kinds of tobacco usage, the annual age-adjusted incidence rates of leukoplakia varies per 1000 population per year.[37,38]The annual screening program reported from Japan showed that the age-adjusted incidence rate for leukoplakia was high in male than in female per 100, 000 persons. The age-adjusted incidence rate for tobacco-associated leukoplakia in males was almost 12 times as compared to females. The reported incidence of oral leukoplakia in Japanese population was somewhat higher to those reported from India. The prevalence of oral leukoplakia varied from 1.1% to 11.7%. Axell[43] *et al*, studied the prevalence in Swedish community. According to Petti[12] *et al* (2003), the global prevalence was about 2.6%. A study done by Gupta[13] *et al (*1980) showed the annual incidence of 1.1 to 2.4 percent among men and 0.2 to 0.03 percent among women.

.

## III. MICROSCOPIC FEATURES OF LEUKOPLAKIA

Leukoplakia can present as benign hyperkeratosis, mild dysplasia, moderate dysplasia, severe dysplasia or invasive carcinomas microscopically. *Mild dysplasia* histologically shows hyperplasia of cells of the basal layers, cytological atypia with mild pleomorphism of cells and nuclei, and minimal architectural changes. Moderate dysplasia histologically shows proliferation of atypical cells seen upto one-thirds of epithelium with prominent cytological changes. Hyperchromatism and cellular pleomorphism are seen. Architectural changes are seen in form of loss of polarity of basal cells. In *severe dysplasia*, abnormal proliferation of cells is seen from basal layer to upper third of epithelium. There is marked pleomorphism of cells with changes in nuclear-cytoplasmic ratio. Architectural changes are severe, with loss of stratification. In *carcinoma-in-situ*, the cytological and architectural changes are clearly evident throughout whole epithelium.

The 14th International Cancer Congress in Hungary suggested the following microscopic changes for the diagnosis of oral epithelial dysplasia: Drop-shaped rete processes, Disturbed nuclear polarity, Basal cell hyperplasia, Disturbed epithelial maturation, Pleomorphic cells, Anisocytosis, Hyperchromatic nuclei, Prominent nucleoli, Increased nuclear - cytoplasmic ratio, Cell crowding, Increased number of mitoses, Abnormal mitoses & Reduced cellular cohesion.

Pindborg in 1997 proposed the following criteria for epithelial dysplasia: Loss of polarity of basal cells, Presence of more than one layer of cells having basaloid appearance, Increased N/C ratio, Drop shaped rete-ridges, Irregular epithelial stratification, Increased number of mitotic figures, Mitotic figures that are abnormal in form, Presence of mitotic figures in the superficial half of the epithelium, Cellular and nuclear pleomorphism, Nuclear hyperchromatism, Enlarged nuclei, Loss of intercellular adherence & Keratinization of single cells or cell groups in prickle cell layer.

WHO[48] classification of head and neck tumors gave cellular and architectural changes for grading leukoplakia. (Table: 1)

**Table 1: Microscopic Features of Leukoplakia**

| Cellular changes | Architectural (tissue) changes |
|---|---|
| • **Abnormal variation in nuclear size (anisonucleosis)** | • **Loss of polarity** |
| • **Abnormal variation in cell size (anisocytosis)** | • **Disordered maturation from basal to squamous cells** |
| • **Increased nuclear/cytoplasmic ratio** | • **Increased cellular density** |
| • **Enlarged nuclei and cells** | • **Basal cell hyperplasia** |
| • **Hyperchromatic nuclei** | • **Dyskeratosis (premature keratinization and keratin pearls deep in epithelium)** |
| • **Increased mitotic figures** | • **Bulbous drop-shaped rete pegs** |
| • **Abnormal mitotic figures (abnormal in shape or location)** | • **Secondary extensions (nodules) on rete tips** |
| • **Nuclear and cellular pleomorphism** | |
| • **Increased number and size of nucleoli** | |

## IV. DISPARITY IN DIAGNOSIS OF VARIOUS GRADES OF LEUKOPLAKIA

The progression rates from oral epithelial dysplasia are varied.[39,40,45] Several studies have shown greater inter and intra - observer variability study in the assessment of various grades of dysplasia.[42] Although generally moderate or severe dysplasias (in-situ carcinomas) show a greater disposition for malignant transformation than mild or non-dysplastic cases[44], carcinomatous transformation may also take place in non dysplastic cases.[28] Very small lesions have been found to exhibit dysplastic changes.[32, 33] The WHO Collaborating Centre for Oral Pre-cancerous Lesions, established in Copenhagen in 1967, recognized three major problems that were attached to the importance of epithelial dysplasia in predicting malignant development: (i) the final diagnosis was essentially subjective. (ii) Not all lesions showing dysplasia would eventually become malignant, and some even regressed. (iii) Carcinoma developed from lesions in which epithelial dysplasia had not been diagnosed in previous biopsies. Nowadays the role of biomarkers is considered to be helpful in evaluation of oral leukoplakia.[47, 48]

## V. MALIGNANT POTENTIAL OF LEUKOPLAKIA

According to Amagasa [16] *et al*., (2006), the percentage of leukoplakia that progresses to invasive squamous cell carcinomas ranges from 0.13 to 17.5 percent, and the rates of five year cumulative malignant transformation ranges from 1.2 to 14.5 percent. Gangadharan[17] *et al*., (1971), in the study of 626 leukoplakias found malignant transformation rate to be 10 percent over a mean period of 8-9 months. Mehta[18] *et al.,* (1972) in a 10 year follow-up study of 117 leukoplakias found mean transformation rates of 0.9 percent only. Gupta[13] *et al.,* (1980) observed malignant transformation rate higher among women than men, and more in persons chewing tobacco. In their study they also found that patients with oral leukoplakia carry a five-fold higher risk of developing oral cancer than controls. Burkhardt[46] *et al.,* (1985) reported that mild, moderate and severe dysplasia has malignant potential of about 3%, 4% and 43% respectively. Zhang et al., in 2001 analyzed 71 epithelial dysplasia cases from the floor of the mouth, ventro-lateral tongue, and soft palate, designated as high-risk sites and 56 epithelial dysplasia cases from other sites of the oral cavity, designated as low-risk sites. The results were not influenced by gender or smoking. They found that epithelial dysplasias from high-risk sites had a higher frequency of loss of heterozygosity and a pattern of loss associated with an increased risk of progression to malignancy. Bouquot[41] *et al*., (2006) reported that severe dysplasia has an overall malignant transformation rate of about 16% with 7-50% range. Speight[19] *et al.,* (2007) reported that moderate dysplasia have malignant transformation potential of 3-15%, whereas mild epithelial dysplasia show a very low risk (<5%). Silverman[21] *et al* (1976) in a study found that 11%of lesions altered clinically and exhibited increased tendency towards.Banoczy[22]*et al*., found 13% of lesions undergoing malignant transformation. studies by silverman[23] *et al*(1984), Holmstrup[24] *et al*., Lind[25] (1987), Schepman[26] *et al*., (2.9%) and shiu[27] *et al*., found malignant transformation rates 17,5%, 20%, 7%, 2.9% and 2.6% respectively.

## VI. TREATMENT OF ORAL LEUKOPLAKIA

### 6.1 BETA-CAROTENES:

The use of beta-carotene has been recommended in order to prevent OL and possibly oral cancer.[49,50,51] The potential benefits and protective effects against cancer are possibly related to its antioxidizing action.[52,53,54] This function is accomplished through a ligation between beta-carotene and oxygen, which is an unstable reactive molecule, thus diminishing the damaging effects of free radicals.[54] According to Liede et al.[52], a diet

supplemented with beta-carotene can prevent changes in the oral mucosa, especially in smoker patients, who present low serum levels of vitamin C and beta-carotene when compared to non-smokers. It has also been shown that beta-carotene has a better therapeutic clinic response in the prevention of oral leukoplakia lesions, and in smoker patients than in the non-smoker ones.[55]

**6.2 LYCOPENE:**

Lycopene is a carotenoid without provitamin A action. Lycopene is considered one of most efficient biological antioxidizing agent.[56] There is a positive relationship between lycopene consumption and a reduction in the risk of the development of degenerative diseases caused by free radicals, such as cancer and cardiovascular diseases[57] Due this property, studies have been done to find out whether or not it could be an alternative to protect patients against the damaging effects of free radicals. Lycopene is believed to modify intercellular exchange junctions, and so effective in potentially malignant disorders. [56]

**6.3 RETINOIDS:**

At the cellular-level, retinoids interact with surface receptors and penetrate the cell. They are subsequently metabolized and transported to the nucleus by various proteins. Retinoids affect diverse processes, such as keratin production, the expression of growth factors and kinases, oncogenesis, apoptosis, production of the collagen matrix, immunologic and inflammatory response, cellular differentiation, embrionary morphogenesis and carcinogenesis.[50] 13-cRA is the retinoid recommended for OL treatment. The use of 13-cRA has been shown to be effective in resolving OL.[53,54] However, the high recurrence rates after short periods of discontinuance, together with its side effects, are limiting factors.[54,58] Various studies have evaluated the therapeutic effectiveness of vitamin A derivatives in the treatment of OL. In one study, patients received a fixed dose of 13-cRA (10 mg/day) plus an escalating dose (beginning at 800 IU/day, until 2000 IU/day) for 4 months. Seventy-one percent of OL patients had complete clinical responses.[59]

**6.4 PHOTOSENSITISERS:**

Among various photo-sensitisers, 5-Aminolaevulinic acid (ALA) has been used for treatment of oral leukoplakia. 5-Aminolaevulinic acid (ALA) was also approved in several countries for the treatment of skin cancer; The ALA is a naturally occurring compound in the haem biosynthetic pathway, which is metabolised to a photosensitive product, protoporphyrin IX (PpIX). The major advantage of ALA when compared to synthetic photosensitisers is the rapid metabolism, which significantly reduces the period of cutaneous photosensitivity. For very superficial skin lesions or premalignant lesions of the oral mucosa, the ALA can be applied topically. For all other indications intravenous application is mandatory[60]. Zakrzewska et al.[61] reported effective role of it than surgery & carbon dioxide laser in treatment of proliferative verrucous leukoplakia.

## VII. CONCLUSION

Disparity in evaluating oral leukoplakia can be minimised by uniform classification systems. Research is going on to evaluate role of specific biological markers in diagnosing and predicting the prognosis of pre-malignant disorders. The role of photosensitisers has shown promising results in its regression.

## VIII. ACKNOWLEDGEMENTS

I am thankful to senior staff and administration for their kind support.

## REFERENCES

[1] World Health Organization Collaborating Center for Oral Precancerous Lesions (1978). Definition of leukoplakia and related lesions: an aid to studies on oral precancer. Oral Surg Oral Med Oral Pathology l46:518-539.

[2] Nomenclature and classification of potentially malignant disorders of the oral mucosa S. Warnakulasuriya, Newell. W. Johnson, I. van der Waal J Oral Pathol Med (2007) 36: 575–80.

[3] Axell T, Holmstrup P, Kramer IRH, Pindborg JJ. International seminar on oral leukoplakia and associated lesions related to tobacco habits. Community Dentistry and Oral Epidemiology 1984; 12:145-54.

[4] Bouquot JE, Gorlin RJ. Leukoplakia, lichen planus, and other oral keratoses in 23,616 white Americans over the age of 35 years. Oral Surgery, Oral Medicine and Oral Pathology 1986;61(4):373-81.

[5] Axell T, Pindborg JJ, Smith CJ, van der Waal I. Oral white lesions with special reference to precancerous and tobacco- related lesions: conclusions of an international symposium held in Uppsala, Sweden, May 18-21 1994. International Collaborative Group on Oral White Lesions. Journal of Oral Pathology & Medicine 1996;25(2):49-54

[6] Pindborg JJ, Reichart P, Smith CJ, Vanderwaal I, WHO histological typing of cancer and precancer of oral mucosa;springer-verlag1997

[7] Gale N, Pilch BZ, Sidransky D, Westra WH, Califano J. Epithelial precursor lesions. In: Barnes L, Eveson JW, Reichart PA, Sidransky D, eds, World Health Organization Classification of tumors. Pathology and Genetics of Head and neck tumours. Lyon: IARC Press; 2005. p. 140-3.

[8] World Health Organization (1973). Report from a meeting of investigators on the histological definition of precancerous lesions. Can/731, Geneva. Cited by Pindborg JJ (1980) in Cancer and Precancer, John Wright and Son Ltd., Bristol

[9] Wynder EL, Bossij.feldmanr;a study of etiological factors of cancer of the mouth.cancer 1957;10;1300-1323

[10] Nielsen H, Norrild B, Praterious F, Reibel J, Holmstrup P.HPV in oral premalignant lesions. European Journal of Cancer Oral oncology. 1996 july;32 B (4):264-70

[11] Miller CS, Johnstone BM., Human papilloma virus as a risk factor for OSCC; a meta-analysis 1982-1987.OOOE 2001;91;622-635

[12] Petti S. Pooled estimate of world leukoplakia prevalence: a systematic review. Oral Oncology 2003;39:770-8011,20

[13] Gupta PC, Mehta FS, Daftary DK, Pindborg JJ, Bhonsle RB, Jalnawalla PN, et al.Incidence rates of oral cancer and natural history of oral precancerous lesions in a 10-year follow-up study of Indian villagers. Community Dentistry and Oral Epidemiology 1980;8(6):283-33.

[14] Mehta, F. S., Pindborg, J. J. &Hamner, J. E. (1971) Oral Cancer and Preeaneerous Conditions in India. Munksgaard Copenhagen, p. 138

[15] Banoczy et al (1983). Oral leukoplakia and other white lesions of the oral mucosa related to dermatologic disorders. I CutanPathol10:238-256.

[16] Amagasa T, Yamashiro M, Ishikawa H. Oral leukoplakia related to malignant transformation. J Oral SciInt 2006; 3(2):45-55.

[17] P. Gangadharan and J. C. Paymaster Leukoplakia—An Epidemiologic Study of 1504 Cases Observed at the Tata Memorial Hospital, Bombay, India Br J Cancer. 1971 December; 25(4): 657–668

[18] Fali S. Mehta, B. C. Shroff, P. C. Gupta and D. K. Daftary Oral leukoplakia in relation to tobacco habits: A ten-year follow-up study of Bombay policemen. Oral Surgery, Oral Medicine, Oral PathologyVolume 34, Issue 3, September 1972, Pages 426-433.

[19] Paul M. Speight Update on Oral Epithelial Dysplasia and Progression to Cancer. Head and Neck Pathol (2007) 1:61–66.

[20] Silverman S Jr. Observations on the clinical characteristics and natural history of oral leukoplakia.J Am Dent Assoc. 1968 Apr;76(4):772-7.

[21] Silverman S, Bhargava K, Smith Malaowalla AM.LW, Malignant transformation and natural history of oral leukoplakia in 57,518 industrial workers of Gujarat, India. Cancer. 1976 Oct;38(4):1790-5.

[22] Bánóczy J. Follow-up studies in oral leukoplakia J Maxillofac Surg. 1977 Feb;5(1):69-75.

[23] Silverman S Jr, Gorsky M, Lozada F. Oral leukoplakia and malignant transformation. A follow-up study of 257 patients.Cancer. 1984 Feb 1;53(3):563-8.

[24] Holmstrup P, Vedtofte P, Reibel J, StoltzeK.Long-term treatment outcome of oral premalignant lesions. Oral Oncol. 2006 May;42(5):461-74. Epub 2005 Nov 28.

[25] Lind PO. Malignant transformation in oral leukoplakiaScand J Dent Res. 1987 Dec;95(6):449-55.

[26] Schepman KP, van der Meij EH, Smeele LE, van der Waal I. Malignant transformation of oral leukoplakia: a follow-up study of a hospital-based population of 166 patients with oral leukoplakia from The Netherlands. Oral Oncol. 1998 Jul;34(4):270-5

[27] Shiu MN, Chen TH, Chang SH, Hahn LJ Risk factors for leukoplakia and malignant transformation to oral carcinoma: a leukoplakia cohort in Taiwan.Br J Cancer. 2000 Jun;82(11):1871-4.

[28] Holmstrup P, Vedtofte P, Reibel J, Stoltze K. Long-term treatment outcome of oral premalignant lesions.Oral Oncol. 2006 May;42(5):461-74. Epub 2005 Nov 28.

[29] Sankaranarayanan R, Thara S (2002).Upper Aerodigestive Tract. In: Franco EL, Rohan,TE eds. Cancer precursors, New York, Springer, 72-95. ISBN: 0387951881

[30] Thomas G, Hashibe M, Jacob BJ, Ramadas K, Mathew B, Sankaranarayanan R, Zhang ZF Risk factors for multiple oral premalignant lesions.Int J Cancer. 2003 Nov 1;107(2):285-91

[31] Bánóczy J, Sugár L Longitudinal studies in oral leukoplakias.J Oral Pathol. 1972;1(6):265-72.

[32] Waldron CA, Shafer WG. Leukoplakia revisited. A clinicopathologic study 3256 oral leukoplakias. Cancer. 1975 Oct;36(4):1386-92.

[33] Neville BW, Day TA Oral cancer and precancerous lesions. CA Cancer J Clin. 2002 Jul-Aug;52(4):195-215.

[34] Pindborg JJ, Reibel J, Roed-Petersen B, Mehta FS (1980). Tobacco-induced changes in oral leukoplakic epithelium. Cancer 45:2330-2336.

[35] Raque CJ, Biondo RV, Keeran MG, Honeycutt WM, Jensen GT (1975). Snuff dipper's keratosis (snuff-induced leukoplakia). South Med J 68:565-568.

[36] Reichart PA, Mohr U, Srisuwan S (1987). Precancerous and other mucosal lesions related to chewing, smoking and drinking habits in Thailand. Community Dent Oral Epidemiol15:152-160

[37] Mehta FS, Pindborg JJ, Gupta PC, Daftary DK (1969). Epidemiologic and histologic study of oral cancer and leukoplakia among 50,915 villagers in India. Cancer 24:832-849.

[38] Mehta FS, Pindborg JJ, Hamner JE III (1971). Oral cancer and precancerous conditions in India. Copenhagen: Munksgaard

[39] Lumerman H, Freedman P, Kempel S (1995). Oral epithelial dysplasia and the development of invasive squamous cell carcinoma. Oral Surg Oral Med Oral Pathol79:321-329

[40] Crissman ID, Zarbo RJ (1989). Dysplasia, in situ carcinoma, and progression to invasive squamous cell carcinoma of the upper aerodigestive tract. Am J SurgPathol13(Suppl):5-16

[41] Bouquot JE, Speight PM, Farthing PM. Epithelial dysplasia of the oral mucosa: Diagnostic problems and prognostic features. CurrDiagPathol 2006;12:11-21.

[42] Fischer DJ, Epstein JB, Morton TH, Schwartz SM. Inter-observer reliability in the histopathologic diagnosis of oral pre-malignant and malignant lesions. J Oral Pathol Med. 2004 Feb;33(2):65-70

[43] Axell T. Occurrence of leukoplakia and some other oral white lesions among 20,333 adult Swedish people. Community Dentistry and Oral Epidemiology 1987;15(1):46-51

[44] Bouquot JE, Whitaker SB (1994). Oral leukoplakia—rationale for diagnosis and prognosis of its clinical subtypes or "phases". Ouintessence \nt25:133-140-41.

[45] Prabu SR.,ORAL DISEASES IN TROPICS., STUDIES ON LEUKOPLAKIA

[46] Burkhardt A.: Advanced methods in the evaluation of premalignant lesions and carcinomas of the oral mucosa. J. OralPathol.*14*, 751- 778, 1985.

[47] Immunohistochemical detection of early-stage carcinogenesis of oral leukoplakia by increased DNA-instability and various malignancy markers M. Iwasa, Y. Imamura, S. Noriki, Y. Nishi, H. Kato, and M. Fukuda Eur. J. Histochemvol 45;pages 333-346,2001.

[48] Piattelli A Prevalence of p53, bcl-2, and Ki-67 immunoreactivity and of apoptosis in normal oral epithelium and in premalignant and malignant lesions of the oral cavity JOMSvolume 60,issue 5,page 532-540

[49] G. Britton, "Structure and properties of carotenoids in relation to function," The FASEB Journal, vol. 9, no. 15, pp. 1551–1558, 1995.

[50] R. S. Parker, "Absorption, metabolism, and transport of carotenoids," The FASEB Journal, vol. 10, no. 5, pp. 542–551, 1996. View at Scopus

[51] R. Sankaranarayanan, B. Mathew, C. Varghese, et al., "Chemoprevention of oral leukoplakia with vitamin A and beta carotene: an assessment," Oral Oncology, vol. 33, no. 4, pp. 231–236, 1997. View at Publisher · View at Google Scholar · View at Scopus

[52] K. Liede, J. Hietanen, L. Saxen, et al., "Long-term supplementation with alpha-tocopherol and beta-carotene and prevalence of oral mucosal lesions in smokers," Oral Diseases, vol. 4, no. 2, pp. 78–83, 1998. View at Scopus

[53] G. E. Kaugars, S. Silverman Jr., J. G. L. Lovas, et al., "A clinical trial of antioxidant supplements in the treatment of oral leukoplakia," Oral Surgery, Oral Medicine, Oral Pathology, vol. 78, no. 4, pp. 462–468, 1994. View at Scopus

[54] G. E. Kaugars, S. Silverman Jr., J. G. L. Lovas, J. S. Thompson, R. B. Brandt, and V. N. Singh, "Use of antioxidant supplements in the treatment of human oral leukoplakia: review of the literature and current studies," Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontics, vol. 81, no. 1, pp. 5–14, 1996. View at Scopus

[55] K. Malaker, B. J. Anderson, W. A. Beecroft, and D. I. Hodson, "Management of oral mucosal dysplasia with β-carotene retinoic acid: a pilot cross-over study," Cancer Detection and Prevention, vol. 15, no. 5, pp. 335–340, 1991.

[56] A. V. Rao and S. Agarwal, "Role of antioxidant lycopene in cancer and heart disease," Journal of the American College of Nutrition, vol. 19, no. 5, pp. 563–569, 2000. View at Scopus

[57] G. Riccioni, B. Mancini, E. Di Ilio, T. Bucciarelli, and N. D'Orazio, "Protective effect of lycopene in cardiovascular disease," European Review for Medical and Pharmacological Sciences, vol. 12, no. 3, pp. 183–190, 2008.

[58] J. A. Olson, "Carotenoids and human health," Archivos Latinoamericanos de Nutricion, vol. 49, no. 3, supplement 1, pp. 7S–11S, 1999. View at Scopus

[59] I. W. Dimery, W. K. Hong, J. J. Lee, et al., "Phase I trial of alpha-tocopherol effects on 13-cis-retinoic acid toxicity," Annals of Oncology, vol. 8, no. 1, pp. 85–89, 1997.

[60] K. Konopka and T. Goslinski, "Photodynamic therapy in dentistry," Journal of Dental Research, vol. 86, no. 8, pp. 694–707, 2007. View at Publisher · View at Google Scholar · View at Scopus

[61] C. J. Kelty, N. J. Brown, M. W. R. Reed, and R. Ackroyd, "The use of 5-aminolaevulinic acid as a photosensitiser in photodynamic therapy and photodiagnosis," Photochemical and Photobiological Sciences, vol. 1, no. 3, pp. 158–168, 2002