CLINICAL STUDIES OF ORAL EPITHELIAL DYSPLASIA

by

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ABSTRACT

This study has investigated a number of aspects of the clinical presentation, aetiology and management of oral epithelial dysplasia (OED) in a large number of affected patients. The results reveal that the peak frequency for occurrence of OED is the 6th decade, a predilection for males was confirmed. The tongue, buccal mucosa and floor of mouth are the most common sites of involvement. Most OED is mild; carcinoma-in-situ is rare, the sites likely to have areas of severe dysplasia are the floor of mouth and lateral border of tongue. There is a positive association between heavy tobacco smoking, especially non-filter cigarettes and risk of OED, although the risk may decline following cessation of smoking. In non-smokers, consumption of alcohol is not a significant predictor of OED but there is interaction between alcohol and some aspects of tobacco smoking. The relative risk associated with tobacco smoking appeared to be highest for OED of the labial mucosa and floor of mouth in males and tongue and floor of mouth in females. While alcohol drinking is not a significant predictor of specific OED subsites in both males and females. OED may occur in non-users of tobacco and alcohol, these patients tend to be older women presenting with erythroleukoplakic type lesions. There is a significant association between reduced serum folate, red blood cell folate and risk of OED. Infectious agents such as Candida albicans may have a slight significance in the aetiology of OED but infection with hepatitis C virus or Helicobacter pylori are not significantly associated with OED. Follow-up of these patients with OED suggest that 5.5% of patients develop oral squamous cell carcinoma while 10.3% develop a second dysplastic lesion and 17.5% develop recurrence.

It is evident that dental practitioners have some knowledge and experience of oral malignancy and premalignancy but they may fail to recognise appropriate signs and symptoms of such disease, do not always provide appropriate preventive advice and may delay referral of patients to appropriate centres.

DEDICATION

To my parents, brothers, sisters, my wife

and my daughter Fatima

DECLARATION

"Except for the help listed in the acknowledgements, the contents of this thesis are entirely my own work. This work has not previously been submitted, in part or in full, for a degree or diploma of this or any other University or examination board".

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LIST OF ABBREVIATIONS

ALA	Aminolaevulinic acid
ALT	Alanine transaminase
APH	Alkaline phosphatase
AST	Aspartate transaminase
BDA	British Dental Association
BDJ	British Dental Journal
BDS	Bachelor of Dental Surgery
BMJ	British Medical Journal
CAL	Computer-assisted-learning
CBC	Complete blood count
CCD	Charged couple device
CFU	Colony forming units
CI	Confidence intervals
CIS	Carcinoma-in-situ
CR	Complete response
DGDP	Diploma in General Dental Practice
DMSO	Dimethylsulphoxide
DNA	Deoxyribonucleic acid
ENT	Ear, Nose, Throat
ELISA	Enzyme-linked immunosorbent assays
EPI5	Epidemiology programme version 5
FDS	Fellow Dental Surgery
FOM	Floor of mouth
GDP	General Dental Practitioners
GGT	Gamma glutamyl transpeptidase
GMP	General Medical Practitioners
HIV	Human immunodeficiency virus
HPV	Human papilloma virus
IARC	International Agency for Research on Cancer
ICD	International Classification of Disease
LDS	Licentiate of Dental Surgery

MSc	Master of Science
NAC	N-acetylcysteine
OED	Oral epithelial dysplasia
OPCS	Office of Population Censuses And Surveys
OR	Odds Ratio
PAS	Periodic acid schiff stain
PBS	Phosphate-buffered saline
PDT	Photodynamic therapy
PhD	Doctor of Philosophy
PML	Potentially malignant lesions
PR	Partial response
PVL	Proliferative verrucous leukoplakia
RAS	Recurrent aphthous stomatitis
RBC	Red Blood Cell
RR	Relative risk
RMA	Retromolar area
SAQ	Self-administered questionnaire
SCC	Squamous cell carcinoma
SPSS	Statistical Package For Social Sciences
SLK	Sublingual keratosis
ST	Smokeless tobacco
STK	Smokeless tobacco keratosis
TIBC	Total iron binding capacity
UICC	International Union Against cancer
WHO	World Health Organisation

CHAPTER 1

Literature Review

Oral epithelial dysplasia

Introduction

Oral epithelial dysplasia (OED) is characterised histopathologically by cellular and morphological changes in the oral epithelium that are indicative of developing malignancy (WHO, 1978). The individual cellular changes are referred to as *atypia*, and the general disturbance in the epithelium is designated *dysplasia*. The presence of epithelial dysplasia is notably the single most important factor predicting risk for subsequent development of invasive neoplasia of the oral mucosa (WHO, 1978; Kramer, 1980; Crissman & Zarbo, 1989;Krutchkoff et al, 1991).

Terminology and definitions

Various epithelial abnormalities can occur in the oral cavity. Most are only benign lesions characterised histopathologically by hyperplasia or inflammation. However, a group of intraepithelial lesions with malignant potential known as dysplasia may also occur in the mouth.

Dysplasia implies bad development or abnormal formation (from the Greek *dys*, 'bad' or 'difficult'; *plasis*, 'forming'). As an adaptive response dysplasia signifies abnormal or atypical hyperplasia and sometimes atypical metaplasia; thus dysplastic change refers to an alteration in the size, shape, and organisation of the cellular components of hyperplastic or metaplastic tissue (Yeldandi et al, 1996). In one standard medical dictionary, dysplasia is defined as "abnormal tissue development". The term is applied to 30 separate diseases, with dysplasia of the uterine cervix being the only one among these referring to a biologic state related to cancer (Linder & Johnston, 1996). The cytopathologic concept of dysplasia can be traced back to Papanicolaou's original

descriptions of what he termed "dyskaryosis" (Papanicolaou, 1954). Cells in a state of gradual transition from normalcy to frank malignancy were described as "dyskaryotic". Their nuclei were characterised by increasing nuclear enlargement, uniform coarsening and hyperchromasia of the chromatin, and thickening of the nuclear membrane. Their cytoplasm exhibited an increasing failure to mature and differentiate. The end stage of this process was considered to be the cells of carcinoma in-situ.

Dysplasia was initially defined as "all other disturbances of differentiation of the squamous epithelium of lesser degree than carcinoma-in-situ" a vague definition modified by the World Health Organisation (WHO) (Poulsen et al, 1975) to "a lesion in which part of the thickness of the epithelium is replaced by cells showing varying degree of atypia" (Pindborg et al, 1977). This definition was later modified to "a spectrum of hetroplastic reactions involving stratified squamous or squamous-like (metaplastic) epithelium" (Patten, 1978). He elaborates further; "fundamentally, the dysplastic reaction is characterised by a combination of hyperplasia and a block in normal differentiation of the component cells as they reach the upper most layers. The resultant effect is a greater number of cells per unit area and abnormally large nuclei in the upper layers of the involved epithelium". Abrams (1978) has defined epithelial dysplasia as "an abnormality in the differentiation of proliferating cells such that there occurs an abnormal degree of variation in the size, shape, and appearance of the cells with a disturbance in the usual arrangement of those cells".

Krutchkoff and co-workers (1991) defined OED as "abnormal epithelial growth characterised by a combination of altered cellular morphology and/or disturbances in architecture or maturational pattern that may represent a "premalignant" state and not a purely developmental or reactive anomaly", although the term dysplasia as applied to squamous epithelium implies disturbances of differentiation and maturation of

epithelial cells and not abnormal epithelial growth. OED is now a collective term for various epithelial changes, seen by light microscopy, considered to indicate an increased likelihood of malignant transformation. The more severe forms represent carcinoma-in-situ (WHO, 1978; Crissman et al, 1987).

Cellular atypia

This denotes any of several abnormalities in cellular morphology or architectural arrangement described by light microscopy that could represent either an unusual reactive change (response to local irritants) or possibly true dysplasia. When there is uncertainty as to whether observed histopathologic abnormalities reflect unusual reactive change or actual dysplasia, the term "atypia" is appropriate (Krutchkoff et al, 1991). The cellular atypia of dysplasia is similar to that of squamous cell carcinoma. (Speight et al, 1996).

Histopathology

As in other sites, oral dysplasia is characterised morphologically by inappropriate intercellular relationships, loss of normal orderly maturation, and nuclear changes. There can be variable amounts of keratinisation and the upper epithelial surfaces may have irregularities such as nodules and chevrons ('church-spires'-pointed projections). Leukoplakia with visible surface chevrons are often referred to histoplathologically as verruciform hyperkeratosis. Below the keratin layer, the microscopic findings are variable and include features of cellular atypia (Table 1.1), although some of these features are also characteristic of regenerating epithelium and epithelial hyperplasias without malignant potential.

Cellular changes in dysplasia

Specific alterations of individual cells are important in the determination of epithelial dysplasia. Cells and nuclei take on a more primitive appearance, similar to those of basal cells with enlarged nuclei (*nuclear hyperplasia*), dark-staining nuclei (*hyperchromatism*), enlarged, often eosinophilic nucleoli (*prominent nucleoli*), and with an increased nuclear cytoplasmic ratio. These cells also appear to be crowded more closely together than normal keratinocytes (*increased cellular density*). There is often *increase in mitotic activity* in dysplastic epithelium, but this also is seen in many reactive lesions. Enlarged, tripolar or star-shaped mitotic figures (*abnormal mitosis*), however, are much more indicative of precancerous changes. Abnormal mitosis may also be defined as mitotic figures found in unusual locations above the basal cell layer. A key alteration of dysplastic epithelial cells is variation in the shape of the cells and nuclei. This *pleomorphism* is unusual outside of cancer and precancers. Premature production of keratin below the surface layer is another important alteration, but it is much more commonly seen in oral carcinomas than in oral potentially malignant lesions. This *dyskeratosis* may be represented by individually keratinized cells or by tight concentric rings of flattened keratinocytes (*epithelial pearls*). Not all keratosis is related to malignancy or potentially malignant lesions. Individually keratinised cells, for example, are also characteristic of hereditary benign intraepithelial dyskeratosis (Witkop disease) (Witkop, 1960; Von Sallmann & Paton, 1960). Cellular necrosis and *loss of cellular cohesiveness* (*acantholysis*) are major signs of poorly differentiated carcinoma but are extremely rare in OED. When present, these features must be distinguished from intercellular oedema, intraepithelial inflammatory cells and degenerating cells with pyknotic nuclei and vacuolated cytoplasm.

When dysplasia is seen in epithelium which otherwise has the microscopic features of lichen planus (liquifactive degeneration of basal cells, saw-toothed rete processes, hyperkeratosis, subepithelial band of inflammatory cells), the lesion is graded according to the above mentioned criteria for epithelial dysplasia, although the term lichenoid dysplasia may be applied to the case (Odukoya et al, 1985; Eisenberg & Krutchkoff, 1992).

Tissue (morphological) changes

Many potentially malignant lesions show excess surface keratin (hyperkeratosis, hyperparakeratosis, hyperorthokeratosis) and most show hyperplasia of the spindle cell layer *(acanthosis)*, but both changes are common to a number of mucosal lesions without a cancer transformation potential and neither is necessary to the diagnosis of dysplasia. *Basal cell hyperplasia* is of major importance to the diagnosis as well as to the grading of dysplasia.

There can be variable degrees of basal cell hyperplasia, although in many cases the basal hyperplasia is confined almost exclusively to the rete processes (Speight et al, 1996), giving rise to elongated processes. In OED the rete processes can appear "drop shaped". Rete process with bulbous enlargement of the lowermost region (drop-shaped rete processes) are worrisome regardless of their size, especially if secondary projections or nodules are seen to arise from the basal layer and branch at indifferent angles into the lamina properia and connective tissue papilla (Speight et al, 1996).

Dysplastic epithelium may be atrophic as well as acanthotic often lack rete processes and may be ulcerated, thereby mimicking a traumatic or inflammatory lesions with thin, regenerating epithelium creeping in from the margins. However regenerating epithelium often has granulation tissue beneath it.

An important morphological alteration of dysplastic epithelium is loss of stratification (*loss of polarisation*) due to an apparent inability to properly differentiate and mature from basal cells to prickle cells to flattened keratinocytes. Cells high in the epithelium have the same immature appearance as those in the basal layers. This feature is especially pronounced in severe epithelial dysplasia and carcinoma-in-situ.

It is unlikely that all the aforementioned features will be present in a single lesion, indeed this variable appearance creates difficulties in achieving consistency of diagnosis and interpretation between histopathologists.

Grading of epithelial dysplasia

When interpreting the changes seen in OED the main aims are to grade reproducibly the degree of dysplasia and to aid the referring clinician in translating the grade into an estimate of risk of malignant transformation. These aims are difficult to achieve with the oral mucosa (Pindborg et al, 1985).

Based on the histopathologist's interpretation of the extent and degree of dysplastic features, OED can be graded into three prognostically significant categories, all of which presume a lack of epithelial invasion into underlying connective tissue (WHO, 1978; Lurmerman et al, 1995; Speight et al, 1996). There may be strong interobserver discrepancy between pathologist's in the evaluation of the presence and the degree of epithelial dysplasia (Pindborg et al, 1985; Abbey et al, 1995) and it is not unusual for different pathologist's to place emphasis on different aspects of dysplastic alterations, thereby arriving at somewhat different diagnostic conclusions. Regardless of individual emphasis, however, certain grading criteria are in general use:

1. The cellular atypia or dysplasia is similar to that seen in squamous cell carcinoma
2. There is no evidence of invasion into undelying stroma.

3. The epithelium with the greatest proportion of atypical cells has the greatest risk of being or becoming a carcinoma.
4. The epithelium with the most extreme atypia of cells has the greatest risk of being or becoming a carcinoma.
5. The final grading or diagnosis should be based on the most severely involved area of change.

The classification system applied to the cervix, based on the proportion of epithelial thickness 'thirds' showing dysplastic changes, is difficult to apply to the mouth. The surface layers nearly always show near-normal cell morphology so some pathologists omit the most superficial few layers before applying the 'thirds' principle (Odell & Morgan, 1998). The use of thickness-affected as only one of several factors to be assessed. It is rare for the full thickness of the oral epithelium to show dysplasia and atypia is sometimes very marked even though very restricted to the basal and parabasal cells. Several calibration studies have revealed a lack of reproducibility in the recording of OED, both between histopathologists and by the same one different occasions. Not surprisingly, therefore, there are no fully defined criteria for grading dysplasia in oral epithelium (Odell & Morgan, 1998).

An estimate is made of the extent and severity of individual dysplastic features and the proportion of epithelial thickness affected, pathologists attempts to grade OED, usually expressed as mild, moderate and severe, the last being essentially in-situ-carcinoma (Crissman et al, 1987; Speight & Morgan, 1993). In mild dysplasia: there is a proliferation of atypical or immature basal cells above the parabasal region but not extending beyond the lower third of the epithelium. In moderate dysplasia the atypical cells extend into the middle one-third of the epithelial cross-section, while in severe dysplasia: this term is reserved for those cases with atypical or primitive cells extending

into, but not completely through, the upper third of the epithelium (Speight et al, 1996). Severe grades of epithelial dysplasia may merge into the lesion designated as carcinoma in-situ, in which the whole, or almost the whole thickness of the epithelium is involved (WHO, 1978). Some authors prefer to combine severe dysplasia and in-situ-carcinoma under a common name, on the assumption that they demonstrate identical biological behaviours (Crissman et al, 1987; Speight & Morgan, 1993).

Apart from the individual features of atypia themselves, a number of other factors should be taken into account when attempting to grade OED. A major factor is site variation. For example, dysplastic buccal and commissural epithelia often show very deep, bulbous rete processes, containing focal keratinization, and atrophic epithelium (Odell & Morgan, 1998). Dysplastic lesions of floor of mouth and palatopharyngeal epithelia, on the other hand, tend to show shallower, budding processes. A comparable degree of basal cell hyperplasia or suprabasal mitotic activity would therefore involve a higher proportion of the epithelial thickness in the floor of the mouth than in the buccal mucosa. Thus a 'thirds'-based grading system would tend to give greater weight to these features in the floor of the mouth and worsen the recorded grade. In the floor of the mouth, involvement of the salivary excretory ducts in the dysplastic process may also be noted and is significant, usually associated with marked cytological atypia in adjacent surface epithelium (Daley et al, 1996).

As well as site of OED, the pattern of keratinization is a further factor to take into account in attempts to grade dysplasia. Many dysplastic lesions are keratotic, usually having undergone parakeratosis in formerly non-keratinizing sites or hyperkeratosis where they are normally parakeratinized or orthokeratinized and such areas appear white clinically (Odell & Morgan, 1998).

Sometimes, the presence of concurrent disease may influence the scoring of OED. For example, nuclear hyperchromatism and basal cell hyperplasia are an expected accompaniment to Candidal infection but comparable histological features would be regarded with more suspicion in mucosal epithelium in which no Candida is present. As several of the criteria for the histological diagnosis of dysplasia are also features of reactive hyperplasia and the healing response, these and any nearby ulceration should be taken into account when assessing possible premalignant changes.

The grading of dysplastic oral epithelium must take into consideration the degree of cellular atypia, and those lesions with marked alteration are elevated into a higher grade level. Dysplastic epithelium may be atrophic and this presents diagnostic grading dilemma because basilar hyperplasia very quickly extend to the surface. There are no standards for this situation but Speight and co-workers (1996) have recommended that these be regarded as severe dysplasias.

This assessment of dysplastic epithelium is subjective one and until now, it has not been possible to devise a scheme for grading epithelial dysplasia that gives consistent and reproducible results. Abbey and co-workers (1995) tested six board-certified oral pathologists for their consistency of histologic diagnosis of 120 oral biopsies, which ranged from simple hyperkeratosis to severe epithelial dysplasia. Exact agreement (mild or moderate dysplasia) with the sign-out diagnosis was achieved in only 50.5%. Moreover, examiners agreed with their own first assessment on a second showing on only 50.8% of occasions. Agreement with the original diagnosis of the presence or absence of dysplasia was 81.5% and agreement by pathologists with their own diagnosis of dysplasia was 80.3%. Thus in nearly 20% of cases expert pathologists could not even confirm their own earlier diagnosis of dysplasia or its absence.

Various attempts have been made to render objective the diagnostic criteria for mild, moderate and severe epithelial dysplasia and carcinoma-in-situ in oral mucosa. One method employed photographic standards (Katz et al, 1985) for degree of severity of each of the main features. This reduce interobserver variability but the weighting that should be applied to individual features has not been determined and this approach has not found favour in routine diagnosis.

Clinical features of oral epithelial dysplasia

Dysplastic lesions does not have any specific clinical appearance though where erythroplakia is present, dysplasia is likely. Thus small and innocent-looking white patches are as likely to show epithelial dysplasia as are large and irregular ones (Scully & Cawson, 1996). Clinically it may present as leukoplakia, erythroplakia, or erythroleukoplakia (Lumerman et al, 1995). Epithelial dysplasia may also seen in verrucous or papillary leukoplakias or in the margins of chronic mucosal ulcers (see below). OED may also seen in the mucosa adjacent to squamous cell carcinoma (Wright & Shear, 1985; Eliezri et al, 1989). Before discussing the lesions that are typically associated with OED, it is important to detail the aspects of oral premalignancy and precancerous disease.

Premalignant lesions may be defined as a morphologically altered tissue in which malignancy is more likely to develop than in its apparently normal counterpart (World Health Organisation (WHO), 1973).

Although in common use, the term(s) precancerous/ premalignant lesion(s) of the upper aerodigestive tract mucosa may be unsatisfactory because they imply that, perhaps irreversible step(s) along the multistage process of genetic alterations leading to invasive carcinoma have occurred. However, the risk of a lesion becoming malignant within the life span of the patient is highly unpredictable and relatively low.

Furthermore, such lesions may regress. A preferable term *is potentially malignant lesions* and *conditions* (Eveson, 1983) among which there is a greater risk of malignant change. There may be several reasons for believing that a given type of lesion is premalignant. For example, if followed over a sufficiently long period the lesion may be seen to undergo malignant transformation, the lesion may be found more frequently in association with a particular type of neoplasm than can be accounted for by chance, or histological examination may show cytological features similar to those present in frankly malignant neoplasms but without evidence of invasion. Based on WHO guide-lines, this broad group is classified under lesions and conditions, the latter being more generalised and widespread, even systemic. The lesions include leukoplakia and erythroplakia, while the conditions include mucosal atrophy (often associated with iron or multiple nutritional deficiencies), oral submucous fibrosis, erosive lichen planus, chronic immunosuppression, chronic Candidal infection and other rare disorders.

Disorders of the oral mucosa which are suspected as being premalignant can thus be classified as follows:

Premalignant lesions

Leukoplakia (homogeneous and speckled)

Erythroplasia (Erythroplakia) (homogeneous and speckled)

Premalignant conditions

a) Conditions associated with oral mucosal atrophy

b) Oral submucous fibrosis

c) Lichen planus

d) Discoid lupus erythematosus

e) Dyskeratosis congenita

f) Syphilis

Leukoplakia

There is considerable confusion regarding the terminology used to describe the various white patches which can arise in the mouth, in particular the precise meaning of the term leukoplakia - the oral lesion most commonly alleged to have a premalignant potential.

In 1978 WHO defined oral leukoplakia as "A white patch or plaque that cannot be characterised, clinically or histopathologically, as any other disease". The term erythroplakia was used to designate lesions of the oral mucosa that present as bright red patches or plaques that cannot be characterised clinically or pathologically as any other condition (Axell et al, 1984). Lesions with both red and white changes were categorised under the heading of erythroleukoplakia. In 1983 it was suggested to avoid the use of the term leukoplakia in cases of known aetiology other than tobacco (Axell et al, 1984) and as a consequence the definition of leukoplakia was rephrased as "A white patch or plaque that cannot be characterised clinically or histopathologically, as any other disease and which is not associated with any physical or chemical causative agent except the use of tobacco". (Axell et al, 1984) a concept later ratified by others (van der Waal, 1986). The latest internationally-accepted definition is "A predominantly white lesion of the oral mucosa that cannot be characterised as any other definable lesion; some oral leukoplakia will transform into cancer" (Axell et al, 1996).

Oral mucosal leukoplakias have been classified into several morphological subtypes. In most cases leukoplakia forms a homogeneous white plaque which may have a tessellated surface. Occasionally these lesions may become ulcerated. Lesions

may also present as white patches and/or nodules on an erythematous background, this is termed speckled or nodular leukoplakia. Rarely lesions can have verrucous appearance and are then termed verrucous leukoplakia.

Leukoplakia is more frequent than erythroplakia (Bouquot et al, 1988). It has a variable prevalence, one recent study suggested a prevalence of 1.1% in a Cambodia population (Ikeda et al, 1995), and overall the prevalence in various populations studied is from 0.4 to 11.7% (Banoczy, 1984) (Table 1.2). Elderly men may be more commonly affected (Bouquot et al, 1986) than other persons. There have been few studies on the prevalence of leukoplakia in unselected populations.

There is limited data on the incidence of leukoplakia. In a 10-year period in the Bhavnagar district of India there was only one new case of leukoplakia in women (incidence 0.03 per 1000 per year) whereas in men the incidence rate was 2.6 per 1000 per year. In the Ernakulam district, the annual incidence for males and females was 2.1 and 1.5 per 1000 respectively. These variations were related to differences in tobacco habits in the genders in each district.

Most investigations suggest that leukoplakia is predominantly a disorder of middle to late life. Waldron and Shafer (1975) found the majority of cases of leukoplakia in Americans in the 40 to 70 year age group. Women developed leukoplakia slightly later than men. Banoczy and Sugar (1972) also found the peak frequency of oral leukoplakia to be in the 41-70 year old age group of European population. However, in India young adults can be affected. For example, in the Gujarat area of India where bidi (a cheap form of cigarette) smoking is common, the majority of patients with leukoplakia were in the 25 to 45 year old age group (Mehta et al, 1971), and 20% of bidi smokers over the age of 60 were found to have a leukoplakia (Roed-Petersen et al, 1972). The increased frequency of leukoplakia with age probably

reflects the duration of exposure to aetiological agents such as tobacco, possibly modified by non-specific local and systemic age changes.

There are striking differences in the reported gender distribution of persons with a leukoplakia. For example, Mehta and co-workers (1969) reported male to female ratio in Bhavnagar, India, of 82 to 1, whereas in Andhra Pradesh, India, where reverse smoking is common practice among women, there was a preponderance of leukoplakia in women (female to male ratio of 1.4 to 1). Indeed, most of the variations in the gender distributions of leukoplakia in India have been attributed to differences in tobacco habits (Roed-Petersen et al, 1972). Waldron and Shafer (1975) showed an increase in the frequency of leukoplakia in US women between 1961 and 1975, possibly reflecting changes in smoking habits during the period of observation.

There are wide geographical variations in the site distribution of oral leukoplakia, possibly reflecting ethnic differences and differences in habits, particularly tobacco usage. It is difficult to compare studies directly as many use different criteria to designate precise anatomical sites. However, Table 1.3 illustrates the range of variation in a single country (India) and the variation between several countries. In most studies the buccal mucosa and commissures are the most common sites of leukoplakia, especially in India (Mehta et al, 1971; Mehta et al, 1972a; Silverman et al, 1976) and Malaysia (Chin & Lee, 1970). The high frequency of palatal leukoplakia in India arises in those districts in which the habit of reverse smoking is practised. Leukoplakia of the gingiva and alveolar ridges is not common in Europe and is rare in India. The high frequency of leukoplakia of the alveolar ridges and gingiva previously reported in some English miners may, however, be related to tobacco chewing (Tyldesley, 1971).

The floor of the mouth is the least common site for leukoplakias in India (Mehta et al, 1971). Whereas in Europe and Scandinavia (Roed-Petersen & Renstrup, 1969; Pindborg et al, 1972; Banoczy, 1977) and the USA (Waldron & Shafer, 1975) leukoplakia of the floor of the mouth is relatively common.

The frequency of OED in oral leukoplakia shows wide variations. Mehta and co-workers (1971) in India found dysplasia in 8-10% of cases of leukoplakia. Some studies have shown dysplasia in 17 to 25% of biopsies from leukoplakias (Pindborg et al, 1963; Waldron & Shafer, 1975; Bouquot & Gorlin, 1986). In a study of 143 leukoplakias none of the homogeneous leukoplakias were dysplastic but 22% of verrucous leukoplakias and 33% of nodular leukoplakias were invasive carcinomas at presentation (Feller et al, 1991). The frequency of dysplasia appears to be higher in Western countries. For example, Waldron and Shafer (1975) found dysplasia in 17% of leukoplakias in the USA and Banoczy (1977) 24% in Hungary. Some of these differences may reflect differences in patient-selection. Most homogenous leukoplakias have little evidence of dysplastic histological changes or aneuploidy: on the other hand speckled leukoplakias, verrucous leukoplakias and nodular leukoplakias show an increasing frequency of such changes (Sugar & Banoczy,1969; Waldron & Shafer, 1975; Banoczy, 1984; Silverman et al, 1984; Lind, 1987; Hogewind et al, 1989; Gupta et al, 1989; Saito et al, 1995; Onofre et al, 1997). Dysplastic lesions appear more likely to be premalignant: for example, a malignant transformation rate of 16% has been demonstrated in a series of nodular leukoplakias followed up for 8 years in India (Gupta et al, 1989) and a rate of 36% has been recorded in the USA (Silverman et al, 1984). Severe degrees of dysplasia are much more common in speckled than homogeneous leukoplakia (Pindborg et al, 1963; Onofre et al, 1997) and it is the

speckled form of leukoplakia which has the greatest frequency of malignant transformation.

The natural history of oral leukoplakia is partly dependent on the nature of any treatment and the removal or persistence of local irritant factors. Lesions can undergo malignant transformation or change to a more severe type, increase or decrease in size or remain static.

There have been few studies on the regression of leukoplakia in general (as opposed to dysplastic lesions) (Table 1.4) but these have shown that substantial numbers of lesions regress and that regression is far more likely than malignant change.

In a 10-year follow-up study of leukoplakia in India, Gupta and co-workers (1980) showed that in the Ernakulam district 47% of leukoplakias persisted throughout the entire observation period and 42% showed spontaneous regression. Those leukoplakias associated with smoking habits were more likely to persist whereas leukoplakias associated with chewing habits (mainly pan and tobacco) were more likely to regress. Similar findings were reported by Mehta and co-workers (1972b) and Silverman and co-workers (1976). Leukoplakias of the labial mucosa, buccal mucosa and commissures may be more likely to regress than lesions at other sites (Mehta & Pindborg, 1974; Banoczy & Sugar, 1975).

Pindborg and his colleagues (1968a) reported an increase in the size of leukoplakias in only 3.3% of 214 patients over a 10-year period, while Banoczy and Sugar (1975) found that 5.3% of leukoplakias increased in size and 3.7% changed from a homogeneous to a verrucous or speckled form. Some leukoplakias of the tongue and floor of the mouth showed progressive changes but there were no instances of regression despite treatment (Banoczy & Sugar, 1975).

The rate of malignant transformation of OED is considered on pages 64 to 67. This present section details studies of the transformation of leukoplakias *per se*. The reported frequency of malignant change in leukoplakia shows wide variations with intraoral sites and patient population, and has shown a striking overall decline in the last few decades. The rate of malignant transformation reported in the older literature was in excess of 30% (Hobaek, 1946). Studies using large numbers of patients, and in most instances also using the WHO definitions of leukoplakia have shown a much lower rate of malignant transformation (Table 1.5). The reasons for the decline in the rate of malignant transformation are not clear, but may be related to improved diagnosis and awareness of intraoral white patches, more precisely controlled epidemiological studies, the reduced incidence of syphilitic leukoplakia. However, it must be emphasised that a patient with leukoplakia may still have a greater risk of developing oral cancer than the rest of the population (Einhorn & Wersall, 1967). In India, although the risk of individuals with oral leukoplakia developing cancer compared with those who develop cancer *de novo* is lower than in the Swedish study of Einhorn and Wersall, the risk is still appreciable, being 4.8-5.8 times that of an individual without oral leukoplakia for men (Gangadharan & Paymaster, 1971; Silverman et al, 1976). Although in the follow-up studies illustrated in Table 1.5 there were differences in the management of lesions and the duration of follow-up, they illustrate some striking differences in the rate of malignant transformation between various countries and within individual countries. Somewhat surprisingly, the rate of malignant transformation in Western studies (mostly in the 2-6% range) appears to be consistently higher than in India (0.13-2.2%), where oral cancer forms a much higher proportion of total cancers. However, this may partly reflect a patient selection bias. The studies in Western countries have been hospital-based whereas most of the studies in India have

been population-based. Indeed, in the study of hospital-based patients in Bombay by Gangadharan and Paymaster (1971) a rate of malignant transformation of 10% was reported. The rate of malignant transformation may be influenced by clinical presentation, patient age and gender, duration, site of lesions, the degree of OED, and associated social habits (Kramer et al, 1978; Silverman, 1988). Pindborg and co-workers (1968) found that 64% of leukoplakias which subsequently became malignant were clinically speckled. Banoczy (1977) found no cases of malignant transformation in homogeneous leukoplakia but 5.5% and 25.9% of cases developed cancer in verrucous leukoplakia and speckled leukoplakia respectively. Other studies have confirmed these observations (Waldron & Shafer, 1975; Banoczy & Csiba, 1976; Burkhardt & Seifert, 1977; Banoczy, 1984; Silverman et al, 1984; Bouquot & Gorlin, 1986; Lind, 1987; Gupta et al, 1989; Hogewind et al, 1989; Onofre et al, 1997). Homogeneous leukoplakia in the floor of the mouth, however, appears to behave differently from the homogeneous leukoplakia elsewhere in the mouth as discussed later.

The risk of malignant change in leukoplakia increases with the duration of the lesion (Einhorn & Wersall, 1967; Banoczy & Sugar, 1972) and the age of the patient. For example, Einhorn & Wersall (1967) showed that 7.5% of leukoplakia patients over 70 years of age developed carcinomas within 5 years of diagnosis whereas for patients under 50 the risk was less than 1%. Banoczy (1977) found that 2.9% of leukoplakia patients in their 4th decade developed carcinomas, 8.2% of leukoplakia patients in the 8th decade are more developed such lesions.

Moore and Catlin (1967) found that over 75% of cases of oral squamous cell carcinomas developed within an area comprising less than 20% of the total oral mucosa. This area of increased tumour incidence was termed the drainage area since it

consists of the floor of the mouth, ventral lingual mucosa, the lingual sulcus and the retromolar region. It may be postulated that any carcinogens present in the mouth drain to these areas before being swallowed. In a study of asymptomatic oral carcinomas Mashberg (1980) found that 96% of the tumours developed in 'high risk sites', which were the tongue, soft palate complex and floor of the mouth. There is evidence that leukoplakias in the drainage area and in some high risk sites have an appreciably greater risk of malignant transformation. Banoczy (1977) for example, showed that 62.8% of cases of leukoplakia affected the commissures and buccal mucosa but only 19% of carcinomas developed at these sites. On the other hand, whereas only 8.2% of cases of leukoplakia involved the tongue, 37.5% of the oral carcinomas were lingual. An even more striking example of the greater tendency for carcinomas to develop from lingual leukoplakia was reported by Roed-Petersen (1971) who found that nearly 45% of oral carcinomas involved the lateral margin of the tongue but this site accounted for only 1.7% of cases of leukoplakia. Roch-Berry (1981) reported a frequency of 17% of lingual leukoplakias developing malignancy in the same area.

Candidal leukoplakia

Chronic infection of the oral mucous membrane by Candida albicans can give rise to persistent, firm white lesions justifying the clinical title of leukoplakia and appearing clinically similar to other varieties. Candidal leukoplakia cannot be adequately diagnosed by its clinical features alone, but the plaque is usually irregular in thickness and texture, is speckled in character and may affect the commissures.

Candidal leukoplakias may be more prone to malignant transformation than idiopathic leukoplakias (Field et al, 1989). Roed-Petersen and co-workers (1970) found 40% of leukoplakias invaded by Candida showed atypia and that in 67% of leukoplakias showing atypia, Candida was present. Acanthosis of the epithelium can sometimes be

extensive with rounded downgrowths, and there may be variable degrees of epithelial dysplasia (Burkhardt & Siefert, 1977). Renstrup (1970) found no fewer than 61% of speckled leukoplakias showed Candidal invasion and of these, 71% were characterised by epithelial atypia. Only 3% of the homogenous leukoplakia showed Candida and in none was atypia present. Cawson and Binnie (1980) followed-up 30 patients with Candidal leukoplakia and they found that 30% developed carcinoma, whereas only 10% of patients with non-Candidal leukoplakias developed carcinoma. All of the Candidal leukoplakias in which carcinoma developed had a speckled appearance. Rindum and co-workers (1994) reported 47% of leukoplakia biopsies contained fungi. Recently Barrette and co-workers (1998) based on a single PAS-stained sections from each biopsy of 1313 keratosis found that 12.3% of dysplastic keratosis contained fungi.

Idiopathic leukoplakia

No aetiological factor can be identified for the majority of persistent oral white plaques, which are thus termed *idiopathic leukoplakia* (Scully & Cawson, 1996). The histopathology is also highly variable, ranging from hyperkeratosis and hyperplasia to atrophy and severe dysplasia.

Most idiopathic leukoplakias are homogenous leukoplakias and show little evidence of dysplastic histological changes or aneuploidy: in contrast, verrucous leukoplakias and nodular leukoplakias frequently have such changes (Waldron & Shafer, 1975; Banoczy & Csiba, 1976; Burkhardt & Seifert, 1977; Banoczy, 1984).

The most extensive follow-up studies on leukoplakia suggest that this idiopathic leukoplakia has the highest risk of developing cancer. One of the largest studies was based on 782 cases of histologically unspecified oral white lesions followed for an

average of 12 years; of these, 2.4% underwent malignant change in 10 years and almost 5% after 20 years. However, even this low rate represent a risk of malignant change 50 to 100 times that in the normal mouth. It was also conspicuous, that in this very large study the rate of malignant change in oral leukoplakias was 10 times higher in non-smokers than in smokers (Einhorn & Wersall, 1967).

By contrast, 13.2% of 500 cases (Banoczy & Csiba, 1972) and 17.5% of 257 patients with leukoplakia (followed in this case for an average of 8 years) had malignant transformation (Silverman et al, 1984). Again, malignant change was more frequent among non-smokers, and this has since confirmed by others (Lind, 1987; Hogewind et al, 1989).

Proliferative verrucous leukoplakia (PVL)

Proliferative verrucous leukoplakia is a slow-growing but highly aggressive form of oral leukoplakia of unknown cause. First described under that title in 1985 (Hansen et al, 1985), it is a white mucosal plaque which virtually always develops nodular, papillary or verruciform surface projections and which gradually, sometimes rapidly, spreads locally to other sites to become multifocal and proliferative. Four of every five affected patients are female and the mean age at diagnosis is 62 years (range 22-89 years). The usual site of female involvement is the buccal mucosa (63% of cases), while the tongue is most frequently affected in men (82% of cases). Etiologic factors are elusive. Two thirds of patients do not have a tobacco habit, but there are especially strong association with human papilloma virus (89%) (Palefsky et al, 1995). At initial diagnosis, almost half of PVL samples will demonstrate epithelial dysplasia, and few cases, are spared this change eventually. This group of lesions had a high risk for malignant transformation with 70.3% of affected patients developing a squamous cell carcinoma at a PVL site in a mean follow-up time of 7.7 years (Silverman & Gorsky,

1997). Careful clinical and microscopic assessment combined with surgical intervention, close follow-up offer the best approaches to management and control of PVL (Zakrzewska et al, 1996; Silverman & Gorsky, 1997).

Sublingual keratosis (SLK)

The term sublingual keratosis (SLK) means a keratotic patch affecting the floor of the mouth or ventral tongue mucosa. In Europe the term SLK is often used clinically as a diagnostic label for sublingual leukoplakias in recognition of the fact that keratosis at this site have a higher risk of malignant change than at most other oral sites (Kramer et al, 1978; Pogrel, 1979). This might be explained by the fact that the floor of the mouth is a more common site for carcinoma, is exposed to higher carcinogens and is very thin. At one time lesions of this type were thought to be developmental in origin and were termed oral epithelial naevi (Cooke, 1956). However, Pindborg and co-workers (1972) believed that leukoplakias of the floor of the mouth were not developmental and could indeed be premalignant, a view repeated and reinforced by the retrospective study of Kramer and co-workers (1978) which reported that out of a total of 63 patients with such lesions 24 (38.1%) developed oral carcinomas. However, the exact scale of risk is unclear and for many lesions it may be low.

Apart from the high risk of malignant transformation, sublingual keratosis is characterised by extremely benign histological features with virtually no dysplasia until the carcinoma develops. Clinical indications of malignant change in these lesions include development of red areas or ulceration, erythema of the immediately adjacent mucosa and spontaneous discomfort.

Oral leukoplakia, tylosis and oesophageal cancer

This is a rare, genetically transmitted syndrome of tylosis (palmoplantar keratoderma) and oesophageal cancer seen in a group of related patients. By the age of 65, 95% of affected patients have developed oesophageal cancer which is fatal in the large majority. Oral leukoplakia has been described in 16 out of 17 patients with this syndrome (Tyldesley, 1974). Microscopy of the leukoplakia showed parakeratosis or hyperorthokeratosis but no significant epithelial dysplasia. Tyldesley (1974) suggested that oral leukoplakia associated with tylosis should be considered as a positive indicator of oesophageal cancer. However, malignant transformation of oral leukoplakia has not been reported in tylosis.

Erythroplakia

Definition

Erythroplakia describes a bright-red, velvety plaque of the oral mucosa which cannot be characterised clinically or pathologically as being due to any other condition (WHO, 1978). Erythroplakia has clearly demarcated margins of variable size and redness need not always be a prominent feature (Shafer & Waldron, 1975). Erythroplakia of the oral mucosa is rare. In a retrospective study of over 65,000 consecutively accessioned surgical biopsy specimens in two dental schools over 15 and 24 years in the USA, only 0.09 per cent were erythroplakias (Shafer & Waldron, 1975). The prevalence of this lesion among 50,915 villagers in India was reported as 0.02 per cent (Mehta et al, 1971) and among 6000 villagers in Burma as 0.1 per cent (Lay et al, 1982) (Table 1.6).

There is little data for meaningful assessment of the gender and age distribution of erythroplakia. In a series of 58 patients from the USA (Shafer & Waldron, 1975), there was no particular gender predilection and most of the affected individuals were in

the sixth and seventh decades. The floor of the mouth was involved most often among men, the alveolar ridge and the gingiva more frequently among women. In a population-based study of over 50,000 individuals in India, of the nine erythroplakias diagnosed, seven involved the buccal mucosa and two the palate (Mehta et al, 1971).

Premalignant nature

Longitudinal studies to indicate the precise malignant potential of an erythroplakia are lacking. The presence of an erythroplakia, however, generally implies a sinister connotation (Daftary et al, 1992), often being an early sign of a symptomatic oral cancer, especially in heavy smokers and drinkers and at sites liable to oral malignancy 'the floor of the mouth, ventral surface of the tongue, and soft palate'. Mehta and co-workers (1971) found epithelial dysplasia in four out of nine cases of erythroplakia and opined that this lesion represents the most dangerous of the oral premalignant lesions. In Shafer & Waldron's (1975) series, 51 per cent of the erythroplakias were histologically diagnosed as squamous cell carcinoma and 40 per cent revealed carcinoma in-situ or severe epithelial dysplasia.

Potentially malignant conditions

Lichen planus

Lichen planus is a common chronic inflammatory mucocutaneous disease which can give rise to white oral lesions, erosive or ulcerative areas and may have a malignant potential (Fulling, 1973; Krutchkoff et al, 1978; Holmstrup, 1992).

The prevalence of lichen planus among different population varies (Table 1.7) and range between 0.02 in India to 3.8 in Thailand (Axell et al, 1990).

The potential of oral lichen planus for malignant change has long been controversial (Table 1.8). The reported frequency of malignant transformation varies from 0% (Andreasen & Pindborg,1963) to 12.5% (Holmstrup & Pindborg, 1979).

Krutchkoff and co-workers (1978) reviewed most of the reported cases of malignant transformation in oral lichen planus using strict criteria for acceptance. Of 223 reported cases of malignant transformation only 15 fulfilled their criteria. However, their first criterion was that there should be photomicrographic evidence of typical lichen planus. But in many departments, typical lichen planus is not biopsied and those atypical cases that are biopsied frequently show atypical microscopical features.

MacDonald (1975), using figures of the reported incidence of lichen planus and oral cancer calculated that lichen planus was 2 to 10 times more common than oral cancer and that patients with lichen planus had a 10 to 20 times greater risk of developing oral cancer than the general population. It is relevant to study the distribution of tumours in reported cases of malignant transformation in lichen planus. For example, in the cases reported by Andreasen & Pindborg (1963) 88% of the tumours were in the tongue and buccal mucosa, two typical sites of intraoral lichen planus. On the other hand, these sites account for less than 39% of oral carcinomas in general (Moore & Catlin, 1967). Also Cernea and co-workers (1971) reported that 8 out of 66 carcinomas arising from lichen planus were on the dorsum of the tongue, is one of the least common sites for oral carcinomas but is a particularly common site for lichen planus.

One of the largest studies involved 611 Danish patients followed for a mean period of 7.5 years and in this group, malignant change developed in 9 patients (1.5%) (Holmstrup et al, 1988). All the latter, except one, were women and malignant change

followed the initial diagnosis after a mean period of 3.6 years (range 1 to 26 years). Compared with the estimated cancer risk in the population of Denmark, lichen planus would thus appear to increase it approximately 50-fold. Voute and co-workers (1992) however, in their follow-up study of 113 patients, concluded that the three cases of oral cancer found after an average period of 7.8 years provided "some but not very strong support" for the belief that lichen planus was premalignant, because of doubt about the original diagnosis. Apparently anomalous findings are those of Murti and co-workers (1986) who concluded from a study of 702 patients in India, where susceptibility to oral cancer appears to be strong, that the 0.4% incidence of malignant change in oral lichen planus could not confirm its precancerous nature.

Barnard and Co-workers (1993) examined retrospectively the records of a further 241 patients in 3.3% of whom carcinoma or carcinoma-in-situ developed. They also reviewed 22 earlier reports, including those mentioned and, in those where more than 100 patients had been followed-up, the rate of malignant change ranged from 0.4% to 2.7%. Holmstrup (1992) on the basis of 5 previous studies reported since 1985, felt that there was no longer any doubt about the risk of malignant change in some lichen planus and that its magnitude was between 0.5% and 2.5% but was unable to identify any predictive factors. More recent report from Italy (Lo Mozio et al, 1998) conclude from a study of 263 patients followed-up for period of 1 to 10 years 14 (5.3%) transform to SCC.

Oral submucous fibrosis

Oral submucous fibrosis is an insidious and chronic fibroelastic disease of the oral cavity and oropharynx. Established criterion for the diagnosis of oral submucous fibrosis is the presence of fibrous bands in the buccal or labial mucosae and the soft

palate. Early forms of the disease leading to submucous fibrosis are now described and these include the presence of petechiae, vesicles, blanching, extensive loss of normal papillary pattern of the tongue and experiencing a burning sensation (Warnakulasuriya, 1992; Zain et al, 1997). There is inflammation and fibroelastic change, initially in the fibrous connective tissue of the superficial corium. This is followed by epithelial atrophy and formation of bands of scar-like tissue which cause trismus. The fibrosis can extend into the underlying muscles so that as well as trismus, mastication, speech and swallowing may be impaired (Pindborg & Sirsat, 1966b).

There is strong evidence that oral submucous fibrosis is a premalignant condition. There is an increased incidence of leukoplakia in patients with submucous fibrosis. Mehta and co-workers (1971) found a prevalence of 12.7% in patients with submucous fibrosis compared with 2% of an appropriate control group (Table 1.9). Epithelial dysplasia is present in between 13 and 23% of lesions of submucous fibrosis (Paymaster, 1956; Pindborg & Sirsat, 1966b; Pindborg et al, 1972). Pindborg & Zachariah (1965) found 40 cases of submucous fibrosis among 100 patients with oral cancer and Paymaster (1956) described oral cancer developing in one third of his cases of submucous fibrosis. Murti and co-workers (1985) reported the development of oral carcinoma in 7.5% of 66 patients with submucous fibrosis followed-up for a median period of 10 years (Table 1.10).

Sideropenic dysphagia and iron deficiency

Ahlbom in (1936) described the relationship between sideropenic dysphagia and cancer of the upper alimentary tract. He found that many patients with hypopharyngeal and oral tumours, especially women, had sideropenic dysphagia. The study of Wynder and co-workers (1957a) also showed a significant association between cancer of the

mouth, hypopharynx and oesophagus and sideropenic dysphagia in Swedish women. They felt that high incidence of cancer of the upper alimentary tract in Swedish women could be caused by the very high incidence of sideropenic dysphagia. Larsson and co-workers (1975) showed the incidence of sideropenic dysphagia in Sweden was decreasing as a result of better nutrition and health care and this was associated with a decrease in the incidence of hypopharyngeal cancer in women. Prime and co-workers (1983) showed that iron-deficient rats developed experimental oral tumours faster than the normal controls and that iron-deficient animals had significantly more lingual tumours. However, Prime and co-workers (1986) failed to show any differences in the severity of epithelial dysplasia in experimental iron sufficient and deficient rats. It is generally accepted that iron deficiency may be related to loss of epithelial integrity and this may cause the epithelium to be more susceptible to chemical carcinogens.

Syphilitic leukoplakia

Leukoplakia of the dorsum of the tongue once a characteristic complication of tertiary syphilis but is now of little more than historical interest. Syphilitic leukoplakia had no distinctive features, but typically affected the dorsum of the tongue and spared the margins. The lesion had an irregular outline and surface. It is usually regarded as having a high risk of malignant change and cracks, small erosions or nodules may be foci of invasive carcinoma. In the past syphilis was a common cause of leukoplakia (Hobaek, 1946) and was frequently associated with carcinomas of the anterior two-thirds of the tongue and the lip (Wynder et al, 1957b). Banoczy and Sugar (1972) found evidence of syphilis in only 2.5% of patients with leukoplakia but in 10% of patients with carcinomas developing in leukoplakia. Wynder and co-workers (1957b) drew attention to the possible influence of medicaments such as arsenicals (and

possibly heavy metals) used in the treatment of syphilis and the subsequent development of oral cancer.

In addition to hyperkeratosis and acanthosis, often with dysplasia, the characteristic late syphilitic chronic inflammatory changes, with plasma cells predominating, may be seen in the connective tissue. Giant cells and, rarely, more or less well-formed tuberculoid granulomas may be present. Endarteritis of small arteries is particularly characteristic. However, any distinctive features of a syphilitic tissue reaction may be totally lacking (Wynder et al, 1957a).

Discoid lupus erythematosus

Discoid lupus erythematosus (DLE) of the mouth has been suggested to be a potentially malignant disorder. Epithelial dysplasia may occur in DLE and a review (Andreasen, 1964) reported malignant transformation of 0.5%. All cases have been associated with lower lip and tend to be more common in men.

Dyskeratosis congenita

Dyskeratosis congenita is a very rare recessive or dominant disorder comprising oral leukoplakia, nail dystrophy and atrophy and pigmentation of the skin and occasionally aplastic anaemia (Cannell, 1971; Gorlin et al, 1976). Many patients also appear to be immunodeficient or have other abnormalities. Oral lesions develop between the ages of 5 and 10 and usually begin as blisters which rupture to leave painful, slow healing ulcers which typically involve the tongue and buccal mucosa. The oral mucosa becomes atrophic and there is depapillation of the dorsal lingual mucosa. Oral white patches are seen in over 80% of patients or there may be inconspicuous red areas. In the majority of affected patients leukoplakia probably does not start to form until adolescence. During the 3rd and 4th decades areas of erosive

leukoplakia or carcinoma develop (Davidson & Connor, 1988). Multiple oral carcinomas can result and the expectation of life is poor.

Risk factors for clinical lesions associated with oral epithelial dysplasia

Tobacco

Although there is much data linking tobacco smoking and excess alcohol consumption in the aetiology of intra-oral cancer (IARC, 1985) and is summarised in (Tables 1.11 to 1.15). Data concerning the role of these habits in the causation of OED is limited and focused largely upon smokeless tobacco (Mincer et al, 1972; Roed-Petersen et al, 1973a; Johnson, 1991). Only two previous case-control studies assessed aetiological relationship between tobacco and alcohol and OED having been reported (Kulasegaram et al, 1995; Morse et al, 1996). Kulasegaram and co-workers (1995) found a higher risk of OED among current and recent ex-smokers relative to non-smokers and ex-smokers of 10 years or more and in a study involving 127 cases of OED and 127 matched controls from US Morse and co-workers(1996) found tobacco smoking to be an important risk factor for OED. The risk increased with current smoking, and total number of cigarettes smoked per day. But the risk decline following smoking cessation, with ex-smokers of 15 or more years demonstrating no excess risk relative to individuals who had never smoked.

Several studies have emphasised that the usage of tobacco in various forms such as chewing, the oral use of snuff, and smoking cigars, cigarettes, and pipes is an important aetiological factor in the development of leukoplakia (Pindborg et al, 1968a,1972b; Tylesley, 1971; Roed-Petersen and Pindborg, 1973b; Banoczy, 1982; Baric et al, 1982; Lay et al, 1982; Salem et al, 1984; Mani, 1985; Zaridze et al, 1986). The aetiological relationship between tobacco and leukoplakia has been investigated

extensively in several population-based studies in India (Metha et al, 1961, 1971, 1972a; Pindborg et al, 1967; Wahi et al, 1970; Roed-Petersen et al, 1972; Smith et al, 1975; Bhonsle et al, 1976; Gupta et al, 1984a).

The roles of cheroot smoking in leukoplakia of the floor of the mouth among Danish women (Pindborg et al, 1972b) and tobacco chewing among English coal miners (Tyldesley, 1971) were highlighted in early work. In France the epidemiology of "tobacco keratosis" is described as similar to oral cancer in that it is located more or less on the same sites, mainly the tongue and the floor of the mouth (Szpirglas, 1992). Cross-sectional studies provide evidence in the form of prevalence rates of leukoplakia among people with and without tobacco habits as well as the influence of different kinds of habit. For example, in house-to-house cross sectional studies comprising over 158,000 individuals aged 15 years and over in different part of India, 56 per cent used tobacco (Metha et al, 1971, 1972a; Bhonsle et al, 1976). The prevalence of leukoplakia was 1.8 per cent among those who used tobacco and 0.03 per cent among those who did not use tobacco: leukoplakia was diagnosed among 7 per cent of claypipe smokers in Bhavnagar, Gujarat and among 4.5 and 2.5 per cent bidi smokers in Bhavnagar, and Ernakulam, Kerala, respectively (Metha et al, 1971).

The importance of bidi smoking in the development of oral leukoplakia is confirmed in several other studies (Pindborg et al, 1967; Metha et al, 1972a; Roed-Petersen et al, 1972). For example, in a study of 100,000 individuals, 521 leukoplakias were diagnosed among 5.3 per cent of bidi smokers, compared to 1.2 per cent among tobacco chewers and 0.5 per cent among non users of tobacco. A multivariate analysis showed bidi smoking to be the most important associated factor (Roed-Petersen et al, 1972).

With regard to tobacco chewing, leukoplakia was diagnosed among 1.8 per cent of betel-tobacco chewers and among 6.1 per cent of those who chewed betel-tobacco as well as smoked (Metha et al, 1971). In another cross sectional study in north India, Wahi and co-workers (1970) distinguished two types of tobacco used for chewing and found that the prevalence of leukoplakia correlated with tobacco chewing but varied with the type of tobacco chewed. Further, the risk for leukoplakia was estimated to be about 60 times higher in daily chewers as compared to non-chewers. The risk was also found to increase with increase in frequency of chewing, earlier initiation of the habit and duration of the exposure to quid. This analysis, however, was not controlled for smoking. Defining dose as the number of quids chewed or bidis smoked per day, Gupta (1984a) reported a positive dose-response relationship between tobacco habits and the prevalence of leukoplakia. The dose-response relationship was stronger, i.e. the prevalence rates were higher, for the smoking habit than for the chewing habit.

Prospective studies provide stronger support for the aetiological role of tobacco in the form of incidence rates among people with and without tobacco habits as well as the influence of different kinds of tobacco usage. In a 10-year follow-up study of random samples of 30,000 people in three districts, the annual age adjusted incidence rate of leukoplakia was zero among those who did not practise any tobacco habits (Gupta et al, 1980). The incidence in Bhavnagar was 6.7 per 1000 among claypipe smokers and 2.9 per 1000 among bidi smokers. Among those who practised chewing as well as smoking the incidence was only 2.5 per 1000. In contrast, in Ernakulam, the incidence was lower among those who only smoked (0.7 per 1000) or chewed (2.5 among men and 3.0 among women) but among men who both chewed and smoked the incidence was 6 per 1000. The smoking and chewing habits in these two areas, however, are not identical. In Srikakulam, the incidence of leukoplakia (excluding

those on the palate) among men who smoked reverse (keeping the burning end inside the mouth) was 1 per 1000 per annum.

The most convincing evidence for the aetiological role of tobacco comes from intervention studies which demonstrated that leukoplakia regressed significantly more often when tobacco habits were discontinued or reduced compared to when the habits remained unchanged (Mehta et al, 1982). Further, there was a significant fall in the incidence rates of leukoplakia in the intervention cohort where special health education regarding the ill effects of tobacco usage was provided, compared to the incidence in the control cohorts (Gupta et al, 1986).

Differences in smoking and chewing habits can be shown to account for many of the geographical variations in the incidence of leukoplakia and the topographical localisation within the mouth (Pindborg, 1980). For example, in those areas of India where hookli (a short stemmed clay pipe) smoking is common, there is a high incidence of labial leukoplakia (Mehta et al, 1969) but in areas where bidi smoking is prevalent there is a high incidence of leukoplakia of the commissures and buccal mucosa. There is a very strong association between reverse smoking and palatal lesions (Pindborg et al, 1971). Chewing betel with tobacco causes clinical changes (either leukoplakia or a precursor lesion some call pre leukoplakia) in the buccal mucosa in over 63% of Indians (Chin & Lee, 1970). In Denmark, the high incidence of leukoplakia of the floor of the mouth, especially in women, has been associated with cheroot smoking (Pindborg et al, 1972b).

The relationship between tobacco smoking and leukoplakia is emphasised by the work of Roed-Petersen and Pindborg who found that in patients who stopped smoking 60% of leukoplakias totally regressed within a year (Pindborg, 1980). However, the relationship between tobacco, and particularly smoking, and malignant

transformation of leukoplakia is less clear. There are frequently discrepancies between the site of leukoplakias and the site of eventual tumour development. Excessive pipe smoking, for example, causes diffuse palatal keratosis and nicotinic stomatitis but oral carcinomas in pipe smokers tend to develop in the drainage areas of the mouth e.g. the lower retromolar area. Similarly, although 60% of leukoplakias in Hungarian patients group involved the buccal mucosa and commissures, less than 20% of carcinomas developed at these sites (Banoczy, 1977). Also several studies have shown that malignant transformation of leukoplakias is more likely in non-smokers than in smokers (Silverman et al, 1984; Lind, 1987; Hogewind et al, 1989). It is possible that the aetiological factors responsible for leukoplakia formation and subsequent malignant transformation are different.

Smokeless tobacco

Smokeless tobacco (ST) may be a risk factor for the development of OED (Kaugars et al, 1989). The association was demonstrated by the high percentage of cases that developed a premalignant lesion at the site of ST placement. Other investigators discussed the strong association between dysplasia/carcinoma and ST (Winn, 1988). Throughout the world there is a considerable degree of heterogeneity in the method of preparation and constituents of smokeless tobacco (Christen et al, 1982). The two most common types of smokeless tobacco are tobacco chewing and snuff dipping. Tobacco chewing consists of placing a quid, of leaf or plug of tobacco in the gingival buccal area where it is held or chewed. Snuff is usually either of the moist or dry (scotch) variety and is placed in the oral mucosa or administered through the nasal passage.

Smokeless tobacco keratosis (STK) is common occurring in up to 60% of smokeless tobacco users (Poulson et al, 1984; Grady et al, 1990; Sinusas et al, 1992;

Greene et al, 1993). The frequency of STK appearance is dependent on the type of smokeless tobacco used. Moist snuff, which is more alkaline than chewed tobacco, more often leads to STK than tobacco (Greene et al, 1993), but moist snuff in pouches causes less pronounced mucosal changes and fewer cases of STK than loose forms (Anderson & Axell, 1989). The keratosis arises at the site of placement of smokeless tobacco use usually within 6 months to 36 months of initiation of the habit (Greer & Poulson, 1983; Greene et al, 1993). The epithelial changes largely are a response to local irritation (Greer & Poulson, 1983).

Oral epithelial dysplasia is not a common feature of STK, occurring in less than 3% of cases of STK (Smith et al, 1970; Roed-Petersen, 1973; Axell et al, 1976; Bouquot and Schroeder, 1993). Even when dysplasia is present in STK, it is usually found in earlier stages than in oral leukoplakias (Mincer et al, 1972; Kaugars et al, 1989).

Likewise malignant transformation only occurs in up to 1.2% of STK lesions over 5 years (Smith, 1975; Christen et al, 1991), indeed one prospective study found no increase of oral malignancy in 1550 persons with STK followed up for 10 years (Smith, 1975). Another study reported no case of oral cancer among 500 regular ST users followed for 6 years (Christen et al, 1991). This low rate of malignant transformation has also been found experimentally in animals. (Park et al, 1986; Chen, 1989). In comparison, as many as 17% of idiopathic oral leukoplakias will transform to squamous cell carcinoma within 7 years (Silverman et al, 1984).

Smokeless tobacco contains several carcinogens including N-nitrosamines, polycyclic aromatic hydrocarbons and polonium 210 and the use of smokeless tobacco results in 10 to 100 greater exposure to N-nitrosamines than with tobacco smoking and

hence may be the major contributor to the carcinogenic potential of smokeless tobacco (IARC, 1985).

Oral snuff appears to cause more severe clinical changes than does tobacco-chewing (Daniels et al, 1992) and this is probably the main way in which smokeless tobacco is currently used. Dysplasia however, is more likely in tobacco chewers (Kaugars et al, 1991, 1992). The main changes are thickening of the epithelium with plump or squared-off rete ridges. There are varying degrees of hyperorthokeratosis or parakeratosis. Chevron keratosis, though sometimes regarded as characteristic, was seen in only 17% of 132 biopsies examined by Daniels et al (1992). Dysplasia may eventually be seen and occasionally, malignant change can follow, but only after several decades of use (Gradey et al, 1990; Daniels et al, 1992). A high proportion of these tumours are verrucous carcinomas (Winn, 1992) and seen in the buccal mucosa/vestibule (Chakrabarti et al, 1991; Link et al, 1992).

The heaviest users of oral snuff in the Western world are in Sweden, but Larsson and co-workers (1991) in an extensive study, found no carcinomas among Swedish users and stated that they had never been seen in the great number of biopsies from all parts of Sweden over many years. These workers also found that the snuff dipper's lesions resolves on stopping the habit even after 25 years of use.

Alcohol

The role of alcohol *per se* in causing white and red premalignant lesions is yet to be established and so far studies describing an association have been limited (Wilsch et al, 1978; Macigo et al, 1995). In the two previous case-control studies on OED (Kulasegaram et al, 1995; Morse et al, 1996) alcohol consumption was found to be an

important factors in the causation of OED and the risk increased with increased consumption of alcoholic beverages.

An association between alcohol habits and oral leukoplakia has been shown in India (Gupta, 1984) but an underestimation of consumption pattern may have led to the weak association reported.

The contributory role of alcohol in leukoplakia is difficult to evaluate as many people who consume alcohol also use tobacco in some form. Roed-Petersen & Pindborg (1973) found that 17 per cent of Danish patients who had snuff-dipper's leukoplakia consumed alcohol daily. In Germany, Wilsch and co-workers (1978) found that alcohol consumption was greater among people with leukoplakia as compared to controls. In India, Gupta (1984b) studied the alcohol habits of over 7000 tobacco habitués and found that 31 per cent consumed alcohol regularly, 25 per cent occasionally, and 44 per cent did not consume alcohol at all. The prevalence of leukoplakia was higher among regular (5.7 per cent) and occasional (3.9 per cent) alcohol drinkers than among non-drinkers (2.9 per cent). Taking into account the prevalence rates adjusted for age, sex, and the type of tobacco used, the author concluded that alcohol, although by it self not an important risk factor for leukoplakia, may produce synergistic effects when combined with the habit of chewing or smoking tobacco.

Possible mechanisms for alcohol related carcinogenesis

Although ethanol has been administered to laboratory animals using various methods and protocols, there is no evidence that ethanol itself is carcinogenic (IARC, 1988). Results from tests for mutagenicity have also shown that ethanol is not mutagenic unless it is metabolised to acetaldehyde and superoxide (Kato & Nomura, 1994). These ethanol metabolites have been found to be mutagenic and cytotoxic, and

acetaldehyde has been found to be carcinogenic (IARC, 1988). Besides ethanol, alcoholic beverages also contain nitrosamines and other contaminants that have carcinogenic properties (IARC, 1988).

Alcohol dehydrogenase and aldehyde dehydrogenase activity have been demonstrated in the oral cavity (Moreno et al, 1994; Dong et al, 1996). And the activity of aldehyde dehydrogenase is much less than that of the alcohol dehydrogenase (Dong et al, 1996) which suggest that it is possible for the cytotoxic acetaldehyde to accumulate in the oral tissues and may thus be a factor in alcohol related carcinogenesis.

Ethanol alters intracellular metabolism in the liver and other sites, resulting in increased activation of certain carcinogens (McCoy et al, 1979). Liver damage caused by alcohol abuse decreases the metabolic clearance rates of some carcinogenic substances (McCoy et al, 1979). Ethanol may act as a co-carcinogen by solubilising a true carcinogen (McCoy et al, 1979), or may act as a promoter by stimulating cell proliferation (Ishii et al, 1989). It has been postulated that alcohol intake may alter hormonal balance in women in relation to hormone-related cancers (Williams, 1976). An indirect effect of alcohol abuse, i.e. nutrient deficiencies, could be more important. Certain vitamins and minerals have been shown to be anti-carcinogenic and deficiencies of these nutrients are frequently observed among alcoholics (McCoy et al, 1979). In Hawaii, men who subsequently developed oral/pharyngeal cancer had decreased levels of serum cholesterol (Chyou et al, 1992) and haematocrit (Kato et al, 1991), suggesting a suboptimal nutritional state, although these might also be signs of subclinical disease (Kato & Nomura, 1994).

It has been suggested that alcohol may have an effect on DNA repair mechanisms (Hsu et al 1991; Mufti, 1992). And chronic ethanol consumption interferes with the repair of

alkylated DNA (Mufti, 1992). Hsu and co-workers (1991) looked at the effect of pulsed bleomycin with the addition of different concentrations of alcohol in vitro. After a bleomycin pulse with incubation of 0.5% alcohol, the frequencies of chromatid breaks steadily dropped as incubation time increased, but if 2% alcohol was used, the number of chromosome breakages remained high, suggesting that DNA repair was inhibited and this inhibition was reversed when ethanol was removed from the growth medium.

Joint exposure to alcohol and tobacco

Tobacco smoking is causally related to cancer of the upper aerodigestive tract (IARC, 1986), and potentially malignant lesions including OED (Kulasgaram et al, 1995; Morse et al, 1996). Because the consumption of alcohol and tobacco are usually strongly correlated, it is difficult to separate the effect of alcohol from that of smoking (Kato & Nomura, 1994). However, some cohort and case-control studies have found an increased risk of upper aerodigestive tract cancer associated with alcohol drinking in non-smokers (Brownson, 1987; Blot et al, 1988; Franco et al, 1989; Franceschi et al, 1990; Kato et al, 1992). Several studies have shown a synergistic effect of joint exposure to alcohol and smoking (Rothman et al, 1972; Vassallo et al, 1985; Brownson, 1987;Guenel et al, 1988; Blot et al, 1988; Franco et al, 1989; Franceschi et al, 1990; Kato et al, 1992; Morse et al, 1996). The definition of the highest exposure combination of alcohol and smoking varied from study to study, but the risk of each site of cancer among persons who drank heavily and smoked heavily increased more than 7 times compared with the reference group (Rothman, 1972). The results also suggest that the risk increases multiplicatively rather than additively. These epidemiological observations are supported by a recent experimental study that showed

that the overexpression of the p53 gene is more likely to be induced among persons who both drink and smoke heavily (Field et al, 1992).

Betel quid and areca nut chewing

There is considerable epidemiological evidence however, linking betel quid chewing and smoking habits in relation to oral precancerous lesions in South Asia, e.g. Papua and New Guinea (Pindborg et al, 1968b), West Malaysia (Lee & Chin, 1970), India (Mehta et al, 1981), Sri Lanka (Warnakulasuriya & Johnson, 1991) and Thailand (Reichart et al, 1987). Furthermore, a dose-response relationship between tobacco habits, and oral leukoplakia was reported from India (Gupta, 1984). Estimation of the risk of leukoplakia among betel quid chewers with and without tobacco has shown that prevalence of leukoplakia among individuals with no type of tobacco habit is generally very low (Gupta et al, 1982). A case-control study in Sri Lanka confirmed that the addition of tobacco to betel quid raised the relative risk by three-fold (Warnakulasuriya & Johnson, 1991) showing the significant importance of tobacco relative to other ingredients in the betel quid mixture. There is now sufficient evidence to conclude that areca nut in betel quid is important in the causation of oral submucous fibrosis, a high-risk, potentially malignant condition (Canniff et al, 1986; Sinor et al, 1990; Lee, 1992) with a malignant transformation rate higher than leukoplakia (Murti et al, 1985).

Dietary components and deficiency states

Mucosal atrophy is a common suggested feature of the various conditions sometimes considered to increase the liability to oral premalignancy and malignancy of the oral mucosa (Cooke, 1975; MacDonald, 1975). Although several causative factors are linked to atrophy, an important contributory factor is deficiency of micronutrients

such as iron, vitamin A and retinoids. Deficiency of vitamin A is known to induce metaplasia and keratinization of certain epithelial structures (Jafarey & Zaidi, 1976). As keratinization is a marked feature of oral leukoplakia, it has been suggested that vitamin A deficiency may be involved in the pathogenesis. The observation that high doses of vitamin A can cause a remission of leukoplakia favours the role of vitamin A deficiency in the development of leukoplakia (de Vries, 1996). The role of vitamin A deficiency and vegetarian and non-vegetarian diets in the occurrence of leukoplakia have been investigated in two population-based studies. While lower serum vitamin A levels among patients with leukoplakia were observed (Ramaswamy et al, 1996), no association between the dietary differences and leukoplakia was found (Wahi et al, 1970).

Occasional investigations have shown the occurrence of both hyperkeratoses and atrophy in deficiency states of iron (Rennie et al, 1982; Ranasinghe et al, 1983).

Although the mechanism is unknown, vitamin B complex deficiency has also been considered as a predisposing factor in the pathogenesis of leukoplakia (Daftary et al, 1992). Overall, there is, however, little evidence for or against a role for vitamin deficiency and nutritional factors in the development of leukoplakia. Nevertheless there are encouraging reports from animal and clinical studies on the treatment of oral leukoplakia with various vitamin A related compounds such as B-cis-retinoic acid (Shah et al, 1983; Lippman et al, 1993) and Betacarotene (Garewal et al, 1990) as well as vitamin A itself (Stich et al, 1991). Although such compounds represent a potential useful method of intervention in populations at high risk of developing premalignant or malignant lesions presently however toxicity including hepatic damage and positive results obtained in the treatment of premalignant lesions have often not been maintained.

There is increasingly strong evidence from several case-control studies linking possible protective effects particularly of fresh fruits and vegetables in primary prevention of oral cancer (Franceschi et al, 1992; Scully & Boyle, 1992). Results of several other studies also show the efficacy of B-carotene and retinoids in the control of oral leukoplakia (Shah et al, 1983; Garewal et al, 1990; Stich et al, 1991; Toma et al, 1992; de Vries et al, 1996).

Vitamin B12 deficiency can cause nuclear abnormalities in cells other than the hematopoietic classes (Boddington & Spriggs, 1959; Whitehead, 1979; Odell & Morgan, 1998).) Indeed, exfoliated oral squamous cells of patients with pernicious anaemia show nuclear enlargement with occasional giant and binucleate forms (Farrant, 1960; Mitchell et al, 1986). Vitamin B12 deficinecy may produce striking atypia in oral mucosa which could be mistaken for genuine dysplasia (Theaker et al, 1989). Random oral mucosal biopsy specimens of patients with pernicious anaemia show epithelial thinning with basal hyperplasia and increased mitotic activity - although the overall changes do not differ significantly from normal (Jacobs, 1960; Mitchell et al, 1986).

Of notable interest, profound vitamin B12 deficiency can cause moderate-to-severe oral mucosal dysplasia that resolves after correction of the vitamin B12 deficiency (Theaker et al, 1989), during which an increased number of mitosis and suprabasal mitosis may be found. Basal cell hyperplasia and increased numbers of mitosis have also been noted in some untreated lesions.

Syphilis

In the older literature syphilis was regarded as predisposing to the development of leukoplakia, as white patches were often seen on the tongue in the tertiary stage.

Currently, however, with modern treatment facilities, progression of syphilis to the tertiary stage is rare and such white patches are unusual (Daftary et al, 1992).

Candidal infection

The relationship between Candida albicans, leukoplakia and carcinoma is controversial. Jepsen and Winther (1965) in an investigation of oral keratotic lesions frequently found hyphal forms of Candida albicans, especially in cases of speckled leukoplakia. Cawson (1966) and Renstrup (1970) also found that Candidal infection could be associated with leukoplakia, especially the speckled type. Jepsen and Winther (1965) and Pindborg and co-workers (1968a) regarded these lesions as infections superimposed on some pre-existing pathology. However, Cawson and Lehner (1968) coined the term Candidal leukoplakia to describe these lesions in the belief that the organisms were the cause of the leukoplakia.

There is strong evidence of an aetiological role for Candida in proliferative mucosal lesions (Cawson & Binnie, 1980), Candidal hyphae have been shown to be intracellular parasites (Cawson & Rajasingham, 1972) these may be able to alter epithelial behaviour. This view is supported by old work on experimentally induced candidosis in rats (Jones & Russell, 1973), in which it was found that while the Candida were confined to the stratum corneum there was disorganisation and mild atypia in the deeper epithelium and increased vascularity and mononuclear cell infiltration of the underlying corium. In addition, Cawson (1973) showed that proliferative plaques could be produced experimentally on the chick chorioallantoic membrane by Candidal infection.

Speckled leukoplakia can have a considerable risk of malignant change. Renstrup (1970) found that 61% of speckled leukoplakias contained Candidal hyphae

and 71% of these plaques were dysplastic. Banoczy and Sugar (1972) also found a 61% incidence of Candida in cases of speckled leukoplakia and in addition reported that 65% of the cases of leukoplakias which became malignant contained Candida. Cawson and Binnie (1980) showed that 30% of patients with Candidal leukoplakias developed carcinoma whereas only 10% of non-Candida leukoplakias became malignant. All the leukoplakias which became malignant were speckled in type.

An association between iron deficiency and chronic mucocutaneous candidosis is known (Higgs & Wells, 1972) and iron deficiency has also been associated with the development of oral and pharyngeal carcinomas in sideropenic dysphagia (Wynder et al, 1957). It is thus possible that the relationship between chronic hyperplastic candidosis and malignancy may be mediated by an iron deficiency (Cawson & Binnie, 1980) or associated cell defect of cell-mediated immunity (Joynson et al, 1972). Nevertheless, the link between chronic Candidal infection and oral premalignancy and malignancy remains unclear, particularly as very few patients with chronic mucocutaneous candidosis have developed oral SCC (Firth et al, 1997) and Candidal isolates from chronic hyperplastic candidosis do not all produce nitrosamines (Krogh et al, 1987; Krogh, 1990).

Mouth washes

Mouthwash has received attention because it contains ethanol in concentrations that exceed 25%, as well as colouring and flavouring agents. In addition, mouthwash is often held in contact with the oral tissues for prolonged periods of time. Recently Morse and co-workers (1997) based upon 127 case-control pairs investigated the potential association between OED and mouthwash use in USA and concluded that regular mouthwash use are not associated positively with OED risk.

Two cases of white lesions affecting large areas of the oral mucosa were reported in patients who used excessive amounts of the mouthwash Listerine (Bernstein, 1978). This mouthwash contains 25% alcohol as well as other potentially irritating chemicals such as methyl salicylate, benzoic acid and others. The importance of this observation is that there have been reports which suggest that there is an excess of cancers of the throat and mouth in users of commercial mouthwashes many of which also contain 25% alcohol. For example, Weaver and co-workers (1979) found that out of 200 patients with oral squamous cell carcinomas only 11 did not smoke or drink alcohol. Of these, 10 out 11 had used a mouthwash for 20 years or more. This and other studies have shown that the normally low risk group of non-smoking, non-drinking women is most at risk (Blot et al, 1983; Wynder et al, 1983). Alcohol containing mouth washes appear not to be considered significant aetiological agents (Kabat & Wynder, 1989) instead, it is suggested that patients use them to disguise the smell of alcohol. There is a need for more detailed analysis of the possible association of alcohol-based mouthrinses and liability to oral premalignancy and malignancy.

Industrial hazards

It has been suggested that leukoplakias may be more common in rubber plant workers or those exposed to soot dust had leukoplakia. There is little other evidence of such links, and it is also possible that supposed occupational links reflect more obvious social habits such as tobacco and alcohol consumption or variable diet (Pindborg, 1980).

Geography and ethnic origin

Available evidence indicates that social and cultural habits (e.g. among Asian populations) underlay any notable geographic variations in oral premalignancy. Nevertheless, within studied populations, poor socio-economic groups are likely to have a higher incidence of oral cancer (Hirayama, 1966; MacFarlane et al, 1996), although this may be linked to greater tobacco use and under-nourishment among individuals with lower socio-economic status rather than any genuine genetic predisposition to oral mucosal disease.

Human papilloma virus (HPV)

Human papilloma virus (HPV) has been implicated in the aetiology of oral premalignancy and malignancy. At least sixteen HPV DNA genotypes has been isolated from oral lesions (Chang et al, 1991; Miller, 1994). The majority being types associated with benign papillomatous lesions of the oral cavity (e.g. squamous papilloma, condyloma acuminatum, verruca vulgaris, and focal epithelial hyperplasia) with little malignant potential. In contrast, HPV genotypes with increased malignant potential (e.g. 16,18,31,33,35) have been found in epithelial dysplasia and squamous cell carcinoma at several anatomic sites including the oral cavity (Durst et al, 1983; Gissmann et al, 1984; Brandsoma et al, 1986; Gupta et al, 1987; Syrjanen, 1987; Chang et al, 1990; Shillitoe, 1991; Yeudall, 1992; Lakshmi et al, 1993). The role of HPVs in the aetiology of epithelial dysplasia is complicated by the fact that viral genomes can be demonstrated not only in dysplastic tissue, but also in normal oral mucosa (Brandsma & Abramson, 1989; Chang et al, 1989; Jenison et al, 1990; Heyden et al, 1992; Jalal et al, 1992; Kellokoski et al, 1992a; Kellokoski et al, 1992b; Lawton et al, 1992; Ostwald et al, 1994; Mao, 1995) and in benign leukoplakia (Greer et al, 1987;

Gassenmaier & Hornstein, 1988; Greer et al, 1990; Kashima et al, 1990; Shroyer & Greer, 1991; Young et al, 1991; Zeuss et al, 1991).

In a recent literature review (Miller et al, 1996), HPV was detected in 18.5% of OEDs. Viruses were detected more often by polymerase chain reaction (PCR) than immunohistochemical techniques. The mean age for HPV positive lesions was 51.7 years. HPV was more prevalent (2:1) in women than men. The tongue was the most common site of HPV positive OED. HPV genotypes 2,6,11,16,18,31,33,35 were identified in 11.2% of cases of oral dysplasia. HPV 16 and 18 were detected more frequently in HPV positive lesions than HPV 6, 11, 31, 33, 35 (Adler-Storthz et al, 1986; Gassenmaier & Hornstein,1988; Greer et al, 1990; Shroyer et al, 1993).

A link between HPV and leukoplakia *per se* has been suggested; HPV-associated antigens were detected in four of seven leukoplakias and, interestingly, one of the four HPV-associated leukoplakias progressed to cancer (Loning et al, 1984).

Malignant transformation of oral epithelial dysplasia

There is a generally held view that assessment of dysplasia in premalignant lesions is important because dysplastic lesions are more likely to undergo malignant change (Mincer et al, 1972; Banoczy & Csiba, 1976; Pindborg et al, 1977; Gupta et al, 1980; Silverman et al, 1984; Lumerman et al, 1995) and because it is believed that the chances of malignant transformation increase with increasing severity of dysplasia. A direct relationship has been established between grade of epithelial dysplasia and risk of malignant transformation in the cervix uteri (Ostor, 1993) (Table 1.16).

Seven published studies have shown a variable rate of development of oral carcinoma in patients with epithelia dysplasia in various population samples (Table 1.17).

Mincer and co-workers (1972) studied 56 patients who had either moderate or severe dysplasia or carcinoma in-situ. Follow-up information was available in 45

patients. Twenty of these had surgical excision of the dysplasias and seven (35%) showed recurrence. Ten patients (22%) were followed-up without treatment and showed no clinical changes in up to 8 years. Five (11%) of the lesion disappeared or decreased in size without treatment other than a biopsy. Five lesions (11%) increased in size or severity in up to 8 years of follow-up. In five patients, squamous cell carcinoma developed in up to 7 years.

Banoczy and Csiba (1976), in a study of Hungarian patients, reported that 120 (24%) of 500 oral Leukoplakias showed dysplasia. The dysplasias were graded in to three categories of severity on the basis of the numbers of histological dysplastic features evident. Sixty-eight of these patients were followed up, and 9 (13.2%) of these dysplasias progressed to malignancy. 10 (14.7%) were improved, 10 (14.7%) were unchanged, 3 (4.4%) increased in size, and 36 (53%) were cured. The percentages of carcinomas arising from severe, moderate and mild dysplasia, respectively were 56, 33 and 11. In their series, it would appear that all of the instances of malignant transformation arose from cases with dysplasia, as assessed by their criteria, evident on biopsy.

Pindborg and co-workers (1977) followed-up 61 Indian patients with various degrees of epithelial dysplasia. Thirty-three (54%) remained the same. Nine (15%) dysplasias showed spontaneous regression in the form of reduction in the size of clinically evident lesions, and four (6.6%) showed malignant transformation at the end of a 7-years follow-up period. In addition, some of the patients had repeat biopsies, which showed regression in the dysplasia. Gupta and colleagues (1980), in a 10 year follow-up study of dysplastic lesions in India, found that less than 10 percent become malignant, 30 percent worsened, 13 percent regressed and 50 percent showed no change.

Silverman and co-workers (1984) Studied 257 patients in the USA with leukoplakia, of whom 45 progressed to carcinoma in the follow-up period of eight years. Only 22 of these patients had dysplasia in the initial biopsy. This is perhaps a very low proportion, but they do not state their criteria for recording a case as dysplastic, and biopsies that other pathologists would have regard as containing mild dysplasia might not have been designated as such. Of the 45 cases that progressed to malignancy, eight were derived from the 22 dysplasias. This is a 36 per cent incidence of malignant transformation and contrasts with the 15.7% incidence in the cases not showing dysplasia on biopsy. The data from Silverman and co-workers (1984), therefore, support the concept that dysplasia is an important predictor of risk. However, another important point is demonstrated and that is the fact that malignant transformation were also reported among patients diagnosed as not showing dysplasia on biopsy.

Pindborg, Daftary and Metha (1977) also investigated carcinomas developing from non-dysplastic lesions in a rural Indian population. They found malignant transformation after a shorter, average time of follow-up in dysplastic lesions, and this change also occurred in a younger age group.

Vedtofte and co-workers (1987), in a series of 61 Danish patients with premalignant oral lesions, treated 47 patients with epithelial dysplasia by excision and followed them up for an average period of 3.9 years (1 to 5 years). Invasive squamous cell carcinoma developed in three patients during this observation period.

Lumerman and his colleagues (1995) recently studied the clinical features and microscopic slides of 308 cases of OED and retrospectively evaluated 44 of these with follow-up data for transformation to invasive squamous cell carcinoma. Forty four patients had follow-up of more than six months with a mean follow-up time of 18.4

months. Twenty (45%) were clinically free of disease and 15 (34%) had recurrence of the dysplasia. Two cases of the lower grade of the disease recurred as carcinoma-in-situ. Invasive squamous cell carcinoma developed in seven patients (16%) in a mean transformation time of 33.6 months.

The conclusions that can be drawn from such follow-up studies were that dysplastic lesions are more likely to proceed to carcinoma than non-dysplastic lesions, and that the severity of dysplasia may be associated with increased risk of malignant change. However, the grade of dysplasia alone may not a reliable predictor of prognosis (Lind, 1987), carcinoma can develop from an epithelium showing only mild dysplasia or no dysplasia and often dysplastic lesions does not inevitably progress to carcinoma.

Clinical markers of the high risk lesion

Clinical markers of potentially malignant lesions and conditions of the mouth are well established (Johnson et al, 1993). However, the clinical phenotype lacks specificity in terms of actually predicting which of these tissue alterations may subsequently lead to cancer. For potentially malignant lesions, the following are of importance (Johnson et al, 1993).

Site

For example: commissure, in bidi smokers. Floor of mouth, in smokers and drinkers. Lateral border of tongue in smokers and drinkers. Cheeks, lower sulcus and retromolar trigone, in betel nut chewers.

Size

Of itself not significant but wide field changes indicate greater risk.

Shape

Nodular, ulcerated and verrucous lesions carry a greater risk. Non-homogenous type of leukoplakia carry more risk than homogenous. However, when present on the floor of the mouth the homogeneous type, termed sublingual keratosis, is known to carry a high risk of transformation in UK patients (Kramer et al, 1978; Pogrel, 1979). Such a high risk of malignant change, however, has not been widely confirmed.

Shade

Erythroplakia carries a much higher risk of malignant transformation and a large proportion (up to 50%) may be invasive at the time of diagnosis (Mashberg et al, 1973; Shafer & Waldron, 1975). Mixed red and white lesions carry a greater risk than white lesions (Pindborg et al, 1968; Banoczy, 1977).

Surroundings

Widespread oral mucosal atrophy or other lesions may indicate systemic predisposition or field change.

Aspects of treatment of potentially malignant lesions

The need for treatment of oral potentially malignant lesions is primarily based on its nature of being a precancerous lesion (Axell et al, 1994). The rate of malignant transformation varies considerably in different studies - from almost zero up to about 20% in 1-30 years (Silverman et al, 1984; Lind, 1987). Non-homogeneous lesions seem to carry a higher risk for cancer development than homogeneous and idiopathic lesions carry a higher risk than tobacco-associated lesions (Silverman et al, 1984). However, homogeneous and tobacco-associated lesions are by far the most prevalent

forms (Axell, 1987). Also it seems that lesions in some oral sites carry a higher risk, e.g. in the floor of the mouth, on the margins of the tongue and on the lower lip (Pindborg, 1980). The strategy of treatment may partly be based on such epidemiological data.

It is true that a few studies have shown that oral leukoplakia may show complete regression even if no or little change has been registered in factors probably causing the lesions to appear, e.g. tobacco smoking (Silverman et al, 1976), but this fact does not motivate refraining from using active measures in the treatment strategy. The strategy may follow the following procedures:

1. Modification of clinical appearance (and of tobacco habits)
2. Removal of the lesion
3. Follow-up procedures

Modification of clinical appearance

It is imperative to have secured the diagnosis before any treatment is instituted. In most cases a biopsy should be undertaken to rule out the presence of malignancy and to establish the degree of epithelial dysplasia, if present. This is so because it may be decisive for the treatment modality to be chosen but also for permitting an adequate follow-up regimen including evaluating the final outcome related to a recently introduced staging system (Axell et al, 1996).

Homogeneous leukoplakias in the commissure and buccal mucosa are by far the most prevalent lesions (Axell, 1987). The risk for malignant development in these lesions is probably very low (Roosaar et al, 1995). For most of these lesions a tobacco smoking cessation programme should be advocated and a follow-up regimen instituted. Non-homogeneous leukoplakias in the same sites should be checked for Candida. If

present, the yeast infection should be eradicated by using antimycotics. If lesions do not change to be homogeneous within 1-2 weeks, compliance should be checked. At this stage taking a biopsy is compulsory.

Removal of lesion

Surgical excision

All antimycotic resistant non-homogeneous lesions and also homogeneous lesions on the lips, on the margins of the tongue and in the floor of the mouth should be surgically removed, thus following a recommendation to treat all white lesion which does not disappear after elimination of possible aetiological factors (Dunsche et al, 1992; van der Waal et al, 1997).

Surgical excision is the most commonly used technique for removing potentially malignant lesions. Using this method permits securing a specimen for histological evaluation - an important aspect. The surgical wound can be closed by direct suturing, by the transposition of a flap or by using mucosal or skin grafts (Vedtofte et al, 1987). Surgical excision is the first method of choice for localised lesions on the lower lip, on the margins of the tongue and in the floor of the mouth. Alternatively, laser surgery or cryotherapy could be considered.

Authors seem to differ in their opinions on which lesions require surgical excision. Tradati and co-workers (1997) suggest surgical excision of all persistent leukoplakias because of poor patient compliance with follow-up. Other authors (van der Waal et al, 1997) recommend active treatment for lesions showing moderate or severe dysplasia, with oral sub-site being the deciding factor in whether or not to treat mild dysplasia. In the UK, treatment of mild to moderate dysplasia varies, with 16% of oral

and maxillofacial surgeons preferring no active treatment. For severe dysplasia and carcinoma-in-situ, the majority favoured excision (Marley et al, 1996).

Recurrence rates after surgery vary from 20 to 35% (Silverman et al, 1984; Vedtofte et al, 1987). Possible explanations for the comparatively high recurrence rates are difficulties in determining the proper margin of the lesion and pathological epithelium extending into and left in salivary ducts after excision.

Laser surgery

The most frequently used surgical laser is the carbon dioxide (CO_2) laser (other are e.g. argon and Nd YAG lasers (Gaspar, 1992)). It may be used either for excision or for vaporisation of the surface structures. At excision, a specimen is secured for histological evaluation. At vaporisation biopsies should be taken before the start of the laser therapy. Because of the physical properties of laser energy, morbidity is minimal also at healing by secondary intention and epithelial regeneration. This minimises wound contraction and impairment of function due to scar formation. Laser surgery may be the first method of choice when leukoplakia involves large areas of the oral mucosa. Other future possibilities for such lesions may involve chemotherapy and photodynamic therapy.

The recurrence rates after laser surgery vary from 9 to 22% (Chiesa et al, 1990; Roodenburg et al, 1991). As for excision with a cold knife there may be difficulties to identify the margins of the lesion. Also at vaporisation, technical difficulties may arise from judging when all epithelium has been removed even if this is facilitated by using magnification and precise beam control equipment.

Cryotherapy

Closed systems using nitrous oxide (N_2O) or carbon dioxide (CO_2) gas are most suitable for outpatient clinics. Healing is completed without scarring. Disadvantages are considerable. There is no possibility of visibly to control depth of treatment and no specimen is left for microscopic evaluation. Thus, a biopsy should always be taken before freezing. Treatment is often accompanied by considerable postoperative pain and oedema for one to two weeks (Rylen & Axell, 1979) and especially so after treatment on the tongue and in the floor of the mouth.

Recurrence rate after treatment with cryotherapy seems to be similar to what is found after surgical treatment (Sako et al, 1972; Rylen & Axell, 1979; Graham, 1993).

Chemotherapy and chemoprevention of potentially malignant lesions

Chemoprevention is defined as intervention in the process of carcinogenesis with the aim of preventing or delaying the development of cancer (de Vries, 1996). In case of potentially malignant lesions, chemoprevention may serve as a method of treating potentially malignant lesions and preventing malignancy. When leukoplakia is surgically excised about 5% of such patients subsequently develop oral carcinoma (Chiesa et al, 1993). There is thus great interest in chemoprevention in context of oral leukoplakia.

Antioxidants such as vitamin A, retinoids, alfa-tocoferol, beta-carotene and vitamin E have been used for treatment and prevention of recurrences of leukoplakias.

Silverman and co-workers (1963) treated oral leukoplakias with topical application of vitamin A in doses of 300,000-900,000 IU for 1-15 weeks, with response in 43%. Both doses were equally effective. Koch randomised 72 patients in three groups: isotretinoine (13-cis-retinoic-acid, 13-cRA), tretinoine (beta-all-trans retinoic-acid) and

etretinate (Koch, 1978, 1981). Partial response (PR) was found in 59-91% in the three groups, however, the mucocutaneous toxicity was considerable. In other, non-randomised studies comparable responses were obtained with these retinoids (Cardero et al, 1981; Shah et al, 1983). In a blinded phase III study in 44 patients, randomized to placebo or 13-cRA 12 mg/kg/day during six month, the response to 13-cRA in 24 patients was 67% (8% CR, 59 PR) versus 10% PR with placebo (Hong et al, 1986).

Stich and co-workers studied the effects of vitamin A with and without beta-carotene in Indian betel nut chewers (Stich et al, 1988). In 130 patients randomised to placebo, beta-carotene 180 mg/week, with and without vitamin A 100,000 IU/week after six months CR of 3, 15 and 28%, respectively were found. In a later study, patients were randomised between placebo or vitamin A 200,000 IU/week during six months. CR was 57% in the treated group, versus 3% CR in the placebo arm. In another study an overall response of 71% was found in a group of 24 patients with leukoplakias (Garewal et al, 1990). Other workers however reported a response of only 27% in group of 24 patients treated with beta-carotene 90 mg/day for six months (Toma et al, 1990). In a study with 13-cRA 1.5mg/kg/day for three months as patients were randimized to maintenance low-doses 13-cRA (0.5 mg/kg/day) or beta-carotene 30 mg/day (Lipman et al, 1993), the response after induction was 55%. Of 53 evaluable patients, there were 22 (92%) in the 13 cRA group, versus 13 (45%) in the beta-carotene group who reacted in the maintenance phase, or had no progression. Carcinoma-in-situ developed in one patients in each groups; invasive carcinoma developed in five patients in the beta-carotene group (Lippman et al, 1993). Others have used 13 cRA in increasing doses in order to try and circumvent the problem of toxicity (Toma et al, 1992).

Richtsmeier and co-workers (1993) analysed a study conducted by (Lippman et al, 1993) and concluded that for oral leukoplakias associated with dysplasia, isotretinoin appeared to prevent the transformation to invasive carcinoma. Hong and co-workers (1993) have noted that even if some studies have shown retinoids to have significant activity in reversing oral premalignancy, caution must be exercised before drawing overly optimistic conclusions, since (i) reversal of leukoplakia has not been shown to reduce the risk of developing cancer; (ii) the most effective retinoids and optimal dosage schedules for reversing oral premalignancy have not been established; and (iii) surgical or laser resection is already used successfully to manage oral leukoplakias and further studies are needed to determine when chemoprevention should replace or adjoin this established therapy.

Other Agents

N-acetylcysteine (NAC) and 4-HPR, based on in-vitro and animal studies, are very promising chempreventive agents, however they have not been tested as combination in oral leukoplakia. Since NAC is active in earlier stages of carcinogenesis and 4-HPR in later stages, the combination seems attractive.

Fenretidine (4-HPR) is already being used in a chemporevention trial in oral leukoplakia and preliminary results are promising (Chiesa et al, 1992). 4-HPR is active in later stages of carcinogenesis. 4-HPR has proven to be safer and less teratogenic than other retinoids and is effective in preventing chemically-induced tumours in various organs in rodents (Rotsmenz et al, 1991; Chiesa et al, 1993). In Milan, patients were randomised after laser excision to 200 mg fenretinide (4-HPR) or no intervention. End points were recurrence of leukoplakia, new leukoplakia-like lesions and/or squamous cell carcinoma (Chiesa et al, 1992). After intervention for one year, 12 recurrences or

new lesions had developed in 41 patients of the control group while only three of 39 patients in the fenretinide group had reached an endpoint.

Chiesa and co-workers (1993) used fenretinide in patients with negative histology for cancer following laser excision of leukoplakias. Fenretinide was well tolerated and the preliminary results showed a significant protective effect of fenretinide during the year of intervention compared to controls in a randomised study.

N-acetyl-cysteine, an anti-oxidant, is used in a large scale intervention trial (de Vries & DeFlora, 1993) and found to be one of the most promising chemopreventive drugs. The rationale for the choice of NAC is based on a variety of experimental data showing its ability to exert protective effects, such as the extracellular inhibition of mutagenic agents from exogenous and endogenous sources, inhibition of genotoxicity of reactive oxygen species, modulation of metabolism co-ordinated with blocking of reactive metabolites, protection of DNA and nuclear enzymes, prevention of the formation of carcinogen-DNA-adducts, the effect of NAC on the mutagen-induced chromosomal sensitivity assay and its anti-carcinogenicity in experimental animals models (de Vries & DeFlora, 1993).

Based on the available data on chemoprevention, the following conclusions can be drawn. First, chemoprevention is not a definite treatment because after the treatment has stopped, lesions recur in a large number of cases. Second, adverse effects are common with many retinoids, and most are teratogenic. Third, the choice of chemopreventive agent is also important: for example beta-carotene supplement is known to be effective in reversing dysplastic oral lesions (Itri, 1993;Garewal, 1994) and is thought to be well tolerated.

Thus chemoprevention to reverse potentially malignant lesions and to prevent their progression to carcinoma seems to be effective in some studies only, and even then is only temporary. Clearly further studies are needed to determine the most effective drugs and treatment schedules (Boyle et al, 1995).

Other modalities

There are probably other future possibilities for treatment of oral potentially malignant lesions. They are variants of treatment methods for cancer, and includ:

Topical application of bleomycin

Resolution of dysplastic leukoplakia was reported following local injection of bleomycin weekly for 8 weeks (Hisano et al, 1978). Topical application of bleomycin in dimethylsulphoxide (DMSO) has been evaluated in open clinical trials, resulting in regression of leukoplakias (Hammersley et al, 1985; Wong et al, 1989; Epstein et al, 1994).

In a study of 6 patients treated with daily topical application of bleomycin in dimethylsulphoxide (DMSO) for 15-18 days Hammersley and co-workers (1985) reported that the white patch peeled off and the resultant row surface epithelised over the following two weeks. Repeated biopsies showed a significant improvement in histology with reduced epithelial dysplasia and keratinization. In another study, 10 patients were treated with topical bleomycin in DMSO (Malmstrom et al, 1988), epithelial dysplasia was reversed in 5 of 10 patients, but was unchanged in the remaining patients; the thickness of keratotic layer was decreased in 5 patients, increased in 4 patients, and unchanged in 1 patient. Recurrence appeared to be slower than that following surgery. Wong and co-workers (1989) applied bleomycin topically

to 12 patients for 2 weeks using 2 concentrations of bleomycin (0.5% and 1.0%). Partial responses were seen in the less concentrated application, complete remission was seen in 60% of those treated with the higher concentration, and 40% achieved a partial remission.

In a prospective, double-blind, randomised trial of topical bleomycin versus placebo Epstein and co-workers (1994) applied bleomycin 1% in DMSO or the carrier for 5 minutes for 14 consecutive days and reported decrease in clinical size of the lesions and histological reduction in dysplasia.

Recently Epstein and co-workers (1998) applied bleomycin once daily for 14 consecutive days to dysplastic leukoplakias in 19 patients. Complete resolution of dysplastic lesions at follow-up biopsies was seen in 75% of patients, 94% of patients attained at least partial responses after a mean follow-up period of 3.4 years and 31.6% of patients had no clinically visible lesions. Clinically benign lesions of homogenous leukoplakias or minimally visible leukoplakias was seen in 47%. In two patients (11%) malignant transformation occurred.

Advantages of this approach in management of potentially malignant lesions include ease of application that does not require treatment at a medical centre, and a relative low cost compared with surgical intervention. But the technique remain at its experimental stage and more work is needed to test the effectiveness of topical bleomycin treatment in the management of potentially malignant lesions including OED.

Photodynamic therapy (PDT)

Photodynamic therapy (PDT) is a complex interaction of a light sensitising drug and the application of a cold laser light. Light sensitising drugs are selectively retained

by tumours tissue, and when laser light applied, a photochemical reaction is triggered resulting in the release of oxygen-drived free radicals causing cell killing. The PDT did appear to have a potential for the treatment of head and neck malignancies (Wenig & Kurtzman, 1990; Gluckman, 1991), however, little studies has addressed the problems of the precise tissue-effects of PDT in the head and neck region in humans. Early animal work suggests PDT was well tolerated by non-malignant tissue (Meyer et al, 1991). Using the rabbit jaw model it was suggested that there was no selectivity of photodynamic effect, that muscle and salivary gland tissue are sensitive at high doses of PDT, bone relatively resistant and gingival tissue and mucosa quite sensitive to the effect of PDT. In a prospective study Grant and co-workers (1992) attempted to quantify PDT effects with a given light dose and found that PDT effects were characterised by small vessels and microvascular damage, with loss of endothelial cells, intensive inflammatory cellular infiltrate with eosinophilia, preservation of collagen fibres but no evidence of a selective effect. This study also showed that the depth of PDT tissue damage varied from 4mm to 12mm despite the same light dosimetry of 50 J/cm being delivered by an argon ion or a copper vapour-pumped ion laser of 630 nm.

Grant and co-workers (1993) has highlighted the potential for this treatment in "condemned mucosal disease" or multifocal oral tumours but also highlighted the problems of failure to control the underlying pathology which predisposes patients to the development of oral cancer.

Prophyrin-drived PDT leaves patients severely photosensitised such that they have to remain out of the sunlight for over four weeks for fear of developing severe sunburn reactions. This has given rise to research for new effective photosensitisers with human applications, and aminolaevulinic acid (ALA) has emerged as a possible photosenstiser. This metabolised intracellularly to protoporphyrin-IX which exhibits marked

photochemical properties. The short half life of this naturally occurring substance raises the possibility of frequent repeated treatments (Hopper, 1996). Topical ALA has been found to have application in the management of skin tumours (Kennedy et al, 1990), thus this type of PDT may well have application in the treatment of orofacial malignancy.

Protoporphyrin-IX, a metabolite of ALA may have application in the mapping of oral cancer and epithelial dysplasia (Hopper, 1996), as protoporphyrin-IX exhibits fluorescence that can be detected using a charged couple device (CCD) camera.

Photodynamic therapy technique is still in experimental phases when it comes to treatment of potentially malignant lesions and much additional work is however still required to determine the precise role of this treatment modality in the long-term management of patients with potentially malignant lesions.

Follow-up procedures

The risk of malignant transformation is not completely eliminated by any of the above described treatment modalities. Spreading and malignant transformation of the lesion may take place in spite of treatment, while the number of lesions prevented from malignant transformation is unknown (Vedtofte et al, 1987).

No strict guidelines can be given with regard to duration and follow-up examination. In general, long-term follow-up examination is advised at 6-12 months intervals, also in patients who seem to be successfully treated for their leukoplakia. Patients who, after treatment, remain disease free for 3 years need perhaps no longer be followed, but any patient with residual leukoplakia should be followed for a lifetime (Bouquot & Whitaker, 1994).

Aims and Objectives

While only few studies of OED has been detailed, most of these studies were undertaken out side the United Kingdom. In view of paucity of information regarding OED in patients resident in the UK, the aims of this thesis were:

1. To study clinical pattern and natural history of lesions diagnosed as oral epithelial dysplasia.
2. To determine the experience and knowledge of a cohort of UK general dental practitioners regarding relevant aspects of oral potentially malignant lesions, and to assess the quality and delays of referral of patients with potentially malignant lesions.
3. To assess the risk factors for the development of oral epithelial dysplasia.
4. To determine the rate of malignant change of oral epithelial dysplasia and factors likely to influence this malignant change, and to describe the clinical characteristics of patients who developed recurrence of oral epithelial dysplasia and second dysplastic lesions.
5. To detail the clinical features and long-term outcomes of oral epithelial dysplasia among tobacco and alcohol non-users and to compare these to those of users of tobacco and alcohol.

CHAPTER 2

Clinical Characteristics of Oral Epithelial Dysplasia

Outline

Oral epithelial dysplasia (OED) does not have any specific clinical appearance though where erythroplakia is present, dysplasia is likely. Thus small and innocent-looking white patches are as likely to show epithelial dysplasia as are large and irregular ones (Scully & Cawson, 1996). OED may clinically present as leukoplakia, erythroplakia or erythro-leukoplakia (Lumerman et al, 1995), and rarely in verrucous or papillary leukoplakias, or at the margins of chronic mucosal ulcers (see chapter 1). It may also seen in the mucosa adjacent to the tumour in patients with invasive squamous cell carcinoma (SCC) (Wright & Shear, 1985; Eliezri et al, 1989). Most idiopathic leukoplakias are homogenous leukoplakias and show little evidence of dysplastic histological changes or aneuploidy: in contrast erythroplakias, verrucous leukoplakias and nodular leukoplakias frequently have such changes (Waldron & Shafer, 1975; Banoczy & Csiba, 1976; Burkhardt & Seifert, 1977; Banoczy, 1984; Silverman et al, 1984; Bouquot & Gorlin, 1986; Lind, 1987; Gupta et al, 1989; Hogewind et al, 1989).
It has been reported that OED and SCC predominantly arises after the fifth decade of life (Mincer et al, 1972; Kaugars et al, 1988; Hindle et al, 1996), the average age of patients with SCC may be declining (MacFarlane et al, 1994), and the frequency of SCC increasing in young adults for example in the USA (Depue, 1986), Scandinavia (Hakulinen et al, 1986) and Scotland (MacFarlane et al, 1987; 1994). Furthermore, oral cancers in young adults can often be anaplastic (Byers, 1975), findings which are likely to account, in part for the poor prognosis of affected patients (Amsterdam & Strawitz, 1982).

As detailed in chapter one there is little available data of the clinical features of OED in patients resident in the United Kingdom, thus the aims of this chapter were:

1. To determine the clinical pattern and natural history of lesions diagnosed as oral epithelial dysplasia in a large cohort of patients resident in the UK.
2. To correlate the degree of histologically-determined epithelial dysplasia with the long-term outcome in a group of patients with known OED.

Materials and Methods

Data collection

The current analysis incorporates data of 630 patients from two centres in the South of England. Eligible cases included all patients diagnosed as having oral epithelial dysplasia at the relevant hospital between 1972 and 1996 inclusive.

Information recorded in patient case notes were collated in a standardised format (Appendix 1), divided in to five sections. *Section A* included patients details: patient gender, date of birth, country of birth, ethnicity divided in to 9 categories (Bedi & Uppal, 1995) White=1, Black Caribbean=2, Black African=3, Black others=4, Indian= 5, Pakistani=6, Bangladeshi=7, Chinese=8, others=9. *Section B* of the form include date of diagnosis of OED, exact site of OED (topographical code according to ICD-9) (WHO, 1977): lip (140.3, 140.4, 140.5); tongue (141.1, 141.2, 141.3); gingiva (143.0, 143.1, 143.9); floor of mouth (144.0, 144.1); Other and unspecified parts of mouth (145.0, 145.2, 145.3, 145.6). Histology was divided into mild dysplasia, moderate dysplasia, severe dysplasia, and carcinoma-in-situ (WHO, 1978; Lumerman et al, 1995), size of lesion determined in cm, treatment of lesion divided into surgery=1, drug therapy=2, advice to reduce smoking and drinking but no active treatment=3, others=4, site of prior lesion, type of lesion divided into five categories white or predominantly

white patch=1, mixed white and red lesion=2, red or predominantly red lesion=3, ulcer=4, lump=5. *Section C* included data concerning history and referral patterns of OED; duration of lesion in weeks, date of first consultation, type of health specialists consulted divided in to six categories dentists=1, general practitioners=2, hospital department=3, oral medicine=4, oral surgery=5, pharmacist=6. Date of referral, source of referral divided in to four categories dentists=1, general practitioners=2, hospital department=3, self referral =4. Symptoms and signs at first clinical presentation was divided into pain=1, dysartheria=2, weight loss=3, dysphagia=4. Medical history, dental history include teeth missing, teeth decayed, teeth filled, patient has dentists, patient has hygienist, tobacco and alcohol consumption habits. *Section D* of the form included data regarding clinical follow-up of patients. Date of last follow-up, status when last seen divided in to four categories, alive disease free=1, alive with OED=2, dead oral cancer=3, dead other causes=4. Recurrence of OED registered as Yes=1, No=2, date of recurrence, treatment of recurrence divided in to surgery=1, drugs=2, no active treatment=3. Recurrence of lesion was considered when a lesion recurred at the same site after treatment for example, if a dysplastic lesion in an individual was excised from the alveolar ridge and a similar lesion later appeared in the adjacent mucosa, this was interpreted as representing recurrence. Second dysplastic lesion recorded as Yes=1, No=2. Second dysplastic lesion were considered when a new dysplastic lesion developed at a site different from that of the initial lesion. Similarly, patients in whom an invasive carcinoma arose after excision of a dysplastic lesion were considered to show malignant transformation of the initial lesion despite the fact that there were several months during which no lesion was clinically visible. *Section E* include haematological analysis and radiological analysis.

Data was collected and loaded onto computer data base for subsequent statistical analysis,the program used Epi 5 (Dean, 1990) .

Statistical analyses

Descriptive statistical analysis of data were calculated within the computer package Epi 5 (Dean, 1990) and SPSS (Statistical Package for Social Sciences,1993). The chi-square test was used to compare differences between groups, with significance level were taken when the *P* value less than 0.05. Fisher's exact test were used when chi-square test not appropriate or does not yield a sufficient analysis of the data. For example when the overall total of the table is less than 20, or the overall total is between 20 and 40 and the smallest of the four expected numbers is less than 5 (Kirkwood, 1997).

Results

Age distribution of patients with oral epithelial dysplasia

The age distribution of 630 patients with oral epithelial dysplasia is indicated in Figure 2.1. The age range was 13 to 93 years with a mean age of 55.0 years. There were no significant differences in the mean age distribution of patients of each gender or degree of oral dysplasia (Table 2.1). The mean age of patients with mild dysplasia was 54.1 years, moderate dysplasia 55.6 years, severe dysplasia 55.9 year, and carcinoma-in-situ 56.3 years. A slight but not significant increase in mean age was noted when comparing patient with mild OED with carcinoma-in-situ.

Patient gender

Just over 55.0% of the patients with OED were male, and 44.2% were female (Table 2.1). There was a slight male preponderance for mild dysplasia (48.4%), female patients with moderate dysplasia accounted for 2.7% more than male patients, while

male patients with severe dysplasia accounted for 23.1% more than female patients. Likewise there was a male predominance in the group of patients with carcinoma-in-situ (Figure 2.2).

Clinical sites of oral epithelial dysplasia

The oral distribution of the dysplastic lesions according to site is detailed in Figure 2.3. The buccal mucosa (21.1%) and floor of mouth (18.8%) were the most commonly affected sites, followed by lateral border of tongue (15.3%). The gingiva and angle of mouth were the least commonly affected sites.

Distribution of oral epithelial dysplasia according to gender and site of occurrence

The distribution of OED sites in relation to patient gender is shown in Figure 2.4. Of the 11 anatomic sites considered in this study, nine had a male predilection whereas the remaining two (gingiva and commissure) favoured females.

Site distribution of oral epithelial dysplasia according to ethnic-background of patients

The site distribution of OED lesions according to the ethnic-background of affected patients is shown in Figure 2.5. The floor of mouth was the common site of occurrence (20.9 %) followed by the buccal mucosa (17.3%) in Caucasians patients. The buccal mucosa was the most frequently involved site of patients with Bangladeshi, Indian, Pakistan or Afrocaribbean origin.

Distribution of lesion according to site and histological grading

Two hundred and ninety seven (47.1%) of the patients had lesions with mild epithelial dysplasia, 183 (29.0%) had moderate dysplasia, 138 (21.9%) had severe dysplasia with only 12 (1.9%) diagnosed as having carcinoma in-situ Figure 2.6. Severe dysplasia was more likely in lesions from the floor of mouth or lateral border of tongue. Mild

epithelial dysplasia was more likely in lesions of the buccal mucosa. Carcinoma-in-situ more commonly occurred on the floor of mouth (58.3% of such lesions).

Distribution of oral epithelial dysplasia according to ethnic-background of patients

The majority of patients with OED were Caucasians (69.6%); the remainder were of Indian (7.4%), Pakistani, Bangladeshi, or Afrocaribbean origin, or belonged to another racial group (Table 2.2).

Distribution of clinical type of oral epithelial dysplasia and patient ethnic-background

Just under 50.0% of Caucasian patients had OED manifested as a white patch and 1.4% as a red patch, 46.0% as mixed (white and red) lesions (Table 2.2). In contrast 41.8 % of the lesions of Indian patients were white patches, 48.8% had mixed white and red lesions. 38.4% of lesions in Pakistani and 67.3% in Bangladeshi patients were described as white patches. 39.2% of lesions in Afrocaribbeans were white patches, 57.1% were mixed lesions, 3.5% red patches.

Distribution according to clinical type of the lesions

Three hundred and nineteen (50.6%) of the OED lesions were described as white or predominantly white by the attending clinicians; 44.1% mixed red and white lesions, 1.9% as red or predominantly red lesions and only 2.8% lesions, were described as ulcers (Table 2.3).

Distribution according to clinical type of lesions and histology

The majority of white patches (43.8%) were found to have mild OED, although 30% and 24.7% had evidence of moderate to severe OED respectively (Table 2.3). The majority of mixed red and white lesions (49.6%) were mildly dysplastic, but again

almost half had evidence of moderate or severe dysplasia. Red lesions were prone to have mild to moderate dysplasia.

Distribution according to clinical presentation of the lesions and patient gender

There were no significant differences between the two genders with regard to clinical type of lesions. Of note however, 8 of the reported 12 red patches occurred in females (Figure 2.7).

Duration of clinical lesions of oral epithelial dysplasia prior to diagnosis

The duration of clinical lesions of OED and patient gender is detailed in Figure 2.8. The majority of females (68.4%) and 31.5% of males self-reported the duration of the lesions to be less than twenty weeks prior to presentation in the oral medicine clinic, 71.4% of males and 28.5% of females had a delay of their presentation to health specialists between 84 and 104 weeks, while in 43.7% of males and 56.2% of females reported duration's of more than 146 weeks.

Distribution according to methods of treatment of dysplastic lesions

Adequate information regarding treatment of OED was available for 427 patients. Surgery was the most preferred method of treatment of OED lesions 63.9% (273/427). Antifungal therapy accounted for 16.6%, whilst giving patients advice to reduce smoking and drinking without any active treatment accounted for 19.4% (Figure 2.9).

Comparison of patients below and above 35 years of age

There were 59 patients who were 35 years of age or younger. Their data was compared with those patients older than 35 years of age (Table 2.4). The data for cases from the lateral, ventral, dorsal border of tongue, were combined. Likewise, the data for the

gingiva, alveolar ridge, and edentulous alveolar ridge of the maxilla and mandible were combined. In the 59 younger patients (<35 years of age), females were slightly more commonly affected than males; and there were no significant differences in the ethnic-background of the younger patients compared with older patients. The younger age group (<35 years of age) were more likely to have mild OED compared to patients older than 35 years of age. Analysis of the site of lesions, clinical type of lesions revealed no significant differences between the two groups. Of note however, all carcinoma-in-situ cases were among patients older than 35 years of age. Follow-up of these two groups revealed 5.0% of the younger group transformed to SCC in contrast to 5.6% of older group but this difference was not significant.

Discussion

The difficulty in correctly diagnosing and classifying dysplastic lesions has been emphasised by others (Pindborg et al, 1985; Abbey et al, 1995; Karabulut et al, 1995), and is an inherent problem of any study of OED. The histologic grading is partially subjective since the pathologist must make a decision which often is based on his or her individual past training and experience. In addition, reactive epithelial changes, which histologically mimic dysplasia, commonly occur in lesions such as papillomas, lichen planus, and denture hyperplasia (MacDonald & Rennie, 1975). The validity and uniformity of the diagnostic criteria in the current study were strengthened by availability of single group of pathologists throughout the study period. Also, the large number of cases minimised the effect that any incorrectly diagnosed ones might have had on the overall data.

Comparing the findings of the present study with previous reviews (Table 2.5) reveals many interesting points. In the present study (47.1%) were diagnosed as mild

dysplasia and that 21.9% were considered severe. This is in contrast to another European study of 120 dysplasias patients which classified 60.0% as moderate and 19.5% as severe (Banoczy & Csiba, 1976). Using the standardised Smith-Pindborg method, Katz and co-workers (1985) found 41% mild, 29% moderate, and 30% severe epithelial dysplasias in their 207 cases. In a larger study Kaugars and colleagues (1988) reported 54.1% of the lesions were diagnosed as focal mild or mild dysplasia and only 8.1% were considered severe dysplastic. Some of these differences may reflect differences in patient-selection or different interpretation of the dysplastic lesions by different pathologists.

If the mild dysplasia category are the first stage in dysplasia, then it could be assumed that the moderate and severe categories would be diagnosed at progressively older ages. One explanation is that a severe epithelial dysplasia becomes symptomatic sooner and, therefore, prompts the patient to seek care at an earlier time. Because of the closeness in mean ages between the four groups of dysplasia, it is not possible, from the present data to render any definitive statement regarding the temporal progression of epithelial dysplasia.

The peak distribution in the 6th decade among both genders accord with other studies (Banoczy & Csiba, 1976; Katz et al, 1985; Kaugars et al, 1988). Mincer and co-workers (1972) found that severe epithelial dysplasia occurs in an older age group, (60-80 year old) than patients with moderate dysplasia. In the current study there was a higher proportion of carcinoma-in-situ in patients in their 6th decades than mild dysplasia, but the difference was not statistically significant.

The male to female ratio in this study was 1.25:1 which is considerably lower than the ratio reported by Katz and co-workers (1985). The ratio varied considerably in different

sites, but there was a male preponderance in mild, severe and carcinoma-in-situ. Female patients with moderate dysplasia accounted for 2.7% more than male patients. In the sample of carcinoma-in-situ reported by Shafer (1980) more females than males showed involvement of the tongue, buccal and alveolar mucosa.

The present findings of the tendency for white males to be diagnosed at an older mean age than black and Asians males, and for black females to be older than white and Asian females is hard to explain. One theory might be that white males and black females seek professional help at a later stage in the development of the lesion, however this seems very unlikely.

The overall figure of 55.7% male patients is similar to the 54.6% in another dysplasia study (Kaugars et al, 1988), and the 54.2% found in the leukoplakia research done by Waldron & Shafer (1975). Of their 3256 cases, 16.7% were epithelial dysplasia or carcinoma-in-situ.

A previous dysplasia study (Pindborg et al, 1972) demonstrated a high incidence on the palate (50.8%) and buccal mucosa (42.6%) which was due to unusual smoking habits among their patient sample from India. Others have reported that either the tongue, commissure, or lower lip is the most common site (Roed-Petersen, 1971; Mincer et al, 1972; Banoczy & Sugar, 1972; Katz et al, 1985). In the present study, the buccal mucosa was clearly the most frequent site of OED. This is similar to the observation made by Kaugars and colleagues (1988). The supposed high risk sites 'the floor of mouth and ventral surface of tongue' accounted for 26.3% (166/630) of the cases in this study. There was also high frequency of severe dysplasia and carcinoma-in-situ in these sites. This correlates with the observation by Kramer, El-Laban & Lee (1978) and Mashberg (1980) that the floor of mouth is a high risk site for the progression of premalignant to malignant lesions.

Red (erythroplakia) lesions have a greater likelihood of showing a moderate dysplasia than leukoplakia, but leukoplakia demonstrated high rates of severe dysplasia compared with erythroleukoplakia (mixed white and red) lesions (Table 2.3). This is in sharp contrast to other studies which demonstrated that leukoplakias which are speckled or erythroplakic are usually dysplastic or frank carcinomas. In a study of 143 leukoplakias none of the homogenous leukoplakias were dysplastic but 22% of verrucous leukoplakias and 33% of nodular leukoplakias were invasive carcinoma at presentation (Feller et al, 1991).

In the present study the buccal mucosa, floor of mouth and lateral border of tongue were more frequently affected in males and females than any other sites, while, gingiva, angle of mouth were more associated with females. This could be related to different habits between genders. Kaugars and co-workers (1988) in their study found lower lip and commissure to be the most frequent sites associated with males. The lower lip involvement as result of the greater likelihood that a male would be exposed to ultraviolet radiation. While commissure involvement was explained as the lower lip involved the contiguous commissure and were interpreted by the clinician as arising from the commissure.

Analysis by anatomic site (Figure 2.6) showed that the "high risk" sites for development of severe dysplasia are the floor of mouth and lateral surface of the tongue, this is in agreement with another study (Katz et al, 1985) but in contrast to other studies (Banoczy & Csiba, 1976; Kaugars et al, 1988) who found the 'high risk' sites for severe dysplasia to be ventral surface of tongue, lower lip, upper lip. The gingival lesions, which accounted for 10.4% (66/630) of all lesions, appeared to be at relatively low risk since 50.0% of these were mild dysplasia. The "low risk" sites are in essential agreement with other studies (Banoczy & Csiba, 1976; Katz et al, 1985;

Kaugars et al, 1988). The high risk site for carcinoma-in-situ was floor of mouth. This is in line with other studies (Banoczy & Csiba, 1976; Amagasa et al, 1985).

It has been shown that carcinomas in younger people are more aggressive (Son & Kapp, 1985) but the current study did not substantiate this for OED. A predilection for females and an unexplained preference for the tongue were noted. The small number of cases in patients 35 years of age or younger (9.3%) is consistent with other studies (Kaugars et al, 1988). Explaining the possible aetiological factors in the development of OED in younger people is difficult. Some authors related the observed increase in mortality from tongue cancer in young males to the increasing use of snuff (Shemen et al, 1984; Depue, 1986) but this habit is not reported among this group of patients. And the recent increase in oral cancer reported among a younger age group than traditionally recognised (under 40 years of age) (MacFarlane et al, 1992) parallels the increased consumption of alcohol in Britain (from 109 litres/person in 1960 to 142 litres/person in 1981) (IARC, 1988). Creath and co-workers (1991) evaluated the prevalence of oral leukoplakia among adolescent smokeless tobacco users and found oral leukoplakia was six times more likely to develop than in non-users. OED is predominantly a disease of men in the sixth to eight decades of life (Mincer et al, 1972; Kaugars et al, 1988). Even though OED is generally more common in males, it is interesting that in this series there was a slight predominance of female patients in the younger age group. Others have reported similar findings with regard to SCC in the younger people (McGreger et al, 1983; Son & Kapp, 1985; Kuriakose et al, 1992;Sarkaria & Harari, 1994).

Histopathological grading of OED in this study showed that the younger age group (<35 years) were more likely to have mild OED compared with older age group (>35years) and could be explained as the mild OED are the first stage in dysplasia while the moderate and severe categories be diagnosed at progressing older age. The

majority of the dysplastic lesions were mild. Odukoya and co-workers (1985) believed that mild epithelial dysplasia had only a slight chance of malignant transformation. But others have suggested that cancer in the adults tend to be more frequently anaplastic resulting in a more aggressive behaviour and poor prognosis (Byer, 1975). Analysis of the clinical type of the lesions in both age groups revealed no significant difference; white patch (leukoplakia) were the most common lesion. Of note however, all but one of the red lesions were found among older age group.

Assessing the response of the OED to treatment showed that in patients < 35 years of age, OED recurred in 12.8% after treatment. While in patients > 35 years of age, 18.1% of patients showed recurrence. However, 5.0% of the younger and 5.6% of the older age group were transformed to malignancy at the end of the follow-up period range between two to 108 months mean 55 months.

It appears prudent to excise all oral dysplastic lesions which are greater than mild in severity and to be cautious of any lesion in a "high risk" anatomic site as even small dysplastic lesions can be followed by multiple carcinomas and a fatal outcome, as confirmed by the report of Shibuya and co-workers (1986) of 522 cases of carcinoma or carcinoma-in-situ (severe dysplasias) of the tongue. They found that the risk of multiple carcinomas was five times greater with carcinoma preceded by leukoplakia than those carcinomas which had no such precursor. This large study, therefore, appears to confirm the possibility that some dysplastic leukoplakia may have a worse prognosis than isolated carcinomas without leukoplakia.

It is concluded that the peak frequency for occurrence of OED is the 6th decade, a predilection for males was confirmed. The tongue, buccal mucosa and floor of mouth

are the most common sites of involvement. Most OED is mild; carcinoma-in-situ is rare, the sites likely to have areas of severe dysplasia are the floor of mouth and lateral border of tongue.

CHAPTER 3

UK Dental Practitioners Experience, Knowledge And Methods of Referral of Oral Potentially Malignant Lesions

Outline

As outlined previously epidemiological studies have shown that cancers of the oral cavity and pharynx (excluding cancers of the lip, salivary gland, nasopharynx) are becoming more common world-wide (MacFarlane et al, 1994). Mortality rates are increasing in all parts of Europe (but particularly in central and eastern Europe) and in Australia. Examination of incidence rates have confirmed that this disease is becoming more common in most European countries, particularly in cohorts born after around 1915 (Moller, 1989; MacFarlane et al, 1992; Plesko et al, 1994; MacFarlane et al, 1996). And there may be an increasing frequency of oral SCC in young adult men and perhaps women (Hindle et al, 1996). The precise reasons for the epidemiological changes remain unknown but may reflect changes in tobacco and alcohol habits and aspects of social deprivation (Moller, 1989; Bundgaard et al, 1995; MacFarlane et al, 1996).

As detailed in chapters one and two some patients with oral malignancy will have had preceding oral premalignant disorders with histopathologic evidence of OED (Scully & Cawson, 1996), thus early diagnosis of lesions with OED could result in a reduction of the oral malignancy associated with the subsequent transformation of OED to squamous cell carcinoma (Pindborg et al, 1977; Gupta et al, 1980; Silverman et al, 1984; Lumerman et al, 1995).

Early diagnosis of oral and oropharyngeal malignancy is important to ensure maximal prognosis (Silverman, 1990; Langdon, 1995). Effective diagnosis requires appropriate communication and referral between primary and secondary health care workers, but there can be significant delay in the referral of patients with oral SCC to appropriate specialists by general dental and medical practitioners in the UK and elsewhere (Cooke & Tapper-Jones, 1977; Amsel et al, 1983; Scully et al, 1986; Guggenheimer et al 1989;

Jovanovic et al, 1992; Schnetler, 1992; Gorsky & Dayan, 1995). Furthermore potentially general dental practitioners may play important role in any planned preventative or screening programmes of potentially malignant and malignant oral disease. It would be expected that primary health care staff such as general dental practitioners or general medical practitioners provide definite help for patients with OED. Nevertheless it would seem reasonable for them to have some knowledge of the clinical, pathological and risk factors associated with OED and related disorders to ensure effective referral of affected patients to the secondary health care workers also to initiate effective preventive measures in patients with known or presumed OED or malignancy.

There is however little information on the knowledge and attitudes of general dental practitioners regarding their appropriate management of patients with potentially malignant lesions, likewise there is no data on the knowledge of primary health care workers concerning related aspects of the aetiology and clinical presentation of OED. There is currently little data on the effectiveness of letter of referrals of patients with potentially malignant disorders such as OED to appropriate secondary health care workers (Zakrzewska, 1995). In view of this paucity of information the aims of this chapter are:

1. To assess the delays in the referral of patients with oral epithelial dysplasia from primary health care staff to secondary health care providers in the UK.
2. To determine the experience and knowledge of a cohort of UK general dental practitioners regarding relevant aspects of oral potentially malignant lesions and SCC.

3. To assess the quality of referral of patients with potentially malignant disorders of the oral cavity.

Materials and Methods

Assessment of quality of referral of patients with potentially malignant lesions

The contents and the quality of the referral letters of 410 adult patients with OED who attended an Oral Medicine Unit in the United Kingdom (UK) over the period 1983-1993 were reviewed. Patient's age and gender, medical history, source of referral, diagnosis of the referring practitioners, degree of urgency expressed in the patient's referral letter, time lapse before the patient seek advice, delay in referral by the practitioners, and hospital delay in diagnosis and management. The delay by the patients was estimated as the period between the day when the patient first admitted being aware of symptoms until the first consultation with dental or medical practitioners. The delay by the practitioners were defined as the time from the day of the first consultation until the date of the referral letter to the specialists clinic. The delay by the hospital was estimated as the period between the date of referral letter and the first visit of the patient to the clinic, from then until the day of histological confirmation of the diagnosis, and from then until the first visit to the operator carrying out treatment.

Details provided by the referring clinicians were compared with a list of the information that might be expected to be included in an ideal referral letter (Table 3.1). In addition, the suggested diagnosis of the referring clinicians was compared with the definitive diagnosis made by the hospital clinicians.

Assessment of experience, knowledge and clinical practice of general dental practitioners

To assess the knowledge, opinions and clinical practice of general dental practitioners (GDPs) regarding aspects of potentially malignant lesions a self-administered questionnaire (SAQ) was constructed (Appendix 2) and posted to 600 UK dental surgeons resident in England, selected randomly from the General Dental Council (GDC) register. Randomisation was undertaken by numbering all the members in the GDC register list, then randomly choosing a number between 1 to 100 to form the starting point. From the list the chosen number was 15 therefore the sample was selected by taking the 15th member of the list on every page. This is known as *systematic random sampling* (Campbell & Machin, 1994).

The questionnaire had been tested by 30 GDPs attending continuing education programmes in Eastman Dental Institute for Oral Health Care Sciences. The questionnaire included demographic variables of the responding practitioners, such as age, gender, professional qualifications, year and centre of qualification, and postgraduate qualifications. Knowledge variables included the knowledge of the dentist's clinical features of potentially malignant and malignant oral lesions and their relevant undergraduate and postgraduate experience of managing patients with potentially malignant and malignant oral lesions. Dentists were questioned on their opinions of the need for and usefulness of screening programmes, their methods of referral of patients to specialists clinics and the methods they employed to motivate patients to reduce their risks of oral malignancy. Statistical analysis was carried out using chi-square test (Swinscon, 1996).

Results

Assessment of quality of referral of patients with potentially malignant lesions

Source of referral

One hundred and ninety five (47.5%) of the patients were referred by general dental practitioners, 158 (38.5%) by general medical practitioners, hospital departments referred 42 (10.2%) patients, and 15 (3.6%) patients referred themselves to the unit.

Patient address, date of birth, gender, contact telephone numbers, occupation, ethnic origin, investigations, main patient complaint.

Three hundred and forty nine (85.1%) of the referral letters detailed the patient address, and the patient gender included in 343 (83.6) of the referral letters. The contact telephone numbers of only 57 (13.9%) patients were provided. The patients main complaint was given in 391 letters (95.3%). However, details of any investigations undertaken prior to referral were included in only 24 (5.8%) (Figure 3.1).

Degree of urgency

Only thirty two (7.8%) of the referral letters indicated that a patient should be seen urgently; 13 (3.1%) of the referring clinicians expected a routine appointment and 112 (27.3%) an "early" appointment. However, two hundred and fifty three (61.7%) referral letters did not mention any degree of urgency (Figure 3.2).

Medical history

Only 29% of the referral letters provided details of the patient's medical history. In comparison with the data obtained from the patient at their initial consultation in the Oral Medicine clinic, many relevant aspects of the patient medical history had been omitted by the referring practitioners (Table 3.2). Cardiovascular, endocrine and

musculoskeletal disease were the most common groups of disorders to be omitted from the letter of referral.

Patient social history

Only one hundred and seventy seven (43.2%) of the letters contained details of the patient's tobacco habit; details of alcohol consumption were only given in 90 (21.9%) of letters. Other habits such as tobacco chewing, or use of betel-tobacco were included in 18 (4.3%), but 36 (8.7%) of the referral letters did not include any details of the patient's personal habits. The ethnic origin of only 43 (10.4%) patients were recorded, while the occupation of only 46 (11.2%) were provided in the referral letters (Figure 3.3).

Clinical problem

All referral letters provided some details of the presenting complaint for which the patient required referral to an Oral Medicine clinic, but precise details were often lacking. For example, in only 386 (94.1%) of letters was the site of the lesion stated, and the size of the lesion was only recorded in 137 (33.4%) of letters, type of the lesion in 263 (64.1%), while the colour and duration of the lesion was only stated in 265 (64.6%) and 221 (53.9%) of letters respectively (Figure 3.4).

Diagnosis of the referring practitioners

About 50% of general dental practitioners did identify patients with premalignant lesions as opposed to only 15.9% of general medical practitioners. In contrast, only 20% of referring general dental practitioners and 42.1% of general medical practitioners accurately determined that the patient referred to the unit had an oral malignancy (Figure 3.5).

Patient's delays

The patients delay varied from one day to 101 days with a mean of 50.5 days. 10.6% (13/122) of patients with intra-oral lesions and 4.7% (1/21) of labial lesions sought medical advice within one to twenty-two days. However, 65.5% (80/122) of patients with intra-oral lesions and 47.6% (10/21) of patients with labial lesions presented to their clinicians within two months. Only 7.6% (11/143) of patients delayed their presentation for more than three months (Figure 3.6).

Practitioner's delay

The majority of GDPs 47.2% (35/74) and GMPs 49.1% (28/57) referred patients with suspicious oral lesions less than one months for specialists opinion. Only 1.3% (1/74) of GDPs and 1.7% (1/57) of GMPs delay patient referral for more than 12 months (Figure 3.7). The mean delay by GDPs was 80 days and by GMPs 73 days.

Hospital delays

Twenty patients commenced treatment on the same day as they had their confirmatory diagnosis. But the majority of intra-oral lesions 22.3% (36/161) were treated after 4 to 8 weeks. However, 42.8% (9/21) of labial lesions were treated within 3 weeks. The mean hospital delays was 39 days range (1 day to 5 months) (Figure 3.8).

Assessment of experience, knowledge and clinical practice of general dental practitioners

Demographics of responding UK dentists

Just over 30% of those GDPs circulated responded to the questionnaire (30.3% response rate). Of the responding dentists one hundred and twenty seven (69.7%) were male and the median age of the dentists was 40 years (range 24 to 75 years). 59.8% of

respondents had a Bachelor of Dental Surgery although a further 7.1% had a Licentiate in Dental Surgery of one of the Royal Colleges of Surgeons (Table 3.3).

Postgraduate education

Only 56 (30.7%) of the respondents had additional postgraduate qualifications, (Table 3.3), this is most commonly being a Fellowship in Dental Surgery (FDS) of one of the Royal Colleges of Surgeons. In contrast, almost all respondents attended postgraduate meetings, usually British Dental Association (BDA) or Department of Health-funded meetings. Eighty two respondents (45.0%) had attended specific courses on potentially malignant or malignant oral lesions. Respondents otherwise seemed to obtain professional information from reading dental journals (particularly the British Dental Journal) although up to 71.1% used Department of Health-funded video tapes. Few practitioners regularly used the facilities of a local postgraduate medical or dental centre.

Undergraduate experience of oral malignancy and premalignancy

During their undergraduate dental studies 31.2% (40/128) of the dentists witnessed less than 10 patients with oral malignancy; 68.7%(88/128) saw more than 10 affected patients during this period. Squamous cell carcinoma was the most frequently observed oral tumour 56.2% (72/128) (Table 3.4)

Thirteen per cent (22/159) of the respondents had examined or witnessed less than 10 cases of potentially malignant oral mucosal lesions during their undergraduate studies. Leukoplakias were the most commonly observed (50.3%) potentially malignant lesion.

Postgraduate experience of oral premalignancy

Up to 84% (108/128) of practitioners reported that they saw up to 10 patients per year with oral mucosal white lesions. Perhaps surprisingly up to 87.1% and 96.5%

suggested that they had examined 1 to 10 patients with red or speckled oral mucosal lesions (Figure 3.9).

Knowledge of likely clinical features of oral squamous cell carcinoma

Thirty-five percent (65/182) of respondents believed that the floor of mouth was the most common site of an oral tumour, while 23.6% realised that the tongue was the most likely site. There were a wide range of other proposed common sites of oral tumours. Few respondents realised that the tumours could be speckled in colour, although they were aware that oral tumours can rarely give rise to a number of other signs or symptoms (Table 3.5).

Management of a patient with a potentially malignant oral lesion

Most respondents indicated that they would attempt to diagnose a potentially malignant or malignant oral lesions by visual examination alone. Ten percent (16/149) suggested the use of toluidine blue as an adjunct to diagnosis (Figure 3.10). only 15.0% indicated that they would routinely undertake biopsies of the oral mucosa.

Thirty-four per cent (62/182) of the practitioners indicated that they would selectively refer a patient with a potentially malignant lesion to an appropriate specialist. And 3.2% (6/182) also suggested that they would routinely consider the removal of likely local factors prior to referring the patient (Figure 3.11). Sixty percent suggested that they would refer a patient to an appropriate specialist by letter (Figure 3.12) which routinely would include a detailed description of the lesion, and or provide a diagram of the lesion. Few, however, would provide measurements of lesion or clinical photograph (Figure 3.13).

Preventive Advice

Only 60% (110/182) of respondents indicated that tobacco and alcohol were the principal causes of oral squamous cell carcinoma. A spectrum of other possible and unlikely causes of oral squamous cell carcinoma were suggested (Table 3.6); of note about twenty percent of the respondents suggested that HIV disease was a risk factor for oral squamous cell carcinoma. Just 29.6% (54/182) of responding dentists routinely recorded the tobacco or alcohol consumption of patients and 93.9% (171/182) provided patients with any advice regarding modification of these habits (Figure 3.14 and Figure 3.15). 82.9% (151/182) of the respondents suggested that clinical screening was an effective means of reducing the frequency of potentially malignant and malignant oral lesions; and the majority suggested this would require clinical examination of each patient every 3-6 months (Table 3.7).

Discussion

In the present chapter the knowledge of the UK general dental practitioners with regard to the diagnosis, prevention and initial management of oral premalignancy and malignancy in general dental practice has been investigated.

The level of response was just above 30.0%, which is lower than the majority of postal surveys of the GDPs in the UK (Williams et al, 1995; Marley et al, 1998; Whiston et al, 1998), this low response rate may reflect the pressures of work in general dental practice in the UK, or more likely the result of the long number of items included in the questionnaire.

The data presented in this study describe the experience, knowledge and management of potentially malignant lesions (PMLs) by the responding UK GDPs. As an unknown proportion of PMLs may be managed by others, including oral & maxillofacial surgeons, ENT (ear, nose and throat) surgeons, these findings may not reflect the total picture of current management of these patients by GDPs in the UK.

It is evident that the present group of practitioners did receive some undergraduate training in the diagnosis of PMLs and oral malignancy, and most were examining patients with squamous cell carcinoma or leukoplakia. Likewise in general practice, most had patients with potentially malignant oral lesions usually manifesting as white patches.

Responding general dental practitioners were often aware of the likely sites of oral squamous cell carcinoma and most would simply refer a patient by letter to an appropriate specialist centre.

While 60% had a knowledge of the major causative factors of oral squamous cell carcinoma, just 29.7% recorded such social factors of their patients and despite the evidence that with the removal of risk factors potentially malignant lesions may regress (Lovas, 1989; Speight & Morgan, 1993) and in contrast to their faith in screening, only a minority either demonstrated active involvement in or favoured investment in health promotion. This is some what disappointing given the well known evidence linking between alcohol consumption and tobacco smoking to the development of OED and malignant oropharyngeal lesions (Binnie, 1976; Mehta et al, 1989; Hogewind et al, 1989; West & Krafona, 1990; Gupta et al, 1990). Whatever the action of alcohol and smoking on the oral mucosa may be (Rajendran et al, 1990; Pillai et al, 1990; Grady et al, 1990; Ernter et al, 1990; Bruerd, 1990; Sankaranarayana, 1990; Sato, 1991; Hu et al, 1991; Kaugars et al, 1991; Creath et al, 1991; Morse et al, 1996), their use should be actively discouraged. However, 81.8% (149/182) of the dentists enquired into the patient's social history either routinely or selectively in terms of the nature and amount of the risk factors (alcohol and tobacco) to which their patients were exposed. However, 18.1% of the dentists confirmed that the patients social history formed no part of their patients' record.

Despite the absence of data from any randomised controlled trial, the vast majority of the dentists 82.9% were convinced of the efficacy of screening, anticipating that an optimal resoursed programme might reduce oral cancer mortality. In fact, though much of the potentially malignant lesions which they currently see must follow a comparatively indolent course, most dentists would elect for a screening interval of three to six months. It would seem that if a regular programme were ever introduced, the arbiter of success or failure would not be professional commitment.

It is thus evident that general dental practitioners in the UK have some knowledge and experience of oral premalignant and malignant lesions, nevertheless they may fail to provide appropriate preventive advice and may delay referral of patients to appropriate centres.

Effective communication between primary and secondary health care workers is essential for high quality care of patients, particularly those with illnesses that are life-threatening or, more commonly, interfering with daily life. While few oral problems may be regarded as life-threatening, the increasing frequency of oral malignancy in some communities in the developed world does indicate that patients can have oral problems of significant morbidity and mortality.

There have been few studies of the quality of information primary health care workers provide to secondary health care staff when referring patients with potentially malignant disorders of the oral cavity. It is thus possible that general dental practitioners may be the first clinicians to diagnose potentially malignant oral lesions and to refer the patients to appropriate specialists.

In the present study basic administrative patient details were often missing, indeed an appropriate patient contact telephone number was provided in only 14% of letters. Details of the social history, particularly patient's ethnic origin and occupation, tobacco

smoking and alcohol drinking were rarely given. These omissions may reflect the referring practitioners not believing them as relevant or the pressures of work in general dental and medical practice in the UK. Although all of the referral letters provided some information about patients presenting complaints, precise details were often lacking, only 94 percent of referring practitioners indicated the site of the lesion, and the size of the lesion was only indicated in 33.4% of the letters. Details of the colour and duration of the lesions was only provided in about 64.6% and 53.9% of referral letters. It is thus evident that secondary health care staff would have difficulty in determining the urgency of many of these patients and thus require to correspond further with the referring clinicians to more accurately determine the urgency of the examination by specialist staff.

As with other similar studies (Jacobs & Pringle, 1990) details of the patient medical history were often omitted. While this is not an uncommon problem due to difficulties of accuracy on the part of both patient and clinician (Jones et al, 1990), it does suggest that the referring clinician might cause undue harm if he undertook a biopsy in a patient whose medical history details were not fully known. Furthermore the omission of important aspects of a medical history may limit the access of hospital clinicians to reliable background details of the patient which might otherwise assist in the diagnosis of the clinical problem (e.g. previous oral malignancy) and avoid delays that may arise as a result of additional correspondence between referring clinician and the patient's attending physician.

Of note, the referring medical practitioners were often more likely to correctly diagnose an oral malignancy than the referring dentists, although dentists could more accurately diagnose oral premalignant lesions, thus confirming previous similar observations (Scully et al, 1986).

It is evident from this study that the quality of referral letters of patients with potentially malignant disorders of the oral cavity are variable and sometimes poor. It would perhaps be charitable to suggest that this reflects a low frequency of patients who attend general dental or medical practitioners with superficial oral mucosal disease, and hence rarely refer patients to Oral Medicine units - but the lack of details of past medical history does suggest there is a need to improve the awareness of the medical and dental practitioners on relevant aspects of such disease and develop more effective methods of communication between primary and secondary health care workers. With training however the positive predictive value of referral can be improved (Warnakulasuryia & Pindborg, 1990).

It is known that there can be delays in the referral of patients with malignant and potentially malignant lesions (Cooke & Tapper-Jones, 1977; Scully et al, 1986) to appropriate specialists, possibly due to misdiagnosis and/or lack of knowledge of effective methods of referral. Furthermore, the practitioner himself is often not suspicious that a lesion may be potentially malignant or even malignant and the lesion is often treated with antifungal therapy, antibiotics, steroids and mouth washes thus contributing to further delay in the ultimate diagnosis and treatment (Langdon, 1995). Another factor is that potentially malignant lesions is not usually painful until such time as either the lesions becomes ulcerated and secondarily infected or transform to malignancy and invades sensory nerve fibres. Delay in seeking care, in making diagnosis, and in instituting treatment add to the lag time between disease onset and treatment, and may impact on survival significantly.

Previous reports indicated that delay was equal in both practitioner groups (Shafer, 1975; Cooke & Tapper-Jones, 1977; Scully et al, 1986). A more recent report by Schnetler (1992) assessed the relative performance of both groups of practitioners, and

suggested that although doctors were better at considering tumours as a diagnosis, they were seeing more advanced disease. The extent of the delay by the clinicians in establishing a diagnosis of oral cancer and precancreous lesions appear to be closely related to the degree of suspicion and diagnostic skill of the clinician, whom the patient first consulted.

Pogrel (1974) showed that "Dentists missed nearly twice as many cases of oral cancer as the asymptomatic incidental cases they spotted". Cooke and Tapper-Jones (1977) found that GMPs tended to delay onward referral for specialist treatment more than their dental colleagues. The present study, however, shows the mean delay of GMPs was 73 days compared with 80 days for GDPs.

The GDPs has much better opportunity to diagnose premalignant and malignant lesions because of training in oral medicine and pathology and routine dental check-ups. This study suggests there is much to accomplish in both GMPs and GDPs education. Since the introduction of charges for dental examination many patients seek advice from their doctor (Bhatti et al, 1995) for conditions such as mouth ulcers. Some degree of training is therefore, required for doctors in examining the mouth (Hutchison, 1994).

The absence of any suggestion of the possibility of a neoplasm or any degree of urgency in the referral letter would account for a 7 to 14 days delay, out of which about 4 days would be included in the postal time. Unless there is a degree of urgency, patients do like to have a weeks notice for appointments. Therefore these patients should be referred to hospital with a note expressing a degree of urgency to avoid unnecessary delay.

The mean patient's delay was 50.5 days (range one day to 101 days) and the proportion of patients who delayed their first visit to a health professional for greater than two months was found to be 34.4% (42/122) for intra-oral lesions and 52.3%

(11/21) for labial mucosal lesions after the first onset of signs and symptoms. The cause of this delay in seeking medical advice is likely to be fear or denial also the problems of the frail elderly patients who may not willing to be troubled by visits to doctors or the hospital as a consequence practitioners is anxious to avoid sending the patients to hospital. There are other problems such as transport or that of having to make an appointment through an institution such as old people's home (Cooke & Tapper-Jones, 1977). If there is no discomfort, it is to easy for the elderly patients to brush aside any possible significance of a swelling or ulcer in the mouth, which in his opinion any way will go just as other ulcers and swellings in his mouth have disappeared throughout his life.

This study has shown that the greatest delay in presentation of oral potentially malignant lesions was caused by patients seeking advice and practitioners delays in referring their patients for specialists opinion and these findings was similar to other studies (Scully et al, 1986; Dimitroulis, 1992).

Considering the time interval of the delay in diagnosis in this study, one should especially try to reduce patients delays. In particular, elderly patients with a history of tobacco and alcohol use should be encouraged to visit their practitioners regularly, as they are known to have a higher risk of developing oral cancer (Graham et al, 1977). Furthermore, as cardiovascular, pulmonary or liver diseases are often related to tobacco or alcohol habits, patients suffering from these diseases are also known to have a higher risk of developing oral cancer (Jovanovic, 1992) for that reason, regular examinations of the oral cavity should be advocated in these patients.

The hospital staff must do their best to expedite the channels of communication between the receipt of the referral note and the making of the first appointment.

Finally, every effort should be made to reduce the delay between the establishment of the definitive diagnosis and the beginning of treatment. This delay would seem to be in most cases administrative and capable of being contained with a limit of 14 days.

The findings of this study, as well as those of others (Scully et al, 1986; Schentler, 1992) are a cause for concern, indicating that patients lack awareness of potentially malignant lesions and do not adequately seek advice. Considering the dramatic change that delay in treatment can bring to survival rates from almost 60% 5-years survival rate for T1 tumours to less than 20% 5-year survival rate for T4 tumours (Easson & Palmer, 1976) suggest the need for improved educational programs about head and neck cancer directed to the public as well as physicians and dentists. The public should be aware not only of the aetiology of the head and neck cancers and the associated signs and symptoms, but also of the availability of the dentist as well as the physicians, as a diagnostic resource.

It is concluded that GDPs in the UK have some knowledge and experience of oral potentially malignant and malignant lesions, but they may fail to provide appropriate preventive advice and may delay referral of patients to appropriate centres.

CHAPTER 4

Risk Factors For Oral Epithelial Dysplasia

Tobacco and alcohol consumption

Outline

The epidemiology of oral cancer has been studied extensively, but less is known about the presumed precursors of the disease OED and carcinoma-in-situ. If these lesions are part of a single continuum, the risk factors identified for oral epithelial dysplasia (OED) and carcinoma-in-situ would be expected to be similar to those identified for invasive oral cancer. Factors acting in the early stages of the carcinogenic process should share similarities, but factors involved in the later stages may differ (Day & Brown, 1980).

Oral epithelial dysplasia can lead to squamous cell carcinoma; reported transformation rates range between 6.6% and 36.4% in a mean follow-up periods of 1.5 to 8.5 years (Mincer et al, 1972; Banoczy & Csiba, 1976; Pindborg et al, 1977; Gupta et al, 1980; Silverman et al, 1984; Crissman & Zarbo, 1989; Lumerman et al, 1995) (see chapter 1). Despite this malignant potential of OED, the current state of knowledge regarding risk factors for OED is limited and focused largely upon smokeless tobacco (Mincer et al, 1972; Roed-Petersen, 1973; Hirsch et al, 1982; Kaugars et al, 1989; Kaugars et al, 1991; Chakrabarti et al, 1991; Kulasegaram et al, 1995; Morse et al, 1996).

Tobacco and alcohol are the principle aetiological factors for the development of oral leukoplakia (Pindborg et al, 1967; Roed-Petersen et al, 1972; Wilsch et al, 1978; Pindborg, 1980; Loftus et al, 1981; Mehta et al, 1982; Baric et al, 1982; Gupta, 1984; Zaridze et al, 1986; Mehta et al, 1991; Evstifeeva & Zaridze, 1992) and oral squamous cell carcinoma (Rothman & Keller, 1972; Graham et al, 1977; van Wyk, 1982; Brugere et al, 1986; Blot et al, 1988; Smith, 1989; Franceschi et al, 1990; Zheng et al, 1990; MacFarlane, 1993; Zheng et al, 1997). For example cohort studies of subjects with high

alcohol consumption find an excess of oral cancer over that expected (Schmidt & Popham, 1981; IARC, 1988; Adami et al, 1992), while alcohol abstainers such as Seventh Day Adventists and Mormons have a low risk of oral squamous cell carcinoma (Lyon et al, 1980). Case-control studies generally report increased risks of oral squamous cell carcinoma in alcohol drinkers compared to non-drinkers and tobacco smokers compared to non-smokers, even after adjustment for the other factors (Graham et al, 1977; Mashberg et al, 1981; Brugere et al, 1986; Blot et al, 1988; Merletti et al, 1989; Franco et al, 1989; Kabat & Wynder, 1989; Talamini et al, 1990; Franceschi et al, 1990; Zheng et al, 1990; Boffetta et al, 1992). Almost all such studies in addition report a dose-risk relation for alcohol and/or tobacco (Rothman et al, 1972; Graham et al, 1977; Elwood et al, 1984; Brugere et al 1986), with increased consumption leading to increased risk. It is also reported that cancer of the oral cavity is more common in males and is rare in persons younger than age 40 (Blot et al, 1988; Merletti et al, 1989; Talamini et al, 1990; Hindle et al, 1996). Reported previous studies have focused upon the risk of tobacco and/or alcohol upon the development of SCC. There is however, little up to date data regarding risk factors for development of OED and site-specific differences of knowing risk factors in a population of patients resident in developed countries. Thus the aims of this chapter were:

Aims and objectives

1. To assess the risk factors for the development of histologically confirmed oral epithelial dysplasia.

2. To investigate the role of alcohol in non-smokers and of tobacco in non-drinkers in the development of OED.

3.To examine the influence of tobacco and/or alcohol consumption on the intra-oral sites of oral epithelial dysplasia.

Materials and methods

Study population

The current analyses incorporated data from two centres in Southern England. Eligible cases included all patients who attended oral medicine clinics between 1972 and 1996 inclusive where histopathological examination of their oral mucosal lesions, revealed evidence of OED (WHO, 1978; Kramer, 1980; Krutchkoff et al, 1991). Cases were identified from histopathological records held in the oral pathology and oral medicine departments and their case notes were obtained from the medical record department. Of the total 850 eligible cases, not all could be included since their records were either incomplete, or missing and untraceable. There were a total of 630 case notes from the two centres (Table 4.1).

A control group was selected from computerised list of patients attending the same oral Medicine clinics with diseases not caused by smoking or drinking. A total of 643 control subjects were selected for the study (Table 4.1). Of these, 20.9% were diagnosed with recurrent oral ulceration, 15.3% as facial pain, 14.9% oral dysaesthesia, 11.3% as trigeminal neuralgia, 6.2% Sjogren's syndrome, 13.2% temporomandibular joint pain dysfunction syndrome, 7.9% dental pain, 6.8% swellings, and 3.1% oral pigmentation.

For each case, one or more controls were identified by matching the date of birth, gender, and ethnic-background. Date of birth was matched to within 5 years. Records of 630 patients with OED and 643 control subjects were eventually collected for the study. For some patients presenting before 1980, information on their smoking habits,

and more particularly their alcohol consumption was incomplete, therefore out of 1273 subjects included in the study 564 being excluded from the analysis because of insufficient knowledge of either smoking (384), or alcohol (428), or both (248).

Data collection

Data were collected on a standardised form (Appendix 1) and entered into a computer database (EPI 5) (Dean et al, 1990). Relevant data included personal information, age, gender, subsites of the OED specified according to the International Union against cancer (UICC). History of past and present tobacco smoking and alcohol consumption, and betel quid chewing. Details of tobacco use included type of tobacco, daily amount used (expressed as cigarettes per day) (Elwood et al, 1984; Blot et al, 1988; Talamini et al, 1990; Winn et al, 1991), duration of habit (where applicable), and the number of years since cessation of smoking. Details of alcohol consumption included type of alcohol and amount consumed per week (expressed in alcoholic units) (Williams & Horm, 1977; Merletti et al, 1989; Franceschi et al, 1990; Barra et al, 1990), and for those who chewed betel nut, the number of pans used per week and duration of the patient's habit.

Unless otherwise stated the following definitions were used in this thesis with regard to tobacco use (Todd, 1972; Lee, 1976). *Smokers*; are current smokers of manufactured cigarettes, hand-rolled cigarettes, pipe, or cigars, or any combination of these. *Non-smokers*; persons who do not define themselves as smokers of cigarettes or pipe or cigar. *Ex-smokers*; persons who do not define themselves as smokers of cigarettes or pipe and who do not smoke as much as one cigar per week but who claim to have smoked in the past, at least a cigarette a day or a pipe a day or a cigar a week for as long as a year. *Smokers of filter cigarettes*; are those whose usual brand is filter-tipped

with a plug made of cellulose acetate fibres or some other material and who smoke no plain brand at all frequently. *Smokers of non-filter cigarettes*; are those whose usual brand is plain tobacco (not filter-tipped), and who smoke no filter brand at all frequently. *Smokers of hand-rolled cigarettes*; are those current smokers of hand-rolled cigarettes by self-definition. *Have never smoked*; persons who have never smoked cigarettes, pipe, cigars, hand-rolled cigarettes or any other form of tobacco.

Data analysis

The association between OED and tobacco and/or alcohol intake was determined using the odds ratio (OR), and corresponding 95% confidence intervals (C.I.), calculated within the computer package SPSS (Statistical Package for Social Sciences, 1993). Forward stepwise logistic regression was employed within SPSS, with entry of any dependent variable tested on the significance of the score statistic, and removal tested on the probability of the Wald statistic (SPSS, 1993). Following initial univariate analysis (Table 4.2), multiple logistic regression was performed to control for potential confounding factors (Breslow & Day, 1980). Tobacco smoking and alcohol drinking behaviours were analysed in detail (Tables 4.3 to 4.15) to determine their combined (synergistic) effect and their independent roles in the development of OED. Interaction components were examined, between smoking behaviour and alcohol use, for specific habits of smoking, and for certain characteristics of alcohol consumption. Dose-response relationships were further assessed by the stratified test for linear trend (Mantel, 1963; Schlesselman, 1982). Differences in proportions were tested for statistical significance using chi-square test, Fisher's exact test.

Sufficient subjects were available to allow for analysis of the effect of alcohol and tobacco on subsites within the oral cavity in both males and females. For each of tongue, labial mucosa, gingiva, floor of mouth, buccal mucosa, soft palate, other non-

specified sites, the effects of two consumption levels (divided in to two categories moderate smokers ≤ 70 cigarettes per week, heavy smokers > 70 cigarettes per week, and for alcohol moderate drinkers ≤ 21 units per week, heavy drinkers > 21 units of alcohol per week) have been related to non-consumption of both alcohol and tobacco while adjusting for other factors.

Results

Background information

Details of the subjects according to demographic characteristics and corresponding odds ratio are presented in (Table 4.1). Age, gender and ethnicity were clearly well-matched, as no relationship emerged between patient's age, gender, ethnic background and risk of having OED. Marital status was also considered but never attained significance.

The results of univariate analysis are detailed in (Table 4.2). Forward stepwise logistic regression was employed within SPSS, with entry of any dependent variable tested on the significance of the score statistic, and removal tested on the probability of the Wald statistic (SPSS, 1993). Age, gender, smoking and alcohol status were initially entered into the model, the results of which are seen in Table 4.3. Age and gender were not significantly determined by the model as predictive of oral epithelia dysplasia. These variables were considered in all subsequent models, but they are never selected, clearly indicating good matching of cases and controls. The first regression model demonstrated an affect in the development of oral epithelia dysplasia only from alcohol and tobacco use. This model incorporated a total of 709 cases and controls, the remaining 564 being excluded because of insufficient knowledge of either smoking status (384), alcohol use (428), or both (248).

Interaction between alcohol drinking and tobacco smoking

When all interaction terms were included, the model determined three significant predictors of disease, namely, 'current smoker *and* never used alcohol', 'current drinker *and* never smoked tobacco', and 'previous smoker irrespective of alcohol use' (Table 4.4) replacing the alcohol variable. Alcohol use alone was not a significant predictor, but tobacco smoking had an independent effect on the propensity of oral epithelia dysplasia. When specific behavioural characteristics were included, such as 'tobacco type' and 'alcohol type', these additional parameters supplanted the interaction terms, and alcohol returned as a significant predictor. But when information on the 'daily numbers of cigarettes smoked' was incorporated, this variable was a significant predictor of disease, and it supplanted the variables for smoking status, 'alcohol type', as well as the interaction terms (Table 4.5).

The fact that smoking status was no longer a significant predictor demonstrates the importance of heavy smoking of non-filter cigarettes over and above the more general categorisation provided by general smoking status. The 'number of years smoking', for those who had stopped smoking the 'number of years since last smoked', and for drinkers the 'number of alcoholic units consumed per week' had also been considered, but the model found no predictive power from these items of information.

Further interaction terms between alcohol and tobacco smoking were considered. These included the interaction between alcohol use and the 'daily number of cigarettes smoked' and the 'number of years of smoking tobacco'. These investigations revealed further insight into the effect of drinking alcohol in conjunction with specific smoking behaviour. The parameters for 'smoking 20+ cigarettes per day *and* a current drinker',

'smoking 20+ cigarettes per day *and* non-drinker' and 'smoking for 20+ years *and* a current drinker' all showed a synergistic effect (Table 4.6).

Interaction between tobacco-related behaviours

When smoking behaviour was examined more closely, the only significant interactive term was between smoking status and 'daily number of cigarettes smoked', where 'current smoker *and* smoking 20+ cigarettes per day' was a strong predictor of oral epithelia dysplasia (Table 4.7).

The inclusion of smoking cessation in the regression model demonstrated a protective effect for those who stopped their habit more than ten years previously (Table 4.8). However there was a higher risk associated with giving up smoking within the last ten years. Furthermore, alcohol again failed to be retained as a significant predictor of disease in the presence of such detailed smoking information. Tobacco type demonstrated an elevated effect for all forms of smoking, but now both filter and non-filter cigarette smoking being highly significant determinants of OED, with non-filter demonstrating the higher risk.

Interaction between alcohol-related behaviours

Upon examination of alcohol use, there appeared to be three main interaction terms: 'current drinker *and* drinks fortified wine', 'current drinker *and* drinks spirits', and a protective effect from 'previous drinker *and* used to drink beer' (Table 4.9). General smoking status was independently a significant predictor of oral epithelia dysplasia. However, with the inclusion of all other variables, the final model determined that smoking status was less important than 'daily number of cigarettes smoked' and 'tobacco type' - a consistent finding throughout this study (Table 4.10). Interestingly,

general alcohol use became a significant predictor of oral epithelia dysplasia at the expense of two interactive terms, the only significant one remaining being 'current drinker *and* drinks fortified wine'.

Tobacco use amongst non-drinkers

In the group of patients who exclusively smoked tobacco initial univariate analyses revealed that, although general smoking status was not a significant predictor of presence of OED, 'cigarette smoking' and in particular 'smoking 20+ cigarettes per day' were both significant predictors of OED (Table 4.11). Multiple regression found that only one smoking-related variable was a significant predictor of disease for non-drinkers, namely the 'daily number of cigarettes smoked'. Tobacco type did not influence the outcome.

Alcohol use amongst non-smokers

In patients who drank alcohol and did not smoke tobacco, there were no significant predictors of disease outcome from univariate analyses. Multiple regression confirmed that there was no significant affect from alcohol use amongst non-smokers (Table 4.12).

Age and gender as risk factors for OED subsites

The distribution of all identified OED cases by subsite, gender, and mean age at diagnosis is presented in Table 4.13. The tongue, buccal mucosa, floor of mouth were the most commonly affected sites and together accounted for more than 66% of all OED for men and women. Among the major sites, OED of the tongue, buccal mucosa, floor of mouth were more common among men whereas OED of the gingiva was most common among women. No statistical differences in mean age of the subjects for any

of the oral subsites. Male to female ratio for OED of the upper labial mucosa, ventral surface of tongue, retromolar region, buccal mucosa were 1.5, 1.3, 1.7, 1.4 respectively.

Influence of tobacco smoking upon sites of oral epithelial dysplasia

The odds ratio for oral subsites related to tobacco consumption and adjusted for alcohol usage are detailed in Table 4.14. The highest OR were of the labial mucosa and floor of mouth. In males moderate consumption of tobacco (<70 cigarettes per week) was strongly associated with OED of the labial mucosa (OR 27.0 95% C.I. 3.20, 597.8) and floor of mouth (OR 10.8 95% C.I. 3.3, 37.4), buccal mucosa (OR 3.5 95% C.I. 1.3, 9.4), in females moderate tobacco consumption was not strongly associated with OED. High tobacco consumption (> 70 cigarettes per week) in males was strongly associated with OED of the labial mucosa (OR 9.5 95% C.I. 1.2, 199.9) floor of mouth (OR 4.5 95% C.I. 1.5, 14.0) and buccal mucosa (OR 2.2 95% C.I. 1.0,4.9). While in females heavy tobacco consumption was strongly associated with OED floor of mouth (OR 3.1 95% C.I. 1.2 ,8.1), and tongue (OR 2.3 95% C.I. 1.1, 5.1). In contrast to the data of male patients the risk of OED of the labial mucosa, gingiva and buccal mucosa was not associated with heavy tobacco consumption in females.

Alcohol and sites of oral epithelial dysplasia

The risk of site of OED related to alcohol consumption in both males and females and adjusted for tobacco consumption and other factors, is given in Table 4.15. In contrast to tobacco smoking alcohol consumption was not significant determinant of OED subsites in both males and females.

Discussion

The present study adjusted for relevant confounding factors in the logistic regression analysis. However, this does not exclude bias due to unmeasured confounders, or residual confounding due to imprecise measurement. The consequences of some types of measurement errors are predictable. Random measurement errors always lead to underestimation of the effect (Rothman, 1986). If there is under-reporting, which is more likely to occur than over-reporting in the case of retrospective studies, the effect is also underestimated (Rothman, 1986). Imprecise and biased measurements not only concern the risk factors of interest but also all potential confounders in the study. The direction of bias due to inaccurate measurement of confounders is less easy to predict. In fact, it may increase or decrease the effect, depending on how the correlation with the risk factor and the disease is changed by the inaccurate measurements (Rothman, 1986). In the current study results regarding smoking and drinking could have been biased if either cases or controls were more likely to accurately or inaccurately report on these habits. It is notable, however, that the potential for recall bias with regard to the primary smoking variable, for example current smoking was reduced given the recent nature of the exposure. Similarly, a differential reluctance on the part of cases and controls to accurately report average levels of alcohol consumption may have also occurred.

Tobacco and alcohol have long been implicated as risk factors for oropharyngeal cancer (IARC, 1986,1988). Previous case-control studies (Blot et al, 1988; Merletti et al, 1989; Franco et al, 1989; Talamini et al, 1990; Franceschi et al, 1992; MacFarlane, 1993; Barasch et al, 1993; Mashberg et al, 1993) have documented an elevated risk of oral cancer and oral dysplastic lesions (Kulasegaram et al, 1995; Morse et al, 1996)

associated with smoking as well as alcohol consumption. But questions remain about the relative impact of drinking compared to smoking, the effects of different types of alcohol and types of tobacco used, and the interrelationship between these factors in the development of OED. The present data helps to resolve these aetiological issues.

The results indicate that tobacco and alcohol consumption are very important factors in the propensity of oral epithelia dysplasia and, in accordance with the available data on the aetiology of OED (Kulasegaram et al, 1995; Morse et al, 1996), potentially malignant lesions (Pindborg et al, 1977; Roed-Petersen et al, 1972; Wilsch et al, 1978; Gupta et al, 1980; Pindborg 1980; Loftus et al, 1981; Mehta et al, 1982; Baric et al, 1982; Gupta, 1984; Zaridze et al, 1986; Mehta et al, 1991; Evstifeeva & Zaridze, 1992) and invasive oral carcinoma (Rothman & Keller, 1972; Graham et al, 1977; van Wyk, 1982; Brugere et al, 1986; Blot et al, 1988; Smith, 1989; Franceschi et al, 1990; Zheng et al, 1990; MacFarlane, 1993). However, the combined role of alcohol and tobacco in the outcome of disease is complex. Although there appears to be a synergistic effect of alcohol and tobacco (when considering just tobacco smokers), heavy non-filter cigarette smoking is a very strong independent predictor of OED. The reverse is not true for alcohol. Alcohol has an important role in the development of disease only when considered in conjunction with tobacco use. The present results indicate that non-filter cigarette smoking is a very specific predictor of OED, as too is alcohol consumption, and the two habits compound one another in the overall risk of disease. However, with more detailed information regarding smoking habits, it becomes apparent that the increased risk of OED from smoking is largely attributable to heavy smoking (20 cigarettes per day or more), particularly non-filter cigarettes. Both heavy smoking and tobacco type have a combined higher risk of OED than alcohol consumption alone.

The sharp decline in risk of OED following cessation of smoking is remarkable. Indeed, the inclusion of smoking cessation in the regression model demonstrated a protective effect for those who stopped their habit more than ten years previously relative to non-smokers. The drop in risk in such a relatively short time suggests that smoking primarily effects the late stages of OED. Morse and co-workers (1996) reported a decline in the risk of OED after tobacco cessation, with ex-smokers of 10 years+ demonstrating no excess risk relative to non-smokers. A finding also observed by other authors with regard to SCC (Wynder & Stellman, 1977; Blot et al, 1988; MacFarlane, 1993), bladder cancer (Hartge et al, 1987), and lung cancer (Surgeon general, 1982).

What is less clear from the present results is the appearance of a minor protective effect from moderate smoking (less than 10 cigarettes per day). This could well be an artefact of the present data, or simply reflect the difficulties of using retrospective medical records. Moderate smoking patterns may incorporate a number of social occasional smokers, or those who move between abstaining and relapsing into the smoking category.

Amongst certain studies (Franceschi et al, 1990; Zheng et al, 1990; Talamini et al, 1990; Boffetta et al, 1992), and initially within this study, the impact of alcohol appears significant and possibly greater than that of smoking *per se*. As more detailed smoking information is included, it becomes clear that the risk of OED attributable to smoking is largely due to heavy smoking (20 cigarettes per day or more) of non-filter cigarettes. The role of alcohol is therefore confused. Although, within multivariate analysis, the individual impact of both tobacco and alcohol is only moderately reduced from that of the univariate analysis. No correlation between degree of alcohol

consumption and risk of OED was observed in the present study. However, increased risk of OED was associated with alcohol consumption of fortified wines and spirits, suggesting a role for ethanol, the common ingredient in these beverages. Kulasegaram and co-workers (1995) reported that the proportion of subjects who drank spirits was significantly higher among cases of OED than control subjects. While others demonstrated a 2-fold increase in the risk of OED with the consumption of beer and hard liquor and no increased risk was observed for wine consumption (Morse et al, 1996). Some studies have suggested spirits and beer (Blot et al, 1988; Merletti et al, 1989) more important risk factors than wine, others have suggested that the highest risk to be associated with wine consumption (Franco et al, 1989; Barra et al, 1990), and other authorities believe that there is no difference in risk potential between types of alcoholic beverages (Doll, 1992; Barasch et al, 1993). A detailed case-control study conducted in Italy reported increased risks of SCC only for those with a very high wine consumption (over 8 drinks per day) (Talamini et al, 1990). The precise reasons for the differential risks are not clear, but it is possible that ingredients in alcoholic beverages other than ethanol are involved, for example, nitrosamines and polycyclic hydrocarbons in whisky (Kleinjans et al, 1996). Experimental studies have not implicated alcohol itself as a carcinogen, although it may promote carcinogenesis by a variety of mechanisms, including nutritional deficiencies associated with heavy drinking, the effect of contaminants and congeners in alcoholic beverages, the induction of microsomal enzymes that enhance the metabolic activation of tobacco or other carcinogens, and the capacity of alcohol to solubilize carcinogens or enhance their penetration in oropharyngeal tissues (Lieber et al, 1979; Tuyns, 1982). Ethanol can enhance the conversion of procarcinogens to mitogens, and its metabolite acetaldehyde can produce DNA abnormalities in human cells (Mufti, 1992). In addition, heavy intake

of alcoholic beverages is correlated with nutrient deficiency (Oth, 1986; Deleyiannis et al, 1996)), which appears to contribute independently to oral carcinogenesis (Marshall et al, 1982; Winn et al, 1984; Ogden & Wight, 1998). Chronic alcohol consumption can lead to salivary gland atrophy and hence reduce salivary flow (Maier et al, 1986). Saliva has been shown to have an inhibitory effect on mutagenicity and clastogenicity (Maier et al, 1986), thus reduced saliva could lead to increased exposure of the oral mucosa to carcinogenes (Simanowski et al, 1995) and hence increased risk for oral cancer. Further studies are needed to determine whether alcohol promotes the effect of other exposures such as human papilloma-viruses.

Within this study, it was not clear if pan chewing included tobacco, and it is debatable whether pan chewing *per se* could be responsible for OED. This is perhaps why the variable explicitly recording tobacco type is a greater predictor of outcome than pan chewing status. Furthermore, problems within this data arose because there were so few controls amongst pan chewers, and it was difficult to incorporate this variable within regression models. With the limited number of cases in this investigation, further studies are needed to test the synergistic effects, if any, between betel quid chewing and its constituents on one hand, and alcohol drinking and cigarette smoking on the other.

The results provide evidence that tobacco use exclusive of alcohol consumption is independently related to the risk of OED. In particular, the number of cigarettes smoked per day was the most significant predictor of risk of OED.

A strong direct association between non-drinking smokers and risk of oral Squamous cell carcinoma (IARC, 1986; Talamini et al, 1990; MacFarlane, 1993) is already

known. It is possible that exposure to tobacco related carcinogen disrupts cellular function in the development of OED and oral squamous cell carcinoma. For example, Lippman and co-workers (1995) reported abnormal p53 expression in 75% of a group of smokers with oral leukoplakias whereas this was not present in oral leukoplakia of non-smokers control subjects. The significance of tobacco in inducing epithelial hyperplasia and dysplasia has been shown by others (Muller & Krohn, 1980) and the role of tobacco in SCC and OED may manifest it self in two ways: by causing morphologically visible alterations and by an increased number of proliferating cells present in not yet morphologically altered epithelium (von Oijen et al, 1998).

The present results suggest that exclusive alcohol consumption was not a significant risk factor in the development of OED, however, combined alcohol and tobacco consumption did have a significant effect in the development of OED. These conclusions are in broad agreement with data concerning the role of alcohol in oral squamous cell carcinogenesis (Zheng et al, 1997). Indeed Squier and co-workers (1986) reported that ethanol enhance the penetration of tobacco carcinogens across the oral mucosa.

Many previous studies have combined non-smokers and light smokers (i.e. <10 cigarettes per day) (Graham et al, 1977; Olsen et al, 1985; De Stefani et al, 1988; Franco et al, 1989) making it difficult to interpret the increased risk of oral and pharyngeal cancer in individuals who report high alcohol consumption. In a population based case-control study from Northern Italy, alcohol consumption did not influence risk of malignancy in light smoking males or non-smoking females (Merletti et al, 1989). Likewise in a house-to-house survey of the association of alcohol habits with oral leukoplakia alcohol consumption was only of aetiological significance when in association with tobacco smoking or chewing (Gupta, 1984). Nevertheless results of

other studies have suggested a doubling of risk of oral and pharyngeal squamous cell carcinoma with alcohol in persons who did not smoke (Rothman & Keller, 1972; Elwood et al,1984). Also, in a large study, Tuyns and co-workers (1988) felt that alcohol has some influence in risk of pharyngeal cancer across all strata of smoking habits, reporting only 9 cases of lifetime non-smokers with tumours of the hypopharynx or epilarynx. The current study did not find any significant association between risk of OED and specific type of alcohol among non-smoking drinkers. This contrasts with a previous study of OED in which consumption of spirits was more closely associated with risk of OED than other types of alcoholic beverages (Kulasegaram et al, 1995). However, in that study the total number of patients studied were small and confounding factors such as tobacco and other demographic characteristics are not controlled. Likewise, some studies of oral and pharyngeal cancer and precancer have suggested that alcohol type may influence the risk of malignancy and leukoplakia (Blot et al, 1988; Merletti et al, 1989; Franco et al, 1989; Barra et al, 1990; Macigo et al, 1995). The mechanisms by which alcoholic beverages induce oral cancer or dysplastic lesions are not well understood. Ogden and Wight (1998) suggested combination of influences, local (e.g. direct effect on cell membrane, alteration in mucosal permeability, variation in tissue distribution and type of enzyme involved in alcohol metabolism) and systemic (e.g. nutritional deficiencies, immunological deficiencies and disturbed liver function), but clear information on risks according to timing of exposure, and on changes in risk following cessation of drinking require further study.

The predominant role played by alcohol and/or tobacco upon the development of OED may depend on the population under study. In a predominantly smoking population,

tobacco may be the main aetiological factor in the development of OED as with oral squamous cell carcinoma (Phillips et al, 1980; Lyon et al, 1980), while alcohol may be relevant in those populations that predominantly drink alcohol as compared to smoking tobacco (IARC, 1988; Adami et al, 1992). A similar reasoning applies to such population groups where neither factor is supposed to be highly prevalent, assuming that a third still unknown or ill defined factor could be shown to play a significant role, this third factor would be the predominant factor.

In the UK smoking of cigarette is by far the most common manner of tobacco consumption (Wald, 1991). World-wide various other tobacco habits are common, like reverse smoking, use of smokeless tobacco with or without lime or betel quid (Winn et al, 1981; Sankaranarayanan et al, 1989). These habits may influence the distribution of OED in various subsites of the oral cavity, for instance, oral squamous cell carcinoma in reverse smokers has a predilection for the palate and in users of smokeless tobacco the sites of predilection are where the product is kept, such as the lower bucco-alveolar sulcus or gingiva (Winn et al, 1981; Sankaranarayanan et al, 1989). Based on a series of 543 white males with oral cancer, Wynder and co-workers (1957) reported that the percentage of heavy smokers (more than 35 cigarettes per day) with floor of mouth cancer was significantly greater than the expected if one presumed that the risk for all sites was the same. Similarly, in a hospital-based study involving 598 case-control pairs of US male veterans, Keller and Terris (1965) found a statistically significant excess of heavy smokers (more than 40 cigarettes per day) among cases of tongue and floor of mouth cancer but not among cases of oral cancer from other aggregated sites. Other studies (Blot et al, 1988; Franco et al, 1989; Franceschi et al, 1992) have compared odds ratio for smoking and cancer of the tongue to odds ratio for smoking and cancer at all other intraoral sites combined. The findings, however, have not been

consistent across studies and cannot be readily compared to the result of the current study. Other studies (Hirayama, 1966; Spitz et al, 1988) provide estimates of the risk associated with smoking for cancers located at two, but not all, of the specific intraoral sites addressed in the current study. Hirayama (1966) reported point estimates for the relative risk of smoking that were higher for cancer of the tongue than cancer of the gingiva. Spitz and colleagues (1988) estimated odds ratio for smoking that were notably higher for cancers of the tongue and floor of the mouth than for cancers at all other oral sites combined, particularly regarding heavy smoking. A number of other published studies (Jussawalla & Deshpande, 1971; Silverman et al, 1983; Merletti et al, 1989; Barasch et al, 1993) have found the percentage of smokers to be notably lower for the gingival cancer groups relative to the case groups comprised of tongue or floor of mouth. However, (Brugere et al, 1986; Boffetta et al, 1992) reported a high percentage of smoking for all case groups regardless of the cancer site.

Although the results of the current investigation are consistent with most previous studies of oral cancer (Jussawalla & Deshpande, 1971; Silverman et al, 1983; Spitz et al, 1988; Blot et al, 1988; Franco et al, 1989; Merletti et al, 1989; Franceschi et al, 1992; Barasch et al, 1993) in suggesting that smoking poses a greater risk of OED at some sites than at others, the present findings further suggest that OED of the labial mucosa and floor of mouth in males and tongue and floor of mouth in females is more strongly associated with smoking than are OED of the gingiva and buccal mucosa. The observed association may be partially explained by a number of a possible mechanisms. Keratin present on the attached gingiva may protect against carcinogenic effects of tobacco. The anatomy of the oral cavity also play a role in that tobacco products pool in the floor of mouth and ventral surface of tongue region, thus allowing increased contact time of noxious substances with these tissues. Finally, it has been suggested that direct

exposure to smoke may also effect the distribution of tobacco-related oral carcinomas (Boffetta et al, 1992). All of these hypotheses, however, require further investigation.

Conflicting results have been reported on the effects of alcohol consumption on the site of squamous cell carcinoma. In the present analysis alcohol consumption is not a significant predictor of various sub-sites of OED in both males and females. Herity and co-workers (1981) found an increased risk for tongue cancer for light drinkers and ninefold risk for heavy drinkers. Llewelyn and Mitchell (1994) confirmed the association between alcohol and cancer, with a site predilection of tongue and floor of mouth. Jovanic and co-workers (1993) also found floor of mouth to be the high risk site for alcohol related oral cancer. Boffetta and co-workers (1992) had reported that cancer of the tongue as well as cancer of the floor of mouth were more strongly associated with alcohol consumption. In the same study tobacco was most strongly associated with the "soft palate complex". Other authors analysed both sexes combined and reported increased risks in non-smoking alcohol consumers (Elwood et al, 1984; Talamini et al, 1990), while Wynder and co-workers (1957) found no such effect. Blot and his colleagues (1988) considered such an effect by sex and reported increased risks among males with inconsistent results noted amongst females. They also found positive trends of alcohol and tobacco consumption and oral cancer risk were slightly weaker for the tongue when compared with other intra-oral sites.

Male to female ratios varied notably by intraoral sites (Table 4.13), with the highest ratio being observed for OED of retromolar area and the lowest being seen for OED of the commissure. However, because the male to female ratio is not closer to 2:1, the possibility of selection bias cannot be ruled out. If cases with OED at a specific sites were more likely to have been included in the case group on the basis of an exposure studied in the current investigation, such a bias could have influenced the results

regarding age, gender, and smoking. Gender-specific differences to environmental risk factors may explain the observed variation in male to female ratios.

In general age was not a significant predictor of specific OED subsite. For most of OED sites considered, the sixth decade of life marked the mean age at diagnosis. A notable exception to this finding was OED of the commissure, with a mean age at diagnosis of 42 years. however, because only three such cases were included in the analysis, caution must be exercised in interpreting this finding.

It is concluded that exclusive tobacco consumption particularly of (20+ of non-filter cigarettes) is more important risk factor for OED than exclusive alcohol consumption. However, there is synergistic effect of alcohol when combined with some aspects of tobacco smoking. The relative risk associated with tobacco smoking appeared to be highest for OED in the labial mucosa and floor of mouth for males and tongue and floor of mouth for females, while alcohol drinking is not a significant predictor of specific OED sub-sites in both males and females.

Some systemic risk factors for oral epithelial dysplasia

Haematinic deficiency and possible association with oral epithelial dysplasia

Outline

It is estimated that diet or nutritional factors contribute to about 35% of all cancer deaths (Doll & Peto, 1981). Epidemiological and experimental data have suggested that some micronutrients, including various carotenoids, retinoids and α-tocopherol may have chemopreventive activity against certain types of cancers (Betram et al, 1987; Moon & Micozzi, 1989; Oth, 1989; Garewal, 1991; Gensler & Magdaleno, 1991). It has been shown that intake of beta-carotene, α-tocopherol (vitamin E) and retinol (vitamin A), or its analogues may cause regression of oral leukoplakia, thus preventing its progression to malignancy (Shah et al, 1983; Koch, 1978; Garewal et al, 1990; Stich et al, 1991; Benner et al, 1993).

There is evidence that folate deficiency may be involved in the aetiology of carcinoma of oesophagus (Jaskewicz et al, 1988), bronchi (Heimburger et al, 1988), colon (Lashner et al, 1989), cervix (Butterworth et al, 1982; Butterworth, 1991), and haematopieotic system (Branda et al, 1978), as well as in certain experimental models of carcinogenesis (Hsieh et al, 1989). There is a growing body of evidence that supports an association between low systemic levels of folate and/or vitamin B12 and an increased risk of cancer and precancer in epithelial tissues (Mitchell et al, 1986; Eto & Krumdieck, 1986; Sankaranarayanan et al, 1989; Ramaswamy et al, 1996). A positive association may also have importance in supporting and expanding the clinical work of Heimburger and collaborators (1988) who have suggested that the treatment of epithelial precancers with vitamin B12 and folate supplements prevents their transformation into neoplasms.

Mucosal atrophy is a common feature of various conditions considered to increase the liability to oral malignancy and premalignancy (Cooke, 1975; MacDonald, 1975). Prime and his colleagues (1983) found that iron-deficient rats developed experimental oral tumours faster than the normal controls and that iron-deficient animals had significantly more lingual tumours. In experimental animals, iron deficiency lead to changes in the cell kinetics (Rennie et al, 1984) and also increase the susceptibility to topically applied carcinogens (Prime et al, 1983).

As detailed in chapters 1 and 4 tobacco and alcohol use are accepted as the most important risk factors for oral precancers (Pindborg et al, 1972; Banoczy, 1982; Mani, 1985; Zaridze et al, 1986; Macigo et al, 1995) and OED (Kulasegaram et al, 1995; Morse et al, 1996). Exposure to cigarette smoke may result in folate deficiency via chemical inactivation and thus render the epithelium more susceptible to neoplastic transformation by the carcinogenic hydrocarbons of tobacco smoke (Heimburger et al, 1987). Several of the hundreds of chemical components of cigarettes smoke, primarily organic nitrites, cyanates, and isocyanates, have been shown to interact with folate and vitamin B12 coenzymes, transforming them into biologically inactive compounds (Francis et al, 1977; Khalid & Krumdieck, 1985; Khalid et al, 1986). That these chemical interactions may have physiological significance is supported by reports of lower circulating folate (Witter et al, 1982; Nakazawa et al, 1983) and vitamin B12 (Dastur et al, 1972) levels in smokers. Heimburger and co-workers (1992) found the plasma folate levels of smokers with potentially premalignant bronchial squamous metaplasia to be lower than those of smokers without metaplasia. Smoking may diminish indicators of systemic folate status (Heimburger et al, 1987) as well as folate concentration in the oral mucosa (Piyathilake et al, 1992).

Epidemiological and clinical evidence suggest that folate deficiency in certain epithelial tissue, regardless of systemic folate status, may be a factor that predispose to the development of neoplasms arising from these tissues (Heimburger, 1992). Folate supplementation thought to have resulted in correction of cellular abnormalities associated with diminished folate status (Heimburger et al, 1988). And profound vitamin B12 deficiency can cause moderate-to-severe oral mucosal dysplasia that resolves after correction of the vitamin B12 deficiency (Theaker et al, 1989).

The aims of this part of the study were:

1. To prospectively establish the baseline circulating levels of the serum folate, red blood cells folate, vitamin B12, ferritin, iron, and total iron binding capacity (TIBC) in normal control subjects with and without tobacco smoking and to compare these levels with the values obtained in cases of oral OED.

2. To evaluate the interaction between folate, vitamin B12 with tobacco smoking and alcohol consumption on the risk of OED.

Materials and methods

Study population

Patient group

The study group comprised a total of 120 patients with histologically confirmed OED (WHO, 1978; Speight et al, 1996) (64 males, 56 females, median age 54 years, range 29-80) attending the Oral Medicine Department of the Eastman Dental Institute for Oral Health Care Sciences between 1995 and June 1997 were selected for the study.

Control group

Control patients were selected from those attending the same clinic with oral diseases not caused by smoking or drinking or related to known haematinic deficiency. 250 controls were selected for the analysis. Of these, 32.4% were diagnosed with dental pain, 22.0% as Sjogren's syndrome, 20.0% as temporomandibular joint dysfunction syndrome, 14.4% as swellings, 6.8% as dental defects, 4.4% trigeminal neuralgia. Case and controls was matched for gender, race, date of birth (within 5 years).

Case and control subjects were interviewed in person and relevant data was collected in a standard, structured questionnaire (Appendix 3). Information on prior use of tobacco and alcohol, type, site, duration of the dysplastic lesions, grade of dysplasia, treatment, dental hygiene, educational level, life long occupational history, past medical history, and family history of OED and cancer were also collected.

A current smoker was defined as someone who had smoked within the year preceding diagnosis, and previous smoker as some one who had smoked but had stopped more than one year prior to diagnosis. Questions regarding the major parameters of tobacco use included: type of tobacco used (use of filter and non filter cigarettes, cigars, pipe, roll-up and chewing tobacco/taking snuff, chew betel quid); duration of smoking in years; average number of cigarette smoked per day or amount of tobacco smoked in (grams) per month.

Data on alcohol consumption included: type of alcoholic beverage used, (beer including lager and cider, wine, fortified wine including sherry, port, martini and spirits), amount of alcohol consumed per week (in unit) (1 unit equal to 1/2 pint of beer or lager, or one glass of wine, one glass of fortified wine, one measure of spirits), and total duration of drinking in years.

Haematological assessment

A venous blood sample was drawn from each patient and control subject and divided for determination of serum folate, red blood cell folate, vitamin B12, iron, ferritin and total iron binding capacity (TIBC). The complete blood count (CBC) included determination of haemoglobin, red blood cell, red blood cell indices, and white blood cells with differential using standard methods. All blood samples from patients and controls were drawn in the morning to provide consistency in interpretation of results. None of the patients or controls were taking any medications at the time of testing.

Data analysis

The crude odds ratio and the corresponding confidence intervals were calculated using the Mantel Hanszel method for each parameter. Adjusted ORs were obtained by conditional logistic regression taking into consideration the matched design. The assessment of the effect modification between tobacco smoking and alcohol drinking was conducted using conditional logistic regression analysis (Breslow & Day, 1980). Analysis of the differences between serum folate, red blood cell folate, vitamin B12, iron, total iron binding capacity and ferritin in cases and controls was carried out using Student's t-test (Swinscon, 1996). Significance was accepted when the *P*-value was less than 0.05.

Results

Age and gender

Age and gender distribution of the study subjects is detailed in (Table 4.16). Most cases were male (53.3%), and white (81.6%). The median age at diagnosis of the patients was 54 years (range 29 to 80).

Age and gender were not significantly determined by the model as predictive of oral epithelial dysplasia. These variables were considered in all subsequent models, but they are never selected, clearly indicating good matching of cases and controls.

Serum folate, red blood cell folate and vitamin B12 among cases and control subjects

Mean serum levels of vitamin B12, folate, and red blood cells folate in normal non-smokers and smokers control subjects compared with OED are detailed in (Table 4.17). A significant decrease in the serum levels of folate, red blood cell folate were found in OED compared to normal tobacco smokers ($P<0.05$). No significant differences in vitamin B12 was found between OED cases and normal control subjects.

Serum ferritin, iron and total iron binding capacity among cases and control subjects

Estimation of serum ferritin, iron, total iron binding capacity among cases and control subjects revealed low mean serum ferritin and iron in the control subjects compared with OED cases and significant differences in serum ferritin level were found between OED cases and normal drinkers of alcoholic beverages ($P<0.05$). And no significant differences in the TIBC level in OED compared with control subjects (Table 4.18).

Folate and vitamin B12 as risk factor for oral epithelial dysplasia

Low levels of red blood cell folate, serum folate were found to be significantly related to oral epithelial dysplasia. The effect of these factors having a significant association with OED, as assessed by 95% C.Is or trend tests, P values, are presented in Table 4.19.

Tobacco and alcohol as risk factors for oral epithelial dysplasia

Forward stepwise logistic regression was employed within SPSS, with entry of any dependent variable tested on the significance of the score statistic, and removal tested on the probability of the Wald statistic (SPSS, 1993).

The regression model demonstrated an effect in the development of OED only from alcohol and tobacco use. This model incorporated a total of 370 cases and controls. When smoking behaviour was examined more closely, the only significant interactive term was between smoking status and 'daily number of cigarettes smoked', 'number of years smoking' where 'current smoker *and* smoking 19+ cigarettes per day' and current smokers and smoking for more than 39 years was a strong predictor of OED (Table 4.20). Increased risks of OED were found regardless of type of cigarette smoked, although more risks associated with cigar and non-filter cigarette OR 82.5 (95% C.I. 18.3, 731) and 10.2 (95% C.I. 4.3, 24.1) respectively.

Alcohol use alone was not a significant predictor, when specific behavioural characteristics were included, such as 'tobacco type' and 'alcohol type', these additional parameters supplanted the interaction terms, and alcohol returned as a significant predictor. But when information on the 'daily numbers of cigarettes smoked' was incorporated, this variable was a significant predictor of disease, and it supplanted the variables for smoking status, 'alcohol type', as well as the interaction terms. The results of alcohol need to be interpreted with caution, as only four patients among OED cases were non-drinkers of alcohol.

Interaction between folate and vitamin B12 with tobacco and alcohol in the aetiology of OED

Further interaction terms between red blood cell folate and tobacco use was considered. These investigations revealed further insight into the effect of red blood folate in conjunction with smoking behaviour. The parameters for current or ex-smokers and smoking more than nineteen cigarettes per day and a current smokers and smoking for more than 19 years all showed a significant interaction with low concentration of RBC

folate (Table 4.21). Vitamin B12 demonstrated no significant interaction with RBC folate in the aetiology of OED.

The 'type of cigarette used', and for drinkers the 'number of alcoholic units consumed per week' had also been considered, but the model found no predictive power from these items of information.

Discussion

The results of this study confirm the significance of tobacco smoking and alcohol consumption as risk factors in the aetiology of OED and provide additional evidence linking the status of folate to OED.

The benefits of folate (Butterworth, 1981; Heimburger et al, 1988; Butterworth et al, 1992), cobalamin (Heimburger et al, 1988) in reducing the risk of cancer or precancer in epithelial tissues have been described in the literature. Furthermore low level of folate was found to be related to an increased risk of epithelial dysplasia or carcinoma-in-situ (Buckley et al, 1992; Correa, 1992). These nutrients are likely to take the active role in the risk reduction effect. The blood concentration of folate, carotene and ascorbate have been shown to be lower in tobacco smokers than in non-smokers (Witter et al, 1982; Nakazawa et al, 1983; Chow et al, 1986) and the buccal mucosal cells of tobacco smokers were shown to have a decreased concentration of folate (Piyathilake et al, 1992). This study shows significantly low levels of serum folate and red blood cell folate in tobacco smokers. Some of the carcinogenic substances present in tobacco smoke 'primarily organic nitrites, cyanates, and isocyanates', have been shown to interact with folate and vitamin B12 coenzymes, transforming them into biologically inactive compounds (Francis et al, 1977; Khalid & Krumdieck, 1985; Khalid et al, 1986) that these chemical interactions may have physiological significance is supported

by reports of lower circulating folate (Witter et al, 1982; Nakazawa et al, 1983) and B12 (Dastur et al, 1972) levels in smokers. Sufficient quantities of folate and cobalamin are required for adequate functioning of a cellular repair mechanism to control damage from cytotoxic and genotoxic manifestation, thus inhibiting neoplastic transformation (Eto & Krumdieck, 1986). The chemopreventive actions of these nutrients are linked. Folate is crucial in the metabolic pathway of DNA synthesis, and methylcobalamin is required as a coenzyme for methionine synthase, an enzyme involved in a critical C-1transfer in the folate metabolism. A deficiency of cobalamine which slows or shuts down the methyl transfer, causes the body folate to accumulate as 5 methyl tetrahydrofolate, unable to be demethylated to tetrahydrofolate, and returned for use in other essential folate pathways. Tetrahydrofolate is required in a pathway that produces the DNA precursor material deoxythymidine monophosphate from deoxyuridine monophosphate. In a state of folate or cobalamin deficiency, deoxythymidine monophosphate may not be present in sufficient quantity, and nucleotide biosynthesis can be slow or halted (Zitton & Copper, 1989). Folate sensitive fragile sites exist in human chromosomes (Krumdieck & Howard, 1983; Chem et al, 1989; Yunis & Hoffman, 1989).

It is generally acknowledged that RBC folate levels provide a more accurate indication of long term nutritional status than plasma or serum folate level, which are influenced by recent ingestion of food. The findings of this study provide evidence that inadequate reserve of folate, as reflected in RBC folate contents, enhance the effect of tobacco smoking on OED risk. The effect of low RBC folate alone was weak. Cigarette smoking had a less strong association with OED among patients with RBC folate above 300μg/l. However, among patients with low RBC folate concentration, tobacco

smoking demonstrated a much stronger effect. A similar observation was reported by other investigators with regard to cervical dysplasia (Butterworth et al, 1992).

The lack of certain micronutrients-rich foods in the diets of tobacco smokers may be associated with increased risk of OED. In a review of the role of vitamin B12 and folate deficiencies in carcinogenesis, Eto and Krumdieck (1986) observed that neither deficiency is carcinogenic by itself but that each may increase susceptibility to the action of other carcinogens. Folate deficiency associated with chromosome breaks (Yunis & Soreng, 1984) which can allow the insertion of viral oncogenes or can cause translocations with resultant alteration in genetic regulatory mechanism (Rowley, 1990). A deficiency of folate and related methyl donors has also been reported to enhance the expression of endogenous and exogenous oncogenes that might other wise be suppressed (Hsieh et al, 1989).

Nutritional factors are of great importance in maintaining the integrity of the oral mucosa (Rennie et al, 1984; Mitchell et al, 1986) and thorough haematinic investigation is recommended in the management of potentially malignant oral lesions, particularly in Asian patients in whom these deficiencies are prevalent (Warnakulasuriya et al, 1992). Serum iron assay alone are of little significance without relating these to total iron binding capacity. Both these values are subject to variability and serum iron level is also subject to diurnal variation and merely indicate the efficacy of iron transportation within the body to sites of erythropioeisis. The diurnal variation is reported to exceed 50 percent (Speck, 1968; Weinberg, 1974). For this reason, all blood samples were drawn in the morning to provide consistency in interpretation. Low serum iron values and a correspondingly low TIBC may represent anaemia of chronic disease, whereas low serum iron values and an elevated TIBC represent true iron deficiency.

A more accurate assay is serum ferritin level, which reflects the level of

total body iron stores. A patient can be sideropenic therefore some time before they are anaemic. In this study serum iron, total iron binding capacity and ferritin level were all within normal limits among OED cases and the normal healthy control subjects. The biochemical changes in iron deficient epithelium including decrease cytochrome C levels and enzyme depletion have been reported (Dagg et al, 1966; Rennie et al, 1982). Iron deficiency whether it is severe enough to cause anaemia or not can lead to mucosal atrophy (Ranasinghe et al, 1983; Rennie et al, 1984). It has been suggested that mucosal atrophy, increased mitotic activity, and diminished repair capacity are among the major common underlying predisposing factors in oral cancer (Morgan & Johnson, 1986). It is recognised however, that in certain cases other associated deficiencies of essential nutrients and vitamins may arise and complicate the situation (Rennie et al, 1982). From this study it is not possible to differentiate between the effect of iron from other nutrients such as vitamin B12 and folate, however the available evidence support that etiological role of iron deficiency in development of epithelial lesions (Rennie et al, 1982).

In this case-control study, the low level of folate concentration together with tobacco smoking was found to be a risk factors for OED. Clinical trials to investigate the effectiveness of supplementation of this micronutrient in reducing the incidence of oral OED and its subsequent malignant transformation may be warranted.

Infection as a risk factors for oral epithelial dysplasia

Outline

Candida albicans, is an oral commonsal in at least 40% to 60% of healthy adults (Krogh et al, 1987). Up to 39% of leukoplakia lesions have evidence of invasion by fungi 'predominantly C. albicans (Pindborg, 1980)', Candida being more common in speckled than homogenous leukoplakias (Daftary et al, 1972). The presence of Candida hyphae may simply represent superimposed infection, nevertheless as non-homogenous type of oral leukoplakia have been reported to have a higher probability of malignant transformation than homogenous form (Pindborg, 1980; Axell et al, 1984; Bouquot, 1991; Speight & Morgan, 1993), and fungal infection is more commonly associated with the former a causal role for Candida in malignant transformation of such lesions has been suggested (Renstrup et al, 1970; Cawson & Rajasingham, 1972; Banoczy & Sugar, 1972). Nevertheless as Candida *spp.* are part of the normal oral flora (Krogh et al, 1987), an aetiological association of Candida *spp.* in oral premalignancy and malignancy may be debatable. Furthermore there has been very few reports of malignant transformation in patient with chronic mucocutaneous candidosis (Firth et al, 1997). Previous studies of the frequency of oral carriage of C. albicans in patients with oral leukoplakia are summarised in (Table 4.22) in these studies the frequency has varied from 6.7% (Daftary et al,1971) to 54.2% (Jepsen & Winther, 1965).

Hepatitis C virus (HCV) is an RNA virus comprising six different types and at least 40 subtypes. The vast majority of subjects infected by HCV develop chronic liver disease, cirrhosis complicates more than 20% of these individuals and some of these patients develop hepatocellular carcinoma (Iwarson, 1994; Dusheiko et al, 1996). HCV has also been reported to occur in association with a wide variety of extrahepatic immunologically mediated abnormalities including the generation of a number of tissue

specific and non specific autoantibodies (Clifford et al, 1995), cryoglobulinaemia (Gumber & Chopra, 1995), autoimmune thyroiditis (Tran et al, 1993), lymphocytic sialdenitis resembling Sjogren's syndrome (Haddad et al, 1992), diabetes mellitus (Allison et al, 1994) and possibly non-Hodgkin's lymphoma (Ferri et al, 1995).

Clinical and epidemiological data demonstrate that chronic HCV infection is a major cause of hepatocellular carcinoma (Blum, 1994). A relationship between liver cirrhosis and oral cancer has been confirmed by epidemiological and experimental studies (Lekholm & Stenman, 1989; Gerson, 1990). Recently an association between HCV-infection and oral squamous cell carcinoma has been suggested as 24% of a group of Japanese patients with oral squamous cell carcinoma were HCV seropositive, and 17% having HCV RNA in serum (Nagao et al, 1995). Of interest Porter and co-workers (1997) described a patient with hepatitis C virus infection with long standing oral lichen planus who developed a squamous cell carcinoma of the tongue. Likewise Carrozzo and co-workers (1997) confirmed the same findings in an Italian woman known to have lichen planus and HCV infection who developed verrucous carcinoma in absence of any other known risk factors.

Helicobacter-pylori is a microaerophilic, gram-negative spiral bacterium. It is the most common bacterial pathogen world-wide, is the major cause of the chronic active type B gastritis and it is strongly associated both with peptic ulcer disease and gastric cancer (Lambert et al, 1995). H. pylori has been found in gastric mucosa, gastric secretions, stools, and saliva. It may be transmitted orally or via the faecal-oral route. (Lambert et al, 1995; Nguyen et al, 1995).

H. pylori has been detected in dental plaque and saliva in healthy persons and in persons with gastric disease. The frequency of detection varies between 1.4% and 10.0%, possibly reflecting differences of methods and patient populations (Majmudar

et al, 1990; Desai et al, 1991; D'Alessandro & Seri, 1992; Bernander et al, 1993; Patel et al, 1994). Nevertheless, studies with the polymerase chain reaction (PCR) suggest that H. pylori may be part of the resident oral microflora (Nguyen et al, 1995). On the basis of the epidemiological observation of an increased prevalence of H. pylori infection in patients with gastric cancer (Correa, 1990; Eurogast Study group, 1993), a relationship between H. pylori and gastric cancer has been proposed. There is no data on association of C. albicans, Hepatitis C virus and H. pylori with OED, thus the aims of this part of the study were:

1. To determine the frequency and amount of the salivary carriage of C. albicans in a large cohort of patients with oral epithelial dysplasia and to describe the frequency of the histopathologically proven invasion of fungi in the lesions and to correlate malignant transformation of oral epithelial dysplasia with presence of C. albicans.
2. To evaluate the HCV status of a group of patients with OED and compared it to control subjects.
3. To investigate the frequency of carriage of serum IgG antibodies to H. pylori in patients with OED.

Materials and methods

Patient groups

Patient group for assessment of Candida albicans in oral epithelial dysplasia

The study group comprised of 223 patients (117 males, 106 females, median age 54 years, range 24-83) who attended Department of Oral Medicine of the Eastman Dental Institute for Oral Health Care Sciences, London, UK between 1980 to 1996 known to have histopathological features of OED (WHO, 1978; Speight et al, 1996).

For the cases medical history, salivary Candida count, Candida hyphae in biopsy reports, malignant transformation, were recorded. For the controls age, gender, medical history, salivary Candida count, were recorded. One hundred and sixty-seven (74.8%) OED patients were found to have detectable salivary Candida counts and compared to those of control subjects.

Counts of C. albicans in saliva were done by requesting the patients to rinse their mouth for 60 seconds with 10 ml of sterile phosphate-buffered saline (PBS, pH 7.2) and expectorated into a plastic universal container. This oral rinse was centrifuged at 1700 x g for 10 min, and the deposit resuspended in 1 ml of sterile PBS. The concentrated oral rinse then inoculated on to appropriate media to assess CFU (colony forming units) per millilitre of rinse sample using a spiral plate prior to incubation. Histological evidence of C. albicans in OED was determined by reviewing histopathological records. All specimens had been stained with periodic acid schiff (PAS) stain.

Patient group for assessment of HCV and Helicobacter pylori antibodies

The study group comprised a total of 120 histologically confirmed OED patients (WHO, 1978; Kramer, 1980; Crissman & Zarbo, 1989; Speight et al, 1996) (64 male, 56 female, median age 54 years, range 29-80) and with no known peptic ulceration or other gastrointestinal disease, attending the Department of Oral Medicine of the Eastman Dental Institute For Oral Health Care Sciences between 1995 and June 1997 were selected for the study. Ethical committee approval to carry out this study was obtained.

Control groups

Control group for assessment of Candida albicans

The control group comprised of 131 healthy individuals (taken from another study) with no oral mucosal disease or local and or systemic disease likely to increase oral carriage of C. albicans. Age and gender were matched with the cases (55 males, 76 females, median age 23.1, range 21-53).

Control group for assessment of HCV antibodies

The control comprised 110 (86 female, 24 males, median age 39, and range 25-65 years), apparently healthy British dental health care workers. None had oral epithelial dysplasia.

Control group for assessment of H. pylori antibodies

Control group consisted of 25 healthy staff members without oral lesions and with no known history of peptic ulceration or other gastrointestinal disease, 75 with recurrent aphthous stomatitis (RAS) and 27 with oral dysaesthesia.

Hepatitis C virus antibodies assay

Serum IgG antibodies to HCV were assayed using two different third-generation enzyme-linked immunosorbent assays (ELISA) (Ortho Diagnostic Systems, Emmeryville, California: and Sanofi Diagnostic Pasteur, Marnes Ia Coquette, France).

Serological estimation of liver function

Serological estimation of levels of total bilirubin, total protein, albumin, hepatic alkaline phosphatase, alanine transaminase and gamma glutamyl transpeptidase were estimated in all patients using conventional methods (El-Kabir et al, 1993).

H. pylori antibody assay

Each subject was investigated for the presence of anti-IgG antibodies to Helicobacter pylori antigen using Helisal™ Rapid Blood test (Cortecs Diagnostics, Clywd, UK) test. A finger-prick sample of venous blood was diluted in the supplied buffer and placed on a nylon membrane coated with H. pylori antigen. The presence of specific IgG antibodies to H. pylori was then determined by an enzyme-linked immunosorbent assay (ELISA) technique. When appropriate substrate is added a reaction with the human IgG antibody occurs as a visible pink/red colour. The colour reagent also reveal human IgG on the control spot. The presence of single spot is therefore a negative result while two spots indicate a seropositivity to H. pylori.

Data analysis

Personal and clinical details of the subjects was collected using standard questionnaire (Appendix 3). The frequency of salivary Candida albicans carriage, HCV IgG antibodies and H. pylori seropositivity between patients and controls were analysed with the chi-square test, with *P* value considered significant if less than 0.05.

Results

Candida count among cases and controls

One hundred and sixty seven (74.8%) of the 223 patients with oral epithelial dysplasia and 42 (32.0%) of 131 controls subjects had C. albicans in their saliva. The frequency of Candidal carriage was significantly greater ($P<0.001$) in patients with OED than healthy controls (Table 4.23).

Candida count and Candida in oral epithelial dysplasia tissue

An increase in salivary Candida carriage was related to increase in Candidal invasion of the dysplastic tissue, those patients with high Candidal counts were more likely to show Candidal hyphae in tissue (Table 4.24).

Correlation between Candida counts and rate of malignant transformation

Clinical presentation of OED and presence of Candidal hyphae and subsequent malignant transformation is detailed in (Table 4.25). 6.6% of white patches shows Candidal hyphae in tissue while 10.6% of mixed type lesions shows positive Candidal hyphae in tissue. Following follow-up period of 27 to 274 months, 3 (50.0%) white patches with Candidal hyphae and 5 (62.5%) mixed lesions with Candidal hyphae transformed into squamous cell carcinoma compared with three (3/6) white patchs and three (3/8) mixed patch with Candida hyphae invasion but not transform to malignancy. Of note only one red patches was invaded by Candida which underwent malignant change.

Anatomical distribution of Candidal hyphae positive oral epithelial dysplasia

Table 4.26. shows the site distribution of Candidal hyphae-positive OED lesions. Slightly more lesions of the floor of mouth (26.6%), invaded by Candida compared with the ventral surface of tongue (20.0%), cheek (20.0%), the gingiva (13.3%). Lesions of the lateral and dorsal surfaces of tongue and retromolar area were least invaded.

Hepatitis C virus seropositivity

Results of 75 patients were available for analysis. Abnormalities in one or more liver function tests were found in 11 of the 75 OED patients (14.6%). These biochemical abnormalities included raised serum levels of aspartate transaminase in 4 patients

(36.3%) of those with abnormalities, raised alkaline phosphatase in 3 patients (27.2%), and raised serum bilirubin in two patients (18.1%), decreased protein levels in two patients (18.1%). Seven (6.3%) of the 110 healthy control subjects had serological abnormalities in liver function tests. These abnormalities included raised aspartate transaminase in three patients (42.8%), raised alkaline phosphatase in two patients (28.5%), and raised bilirubin in two patients (28.5%).

The results of the ELISA of serum samples from the patients with OED and controls are summarised in Table 4.27. Two (2.6%) of the 75 patients with OED had serological evidence of IgG antibodies to HCV. But none of the control group were HCV seropositive.

H. pylori seropositivity

The frequency of detection of serum IgG antibodies to H. pylori is detailed in Table 4.28. Twelve percent of OED cases were H. pylori seropositive compared with 24% of the healthy control subjects and 30.6% of patients with RAS, and 40.7% of patients with oral dysaesthesia. The frequency of carriage of H.pylori antibodies was slightly less than that of the control subjects.

Discussion

The association of leukoplakia with yeasts, in particular C. albicans, has long been known by means of mycological isolation technique, Jepsen and Winther (1965) found yeasts in oral leukoplakias, particularly lesions of the speckled type. Since then, a number of studies have confirmed that a considerable proportion of leukoplakias can have an associated Candida infection. The exact extent of such an association, however, is influenced by the technique used for isolation and identification of the yeasts and by the type of the leukoplakia from which the yeasts have been isolated (Roed-Petersen et al, 1970; Cawson & Binnie, 1980; Axell et al, 1984; Krogh et al, 1987). In the present

study 74.8% of the OED showed C. albicans. Furthermore the salivary Candida count are significantly elevated in cases of OED compared to healthy controls.

It is generally considered that non-homogenous leukoplakia have a higher probability than homogenous types for malignant transformation (Daftary et al, 1972; Axell, 1976; Pindborg, 1980; Banoczy, 1984; Silverman et al, 1984; Lind, 1987; Hogewind & van der Waal, 1988; Gupta et al, 1989; Saito et al, 1995; Bouquot, 1991) and the fact that the majority of non-homogenous leukoplakia may be invaded by C. albicans suggests a causal role of C. albicans in malignant transformation of oral leukoplakia (Renstrup, 1970; Cawson & Binnie, 1980; Axell et al, 1984). Renstrup (1970) found 61% of speckled leukoplakias showed Candidal invasion and of these, 71% had epithelial atypia. Only 3% of the homogenous leukoplakias showed Candida and none had atypia. Candida infection may be associated with increasing severity of epithelial dysplasia (Burkhardt & Seifert, 1977). Barrett and co-workers (1998) reported a significant positive association of fungal infection with moderate and severe epithelial dysplasia and subsequent biopsy reveals 21.9% of dysplasia which were infected with fungus worsened in histological severity as compared with 7.6% of dysplasia which were not infected. Roed-Petersen and co-workers (1970) found 40% of leukoplakias invaded by Candida showed atypia and that in 67% of leukoplakias showing atypia, Candida was present, but these workers still had reservations about the relationship of the fungus to the epithelial changes. Cawson and Binnie (1980) followed-up 30 patients with chronic hyperplastic candidosis (Candidal leukoplakia), and they found that 9 (30%) are known to have developed carcinoma. Of 153 other (non-Candidal) leukoplakias only 16 (10%) were known ultimately to have developed carcinoma. C. albicans is the species by far the most commonly isolated from oral leukoplakia, and the biotypes of C.

albicans associated with leukoplakia have been shown to differ from those isolated from normal oral mucosa (Krogh et al, 1987).

All of the Candidal leukoplakias where carcinomas developed within the period of observation had a speckled appearance. This suggests that speckled plaques are more likely to be due to or at least associated with candidosis than other causes and more likely to show dyskeratosis or to undergo malignant change. Pindborg and colleagues (1963) emphasised the importance of speckled leukoplakia as showing an abnormally high incidence of malignant change. In their later study, Pindborg and co-workers (1968) again noted that a majority of their patients who had speckled lesions developed carcinoma.

In the present study the frequency of Candida invaded OED was highest in mixed red and white lesions (10.6%), and floor of mouth was the commonest site (26.6%). Similar findings has been reported by others (Jepsen & Winther, 1965; Renstrup, 1970; Barrett et al, 1998). Epithelial atypia was found in 56% of the leukoplakias with Candidal invasion by Renstrup (1970), 40% by Cawson (1966). In the present study, however, 50.0% of white patches and 62.5% of mixed red and white lesions invaded by Candida underwent malignant transformation compared with 50.0% of white patches and 37.5% of mixed patch with evidence of Candida invasion but not transformed to malignancy within follow-up period of 27 to 274 months.

Pathogenic yeasts are able to catalyse the formation of *N*-benzylmethylamine from precursors at neutral pH, and this nitrosation potential is strain dependent (Krogh et al, 1987; Krogh, 1990). The products of nitrosation may be carcinogenic (Field & Martin, 1989). As yeasts are found in the oral cavity of 40 to 60% of adults, and OED occurs in much lower frequency, it is essential for the plausibility of the hypothesis of a causal role for yeasts in oral cancer that not all, but only specific, types of yeasts have a high

nitrosation potential (Krogh et al, 1987). The strains with a nitrosation potential exceeding 0.45μg *N*-benzylmethylamine/ 10^6 cells were the C. albicans biotypes 051, 147, 151, 153, 157 and 353 which are among the more rarely occurring biotypes in oral cavity (Krogh et al, 1987). Hyphae of C. albicans are frequently seen invading the outer epithelial layer in oral white and red patches that have known malignant potential, and sometimes in oral cancer itself. There continue to be controversy as to whether these Candida are secondary opportunistic invaders or of aetiological significance (Arendorf et al, 1983; Holmstrup & Bessermann, 1983). The hyphae may provide a method of transportation of precursors from saliva on the mucosal surface to the epithelial cells, in some cases including stratum spinosium, where the nitroseamine production may occur thus initiating malignant transformation (Arendorf et al, 1983; Krogh et al, 1987).

In this study there were no significant differences in the frequency of serum IgG antibodies to HCV between OED patients and control subjects. This is in sharp contrast to the findings of Nagao and colleagues (1995) who demonstrated that 24 of 100 patients with oral squamous cell carcinoma in Japan were HCV seropositive compared to 10.6% of control subjects, although in this Japanese study none of the lesions from the HCV infected patients expressed HCV.

The present study was the first to examine the HCV status of a substantial group of British patients with OED. No significant association between HCV and OED was found, furthermore none of the patients with OED had serological evidence of chronic hepatic disease and these data thus agree with previous studies of the prevalence of chronic hepatic disease in British patients with lichen planus (Scully et al, 1985; El-Kabir et al, 1993; Ingafou et al, 1998).

Although a positive association between HCV and premalignant lesions (e.g. lichen planus) has been detailed in some populations (Pawlotsky et al, 1994; Cribier et al,

1994; Mignogna et al, 1996), an explanation for the mechanism of this association has not been found (Lodi & Porter, 1997). A number of studies (Knudson, 1985; Vogelstein et al, 1988; Vogelstein et al, 1989; Nigro et al, 1989; Pawlotsky et al, 1994) indicated that cells in the oral region are continuously exposed to HCV, which in turn might increase the risk of genetic instability in the cells.

The apparent lack of an association between HCV infection and OED patients parallels the lack of association between lichen planus and HCV infection in British patients.

The prevalence of HCV infection in the UK is low (0.088% to 0.55%) (Crawford et al, 1994; Maclennan et al, 1994) compared with Southern European countries such as Italy (0.7% to 1.3%) (Chiaramonte et al, 1991; Arranconi et al, 1994). Of note the two HCV seropositive patients found in this study were of Italian and Spanish origin, countries known for high prevalence of HCV infection, thus this association might reflect the effect of local prevalence elsewhere.

Helicobater pylori is a very common pathogen, described to be associated with B cell lymphoma of the stomach and possibly gastric carcinoma. H. pylori is present in the dental plaque of the majority of individuals (Nguyen et al, 1995) but there is no data on its positive association with oral OED or malignancy.

In the present study, 25% of the healthy control subjects were anti-H. pylori IgG seropositive, a frequency that is consistent with available data in healthy persons in developed countries (Smoak et al, 1994; Webb et al, 1994; Veldhuyzen et al, 1994). In the UK, H. pylori infection is found in at least half of those over the age of 50 years (Drug and Therapeutic Bulletin, 1993) and the prevalence increases with age, from about 25% in those aged below 35 years, rising to about 50% in those above 55 years (Newell et al, 1990), also low socio-economic status and poor hygiene owing to person-to-person transmission of the organism by oral-faecal route (Neri et al, 1996).

Only a minority of people infected with H. pylori develop disease mainly duodenal or gastric ulcer (Webb & Forman, 1995) but the organism may also be a risk factor for gastric cancer (Blaser, 1992) and is classified as a class I carcinogen by the WHO. In this study the frequency of anti-H. pylori seropositivity was not significantly elevated in patients with OED compared with healthy controls. Indeed a significantly higher frequency of H. pylori antibodies was found among patients with recurrent aphthous stomatitis and oral dysaesthesia although the later may simply reflect the older age distribution of this group of patients. H. pylori DNA has been reported in the lesional tissue of 6 of 29 patients with ill-defined oral mucosal ulceration, however, this was not compared with the presence or absence of the bacterium orally in healthy control patients (Leimola-Virtanen & Happonen, 1995).

The immune response to H. pylori is complex with each infected person having a unique antibody profile (Megraud, 1995). Nevertheless about 2% of people fail to mount a response despite proven infection (Glupczynski et al, 1992). Serological tests remain positive for at least 6 months after bacteria have been eradicated (Kosunen et al, 1992) so a positive serological test may indicate past rather than current infection. Helisal Rapid Blood is an ELISA-based assay reported to have sensitivities and specificity of around 90% in comparison with histology, microbiology, rapid urease testing and ^{13}C urea breath testing as standard (Borody et al, 1996; Moayyedi et al, 1997). Hence this test (Helisal Rapid Blood) is probably appropriate, although it does not estimate levels of antibodies.

Khulusi and co-workers (1995) reported that gastric mucosal metaplasia (precancerous gastric lesion) presents to a greater extent in H. pylori positive subjects, and that the metaplasia declines following eradication of H. pylori. The relationship between H. pylori and gastric carcinoma per se is thought to be indirect with gastric carcinoma as a

long-term consequences of increased cellular turnover and the development of intestinal metaplasia (Correa, 1991; Parsonnet, 1993).

H. pylori itself does not appear to be either genotoxic or mutagenic. Infection is, however associated with increased cellular turnover, a chronic immune response accompanied by increased levels of reactive oxygen metabolites all conditions that could favour the development of cancer (Webb & Forman, 1995).

The current investigation does not support the etiological role of H. pylori in the development of OED, this is in agreement with another recent study (Masci et al, 1996) who found that H. pylori was rather infrequent in type III intestinal metaplasia and found even less often in cases of gastric dysplasia. This could be related to the condition of gastric dysplasia becoming non-hospitable for the bacteria. Likewise two recent reports (Abbas et al, 1995; Ricaurte et al, 1996) reported a lack of association between H. pylori and Barrett's oesophagus (a known precancerous lesion) and its subsequent malignant progression. Future studies to assess the lesional carriage of H. pylori in affected patients to more precisely define any association of H. pylori with OED might be warranted.

It is concluded that there is little evidence for an association between OED and infection with HCV or H. pylori. But infectious agents like C. albicans might have a role in causation and subsequent malignant transformation of OED.

CHAPTER 5

Long-term Outcome of Oral Epithelial Dysplasia

Outline

There is a generally held view that assessment of epithelial dysplasia in premalignant lesions is important as dysplastic lesions may undergo malignant change (Mincer et al, 1972; Banoczy & Csiba, 1976; Pindborg et al, 1977; Gupta et al, 1980; Silverman et al, 1984; Lumerman et al, 1995) and because it is felt that the chances of malignant transformation increase with increasing severity of dysplasia. Indeed a direct relationship has been established between the degree of epithelial dysplasia and the likelihood of the lesion progressing: this is clearly shown, for example, by Ostor (1993) in relation to lesions of cervix uteri and summarised in Table 1.16.

A number of studies addressing the relationship of OED and subsequent malignant transformation have been published (Mincer et al, 1972; Pindborg et al, 1977; Gupta et al, 1980; Silverman et al, 1984; Lumerman et al, 1995), which suggest that dysplastic lesions are more likely to proceed to carcinoma than non-dysplastic lesions, and also provide some evidence in support of the concept that the degree of dysplasia influences risk of subsequent malignant change. However, it still seems difficult to predict the behaviour of these lesions on the basis of clinical and histopathological techniques alone.

While many markers of possible malignant transformation have been reported, few have proven to be reliable or of any notable clinical relevance; furthermore there remain few detailed studies of long-term outcome of patients with OED. Hence the aims of the present chapter were to determine the rate of malignant change of OED in a group of patients followed-up for a number of years, and hence determine factors likely to influence this malignant change, and to describe the clinical characteristics of patients who developed recurrence of OED and second dysplastic lesions.

Materials and methods

Only 359 (57.0%) of the 630 patients with OED as described in chapter 2 had sufficient data for examination of their long-term outcomes. Patients were followed up for period of 2 to 274 months with a mean follow-up time of up to 40 months. Malignant transformation was considered if a histopathologically proven oral squamous cell carcinoma arose in the lesion which had histopathological evidence of OED; recurrence of a dysplastic lesion was considered if a second histopathologically proven dysplastic lesion developed at the same site during follow-up while second dysplastic lesions were considered when a new histopathologically proven OED lesion developed at a site different to that of the index dysplastic lesion.

The chi-square and Fisher's exact tests were used for statistical analysis of the results with *P* value considered significant if less than 0.05.

Results

Outcome of long-term follow-up of 359 patients with oral epithelial dysplasia

Twenty (5.5%) of the 359 patients developed a SCC of the oral mucosa over a period of 2 to 274 months with mean transformation time of 3.3 years (Table 5.1).

Demographic characteristics of 20 patients who developed squamous cell carcinoma subsequent to oral epithelial dysplasia

Nine of the patients with SCC (45.0%) were male and 11 (55.0%) female. The mean age at time of diagnosis of SCC was 52.6 years with range of (15 to 84 years). The mean age for males was 57.1 years (range 44 to 81 years), the mean age for females 49.0 (range 15 to 84 years). Eleven of the 20 SCC developed in patients older than 50 years. 75.0% of patients were Caucasian, 15% Pakistani or Bangladeshi, 10% Indian (Table 5.2).

Clinical type of oral epithelial dysplasia and subsequent development of squamous cell carcinoma

Nine of the 20 SCC (45%) developed from mixed lesions (erythro-leukoplakia), 4 (20.0%) from white lesions, 6 (30.0%) from red lesions and one (5.0%) from an area of ulceration (Table 5.3).

Site of oral epithelial dysplasia and subsequent development of squamous cell carcinoma

The floor of mouth (40.0%) was the most common site of malignant changes. But of note a significant number of gingival lesions transformed to invasive cancer ($P<0.01$). Malignant transformation was uncommon on the dorsal surface of tongue, alveolar ridge and retromolar area (Table 5.4).

Size of oral epithelial dysplasia and subsequent development of squamous cell carcinoma

Half of the tumours arose from dysplastic lesion with initial sizes of about 2 cm while 45% arose from lesions of one cm size. Only 5.0% of tumours arose from a lesions with a size of 3 cm (Table 5.5).

Histology of oral epithelial dysplasia and subsequent malignant transformation

Malignant transformation was more likely with lesions already having features of moderate or severe OED (Table 5.6).

Treatment method of oral epithelial dysplasia and subsequent development of squamous cell carcinoma

Three of the 20 SCCs (15.0%) arose from dysplastic lesions treated surgically. However, a significant number ($P=0.001$) of SCC arose in lesions treated with topical antifungals drugs, only 5 (25%) tumours originated in patients kept under clinical

review with advice to reduce tobacco smoking and alcohol drinking habits only (Table 5.7).

Tobacco smoking habits in patients with oral epithelial dysplasia with and without subsequent malignant transformation

The tobacco smoking and alcohol drinking habits of the patients are detailed in Table 5.8. Although 43.7% of the 20 patients were current tobacco smokers and 80.0% current drinkers of alcohol these features did not significantly differ from patients with an OED who did not develop a malignancy.

Demographic characteristics of patients with recurrent or additional oral epithelial dysplastic lesions

Sixty-three (17.5%) of the 359 patients had a recurrence of OED and 37 (10.3%) developed additional dysplastic lesions. The age, gender and ethnic-background of the affected patients detailed in table 5.9. The majority of these patients were over 50 years of age, there was a slight male predominance in those who had a recurrence of OED but slightly more females than males developed additional dysplastic lesions but the differences were not statistically significant.

Clinical and histological aspects of oral epithelial dysplasia lesion that recurred or had associated second dysplastic lesions

Details of the clinical type of the lesions that recurred together with their histological grading and method of treatment were detailed in Table 5.10. Recurrence of OED was most commonly associated with (erythroleukoplakias) lesions in contrast to second dysplastic lesions which usually arose in patients with an initial lesions having the appearance of (leukoplakia). The tongue, buccal mucosa and floor of mouth were the most common sites of recurrent or second OED lesions. Patients treated surgically

and/or with antifungals were at greater risk of showing recurrence or additional dysplasia.

Tobacco and alcohol consumption of the subjects with recurrent or second dysplastic lesions

The majority of patients with recurrence of OED, or with development of new OED lesions were current smokers of more than 21 cigarettes per day, likewise most were current drinkers of more than 59 units per week. Of note however, more wine drinkers developed recurrence of OED than other types of alcoholic beverages and more spirits drinkers developed second dysplastic lesions but these differences were not statistically significant (Table 5.11).

Tobacco and alcohol habits of patients with recurrence or second oral epithelial dysplastic lesions at last clinical appointment

It is evident the stopping smoking and drinking habits were more likely to reduce a risk of recurrence of OED ($P<0.006$). It is also clear that quitting these habits will reduce the risk of development of second dysplastic lesions ($P<0.002$) (Table 5.12).

Discussion

Establishing a histopathological diagnosis of OED is some times but not always straightforward (Pindborg et al, 1985; Abbey et al, 1995). Examination of the sequentially excised specimens may reveal a range of grades of dysplasia in various portions of the same OED specimen, suggesting that incisional biopsy samples may not be representative of the true nature of the lesion and histologic examination of the entire clinical lesion may be necessary for accurate grading of dysplastic lesions (Wright & Shear, 1985; Lumerman et al, 1995). Nevertheless in practical terms it was often be very difficult to excise entire OED lesions.

Several studies have been published on the biological behaviour of OED (Mincer et al, 1972; Banoczy & Csiba, 1976; Pindborg et al, 1977; Gupta et al, 1980; Silverman et al, 1984; Vedtofte et al, 1987; Gregg & Cowan, 1992; Lumerman et al, 1995).

The results of nine studies of long-term follow-up of OED are summarised in Table 5.13. The transformation rates varied between 5.5% to 36%, and it is evident that 86 (10.0%) of 854 reported patients developed invasive squamous cell carcinomas within follow-up periods of 20 years.

In this study 5.5% of patients with OED diagnosed histopathologically from incisional biopsies developed a subsequent SCC within 2 to 274 months after initial diagnosis, with a mean time to malignant transformation of 40 months. This frequency of development of malignancy is less than that of other studies (Mincer et al, 1972; Banoczy & Csiba, 1976; Pindborg et al, 1977; Silverman et al, 1984; Gregg & Cowan,1992; Lumerman et al, 1995) (see table 5.13).

In the present study SCC were commonly in patients who had had previous severe dysplasia in contrast to mild and moderate dysplasia, thus like other studies the lesion degree of dysplasia alone is not a reliable predictor of prognosis (Lind, 1987), indeed carcinoma can develop from an epithelium found by incisional biopsy to have only mild dysplasia. It is also important to note that not all dysplastic epithelial lesions inevitably progress to carcinoma (Mincer et al, 1972; Banoczy & Csiba, 1976; Pindborg et al, 1977).

Previous studies have suggested certain morphological characteristics are associated with the risk of malignant transformation of OED (Pindborg et al, 1961; Amagasa, 1981; Shibuya et al, 1986). In this study 45.0% of malignancies arose from areas of erythroleukoplakic lesions and 30.0% from red lesions a findings similar to the observations of Amagasa (1981).

Hence, while oral malignancy may arise from areas of pre-existing leukoplakia (known to have OED), it seems that malignancy is more likely to arise in lesions with areas of redness. The site of pre-existing OED has been suggested to influence malignant changes. For example the floor of mouth has been reported to be a possible risk site for malignant transformation (Kramer et al, 1978; Pogrel, 1979). In the present study the floor of mouth was among the most common sites of malignant transformation and does suggest that clinicians would be advised of risk of malignant transformation of OED of sublingual region.

As with some other studies females were more likely to be developed malignant transformation within an area of OED than males (Roed-Petersen, 1971; Banoczy & Sugar, 1972).

The biological behaviours of OED on patients who received no further surgical treatment after diagnostic biopsy is summarised in table 5.14. Of the 91 patients, 58.2% were remained the same after period of follow-up, but 17.5% improved or regressed. However, 15.3% were transformed to malignancy.

Results of the follow-up studies of patients whose lesions were surgically excised is summarised in table 5.15. Out of 126 patients incorporated in the analysis, 78.5% were cured, 15.8% recurred, but 5.5% transformed to malignancy.

From the previously reported data it appears to be beneficial to treat OED with surgery in as much as only 7 (5.5%) of 126 patients who had excision had invasive squamous cell carcinoma as compared with 14 (15.3%) of 91 patients who received no treatment. Furthermore, 99 (78.5%) of 126 were cured by surgery, 20 (15.8%) had recurrence of disease, and only 16 (17.6%) of 91 were improved without treatment.

The standard treatment of OED is surgical excision (Vedtofte et al, 1987). Recent studies have suggested that 13 cis-retinoic acid therapy is effective in preventing

progression of OED in patients with premalignant oral lesions (Hong et al, 1986; Lippman et al, 1993). Thus, the aforementioned cumulative surgical cure rate of 78.5% reported in the literature may be maintained with adjuvant use of 13-cis-retinoic acid in a very low dose and this should be investigated in a clinical trial. Recurrence of OED after surgical excisions occurred in 25.3% of the patients. This is lower than 35% recurrence rate of oral leukoplakia after surgical excision reported by Silverman and co-workers (1985). Previous studies of surgical treatment of potentially malignant lesions has also demonstrated that the risk of malignant development is not completely eliminated (Mincer et al, 1972; Stell et al, 1982; Vedtofte et al, 1987). The number of lesions prevented from malignant transformation by surgical excision is unknown however, a significantly lower risk of malignant development has been found in surgically treated OED than in a comparable group treated by antifungal drugs alone. The recurrence often located adjacent to the surgically treated lesions. The explanation may be that although the lesion was excised, pathologically changed tissue left behind. Difficulties in determine the exact margin of the lesion were particularly evident in the floor of the mouth and tongue and in Erythroleukoplakia. This may be the main reason for the high recurrence rate found in these lesions, another possible explanation for the recurrence of sublingual dysplastic lesions is that dysplastic tissue in the salivary duct is left behind after the surgical excision of the lesion (Browne & Potts, 1986). Indeed recently Daley and co-workers (1996) screened 1216 tissue sections of OED and SCC and found 26 (21.4%) exhibited ductal involvement, clinical follow-up on 23 cases showed that recurrence rate of dysplastic lesions with ductal involvement was equal to SCC. Concerning possible etiological factors, smoking was associated with 68.2% (43/63) of recurrence cases and 70.2% (26/37) of second dysplastic lesions. However, although tobacco and alcohol use may be associated with recurrence and development

of additional dysplastic lesions one cannot say with certainty that these habits promotes these changes and combination of several etiologically factors perhaps responsible for these changes.

Most patients didn't change their smoking or drinking habits after treatment only 15.2% of patients stopped smoking and drinking before development of recurrence and 16.2% before development of additional dysplastic lesions. These factors were significant in this study with risk of recurrence and development of second dysplasia significantly different between those who stopped these habits and those who continue these habits which indicated the need to encourage all patients to modify there habits when dysplastic lesions were diagnosed and treated. Chiesa and colleagues (1993) found modification of these habits is not significant predictor for development of relapses in operated oral leukoplakia while others found that cancer and other changes developed more frequently in those patients with leukoplakia who did not stop smoking and drinking (Banoczy, 1977). It is also reported that leukoplakia may disappear if patients stop smoking (WHO, 1978; Silverman et al, 1984; Chiesa et al, 1986).

The present study indicates that OED do not invariably progress to carcinoma. However, with current knowledge of potentially malignant lesion, regular follow-up examination as the only intervention are not justified for all lesions, in other hand as only a small proportion of minor degree of dysplasia develop malignancy it is redundant to treat all lesions surgically. The validity of using combined clinical and histological criteria for identification of a high risk lesions are supported by the actual number of carcinomas diagnosed in the follow-up period of the present study. The value of histological criteria as predictive of malignant development has been documented (Banoczy, 1977; Burkhardt & Maerker, 1978) but it must be emphasised that the correlation between histologic appearances and clinical behaviour is not always

a very close one. For example, there have been cases in which carcinoma has subsequently developed in lesions showing only the slightest degree of dysplasia, while on the other hand lesions showing severe dysplasia have persisted with little changes for years. The necessity of including criteria other than the histology is therefore evident. Taking into account all the uncertainties and anomalies of behaviour of these lesions, it does seem justifiable to consider in general term that the degree of dysplasia is linked to the degree of probability of the development of malignancy, and the patient should be managed accordingly. In any event, it is a safe rule to advice the complete removal, whenever possible, of all lesions showing more than slight degrees of dysplasia. Also, it should again be emphasised that there are certain 'high-risk' sites especially floor of mouth and ventral surface of tongue, where greater attention should be given to even lesser degrees of dysplasia.

Although the reason for the high frequency of additional dysplastic lesions is not clear, the most convincing hypothesis is the 'field cancerization' theory originally proposed by Slaughter and co-workers (1953), several carcinogen act on area of squamous mucosa, producing an irreversible changes toward cancer extensively, but not uniformly. Cancer develops first where the carcinogenic stimuli have been maximal and cancers develop later in areas subjected to lesser carcinogenic stimuli. It is also confirmed that dysplastic changes are multicentric in which they reported presence of skip areas in the dysplastic epithelium where the intervening epithelium either normal or hyperplastic (Wright & Shear, 1985; Lumerman et al, 1995).

It appears that much of the dispute about the progression of epithelial dysplasia and carcinoma-in-situ involves the problem of clearly identifying the dividing line between reversible atypia and irreversible carcinoma-in-situ. Although the histologic characteristics of epithelial dysplasia have been well defined (WHO, 1978; van der

Waal, 1986), the interpretation of these criteria depends on extent and severity of changes and, therefore, is largely subjective (Pindborg et al, 1985; Abbey et al, 1995). Smith & Pindborg (1969) suggested a step toward clarification of this problem by concentrating the observer's attention on one photographically standardised microscopic feature at a time and secondly, by enabling the observer to assess each features individually and allocate a weight score to each. In this way, observer bias or errors may be considerably reduced.

It is concluded that follow-up of OED patients from two to 274 months with a mean follow up of 40 months, reveals 66.0% were clinically free of disease and, 17.5% exhibited recurrence of the dysplastic lesions. Second dysplastic lesions were developed in 10.3% of cases. Squamous cell carcinoma developed in 5.5% of patients. The high risk of malignant transformation of OED seems to be related to patients older than 50 years when lesions were on the floor of mouth. Red or mixed lesions with severe dysplastic changes. The high risk for development of recurrence and second dysplastic lesions seem to be related to patients older than 50 years of age. Severe dysplastic lesions of the tongue. Erythroleukoplakic or leukoplakic lesions following surgical excision or use of antifungal therapy.

CHAPTER 6

Oral Epithelial Dysplasia among Non-smokers and Non-drinkers

Outline

The role of tobacco and alcohol as the two major risk factors of oral cancer (IARC, 1986,1988; MacFarlane, 1993) and precancers (Pindborg, 1972,1980; Banoczy, 1982; Salem et al, 1984; Zaridze et al, 1986) have been well documented and their effect upon OED has been detailed in chapter 4.

There is however a paucity of information on OED developing in persons who have never used tobacco and alcohol, thus the aims of this chapter was to detail the clinical features and long-term outcomes of OED among tobacco and alcohol non-users and to compare these to those of users of tobacco and alcohol.

Materials and methods

The hospital records of the 630 patients detailed in chapter 2 were reviewed. Two groups of patients were selected based upon tobacco and alcohol usage: 37 patients who had neither smoked tobacco nor drank alcohol and 419 patients who both smoked tobacco and drink alcohol. Demographic and other data including age, gender, site of the primary lesions, histopathology, treatment, and disease behaviour, were examined using EPI 5 (epidemiological computer package) (Dean, 1992).

Statistical procedures were carried out using SPSS programme (SPSS, 1993), Chi-squared test were used to compare between the two groups (Swinscon, 1996) and the results were considered significant when P was less than 0.05.

Results

Patient demographic

Thirty-seven (5.8%) of 630 patients with oral epithelial dysplasia had never taken tobacco and alcohol. There was a male to female ratio of 1:1 in the group who neither

smoked tobacco or drank alcohol. Most of the patients were in the 6th decade. There were no significant differences in the gender or race of patients with OED who were non-smokers and non-drinkers compared with those who both smoked and drank (Table 6.1).

Site of oral epithelial dysplasia

There was no significance difference in the distribution of oral sites of OED in the non-users and users of tobacco and alcohol. Although more of the users had lesions of the retromolar area and commissure (Table 6.2).

Clinical type of primary lesion

There was no significance difference in the clinical presentation of OED in the non-smokers and non-drinkers compared with the other patients with OED (Table 6.3).

Histology of oral epithelial dysplasia

The histology of OED in patients who neither smoked tobacco or drank alcohol was detailed in (Table 6.3). There were more mild dysplastic lesions (51.3%) in the non-users than users of tobacco and alcohol (35.3%), but the users group had more moderate dysplasia (30.5%) than the non-users group (18.9%).

Treatment and clinical behaviour of the primary dysplastic lesions

Fifty-four percent of non-users and 41.2% of users were surgically treated, the remaining patients being managed with other treatment modalities (Table 6.4). The rates of OED recurrence and development of second OED were not statistically significant in the two groups although second OED lesions were more common in the non-users (13.5%), than users (7.7%) of tobacco and alcohol. Transformation to

invasive SCC was more common in non-users (10.8%) than users of tobacco and alcohol (4.0%).

Characteristics of squamous cell carcinoma in tobacco/alcohol users and non-users

Details of the SCC in both groups are presented in Table 6.5. Among the non-users, the gingiva was a common site of malignant transformation 50% compared with none of the tobacco and alcohol users. Forty-six percent of the tobacco and alcohol users however, had SCC of the floor of mouth compared to 25% of non-users.

Discussion

The current study has investigated the demographics, clinical, histological and prognostic aspects of OED in patients who had not been exposed to both alcohol and tobacco, the two widely recognised risk factors for OED and invasive SCC. The presence of OED in patients who do not smoke tobacco or drink alcohol would suggest that risk factors other than alcohol and tobacco may exist.

In the present study, several demographic observations of the disease population were made. Only 5.8% of the group of 630 studied patients were identified as non-users of both tobacco and alcohol. This is higher than the figure of 3.4% reported by Hodge and co-workers (1985) in one of the few studies on oral SCC solely in non-users of tobacco but less than the figure of 31% reported by Wey and co-workers (1987). Perhaps the acknowledged contributions of tobacco and alcohol, and the prevalence of their use within western society obscures the attention given to other etiologic factors in the development of oral precancers and cancer.

Just over 50% of the patients who neither smoked tobacco or drank alcohol were female, this lack of gender differences is not unsurprising in view of the results of

chapter 2, but perhaps contrast with Daly's study (1982) at Centre Claudius Regaud, France of a series of 95 women with head and neck cancer in whom up to 82% of the lesions lacked identifiable epidemiological factors, including tobacco and alcohol.

The number of cases in this study were adequate for detailed analysis of the differences between adjacent locations within the oral cavity with respect to the percentage of non-smokers and non-drinkers. Among sites of initial presentation, the non-users had fewer lesions involving the floor of the mouth. This observation supports the theory that tumourogenesis may be distinctly related to the direct effects of tobacco and alcohol at the site where the greatest dose is supplied. Furthermore, the carcinogens in alcohol, thought to be promoters of oral cancer, are reportedly delivered in a greater concentration to the floor of the mouth compared to the relatively dilute carcinogens of tobacco smoke (Decker & Goldstein, 1982). In a combined exposure, tobacco saliva suspensions which collect within the "pooling area" of the mouth, may allow for a much more substantial mucosal injury (McGuirt, 1983).

A difference was noted between users and non-users when comparing the histologic severity of the OED. Of the tobacco and alcohol non-users 51.3% were diagnosed as mild dysplasia and only 27.0% were in the severe category, while among the tobacco and alcohol users 35.3% were diagnosed as mild dysplasia with 22.9% were severe dysplasia. In contrast Kaugars and co-workers (1989) reported 66.7% mild dysplastic lesions among smokeless tobacco users and 5.4% were severe dysplastic lesions.

A previous investigation stated that an epithelial dysplasia associated with snuff had a better clinical course and less severe histologic grade than an unrelated one (Mincer et al, 1972). Confirmation of this finding toward a milder degree of epithelial dysplasia was noted in a study by Schroeder and colleagues (1987) which identified seven mild epithelial dysplasia (20.5%) among 34 smokeless tobacco users with mucosal lesions.

In the present study tobacco and alcohol usage were not good prognostic indicators of early behaviour of the primary dysplastic lesions as frequency of recurrence after the initial treatment was similar in the both groups. This may reflect the relatively small series of patients examined, however, the results do stress the importance of recognising the prevalence of recurrent disease in both populations.

There was a trend of an increased risk of second OED lesions in the non-users of tobacco and alcohol compared with users. Tepperman and Fitzpatrick (1981) has presented an explanation of development of second primaries in tobacco and alcohol users, in which the chronic mucosal irritation by the agents "primes" the entire oral mucosa for neoplasia even before the clinical appearance of disease." Therefore, the user population would be at a greater risk for the development of multiple primaries even upon subsequent cessation of their habits. The present results do not support such a concept.

Studies report progression of OED to SCC at rates ranging from 6.6 to 36.4% after mean follow up periods of 1.5 to 8.5 years (Mincer et al, 1972; Banoczy et al, 1976; Pindborg et al, 1977; Gupta et al, 1980; Silverman et al, 1984; Lumerman et al, 1995). In the present study 4.0% of the users group and 10.8% of non-users transformed to malignancy after follow-up period between one year to 23 years with mean 3.3 years. Results of other studies have shown that malignant transformation of leukoplakias is more common in non-smokers than in smokers (Silverman et al, 1984; Lind et al, 1987; Hogewind et al, 1989).

As oral SCC and OED can develop in the absence of tobacco and alcohol a number of non-tobacco and non-alcohol etiological factors must be at play, these may includes chronic mechanical irritation (Thumfart et al, 1978; Decker & Goldstein, 1982), or

sideropenic anaemia (Ahlbom, 1936; Larsson et al, 1975). Other dietary deficiencies (Wynder & Klein, 1965; Hormozdiari et al, 1975; Graham, 1984), immunosuppression (Toto et al, 1981) or chemical irritants may also be an etiological factors. Infectious agents such as Candida albicans (Cawson & Binnie, 1980) and human papilloma virus (Shroyer et al, 1993; Miller et al, 1996) may play some role, but as noted in chapter 4 the evidence is somewhat tenuous.

CHAPTER 7

Summary and recommendations

To generate a statistically significant number of cases upon which valid conclusions could be based, data were gathered from 630 patients with oral epithelial dysplasia (OED). It was established that OED in a UK patient group is usually of a mild grading and very rarely carcinoma-in-situ. Males are more commonly affected than females. The peak frequency of development of OED for males and females was in the 6th decade. Most OEDs were diagnosed in people of Caucasian origin. The most common sites of involvement are the tongue, buccal mucosa, and floor of mouth, although the buccal mucosa is the most frequently involved site among Asians. The sites with the greatest percentages in the severe dysplasia are the floor of mouth and lateral border of tongue. Just over half of all OED are white patches, 44.1% are mixed lesions and 1.9% are red patches. There is no significant different between patients younger or older than 35 years of age with regard to race, site of lesion, or clinical type of OED lesions.

The experience and knowledge of OED and related lesions of UK dental practitioners and relevant aspects of referral of patients with PMLs by UK practitioners were investigated. While most of the referral letters of patients with potentially malignant lesions were legible and provided some details of the patient's, gender, date of birth, address, the majority lacked data on contact telephone number and patient's social and medical history. Degree of urgency was only indicated in 7.8% of the letters. Dental practitioners were the main source of referral (47.5%) followed by general medical practitioners (38.5%).

Almost all dentists attended postgraduate meetings usually BDA or Department of Health-funded meetings. 45.0% had attended specific courses on oral malignancy and premalignancy. 68.7% (88/128) of dentists saw more than 10 cases of oral malignancies and 13.8% examined or witnessed less than 10 cases of potentially

malignant lesions during their undergraduate studies. Few realised that the carcinoma would be speckled in appearance. Only 60.4% of respondents indicated that tobacco and alcohol use were the principle cause of oral SCC and 19.7% suggested that HIV disease was a risk factor for oral SCC. 29.7% of dentists routinely recorded the tobacco or alcohol use of patients and 93.9% offered advice to the patients regarding modification of these habits. The vast majority of dentists (82.9%) were convinced of the efficacy of screening, and the majority suggested that screening at an interval of 3 to 6 months was an effective means of reducing the frequency of potentially malignant and malignant oral lesions.

GDPs in the UK have some knowledge and experience of oral premalignant and malignant lesions, but they fail to provide appropriate preventive advice and may delay referral of patients to appropriate centres.

In view of the gradual rise in oral malignancy in the UK, there is an increased need for general dental practitioners to have some appropriate training, to recognise the signs and symptoms of oral premalignancy and malignancy, and be aware of the appropriate early management of patients with such oral lesions. Undergraduate and postgraduate education of general dental practitioners in the UK with regard to the recognition and management of oral premalignancy and malignancy is thus still warranted. To increase the amount of appropriate information provided by the referring clinicians and to reduce unnecessary delay in the patient management, a standard referral form is proposed.

Despite the malignant potential of OED the current state of knowledge regarding aetiological risk factors associated with OED is limited. To evaluate risk factors for

(OED), data were collected in a case-control study based on 630 patients and 643 controls. Conditional logistic regression was used to calculate measures of association and statistical significance. Non-filter cigarette smoking is a very specific predictor of OED, as too is alcohol consumption, and the two habits compound one another in the overall risk of disease. The increased risk of OED from smoking is largely attributable to heavy smoking (20 cigarettes per day or more), especially non-filter cigarettes relative to non smokers. Tobacco cessation was associated with a significant decline in the risk of OED. For alcohol consumption the association was considerably stronger for drinkers of fortified wines and spirits.

The effect of alcohol or tobacco was also considered specifically in non-consumers of the other product. Among persons who had never consumed alcohol the risks of OED increased with tobacco smoking of more than 20 cigarettes per day particularly non-filter cigarettes and the risk of OED declined following smoking cessation, with ex-smokers of 10 or more years demonstrating no excess risk relative to non-smokers. In the non-smokers, consumption of alcohol was not a significant predictor of OED.

Sufficient subjects were available to allow an analysis of the effects of alcohol and tobacco on sub-sites within oral cavity. The relative risk associated with tobacco smoking appeared to be highest for OED in the labial mucosa and floor of mouth for males and tongue and floor of mouth for females, whereas the lowest relative risk was found in the buccal mucosa. While alcohol drinking is not a significant predictor of specific OED sub-sites in both males and females.

A further study to clarify the etiological significance of serum folate, red blood cell folate and vitamin B12 was conducted. A significant association between red blood cell

folate, serum folate and risk of OED was found and these factors interact with other risk factors in the aetiology of OED. Infectious agents such as Candida albicans may have an aetiological significance in the OED but no significant association was found with regard to Helicobacter pylori and hepatitis C virus.

Based on these findings the following recommendations are suggested:

1. Research to investigate the effectiveness of supplementation of folate and vitamin B12 in reducing the incidence of OED and its subsequent malignant transformation.
2. Investigation of biological and other clinico-histological factors which have any etiological or prognostic significance.

Follow-up of OED patients from two to 274 months with a mean follow up of 40 months, reveals 66.0% were clinically free of disease and, 17.5% exhibited recurrence of the dysplastic lesions. Squamous cell carcinoma developed in 5.5% of patients within two months to 274 months. The mean transformation time was 3.3 years. 45.0% of patients were men and 55.0% were female. The mean age at time of diagnosis of squamous cell carcinoma was 52.6 years with range of 15 to 84 years. Second dysplastic lesions were developed in 10.3% of cases. Risk of malignant transformation of OED seems to be high in patients older than 50 years of age when lesions were on the floor of mouth and red or mixed lesions with severely dysplastic changes. The risk of development of recurrence and second dysplastic lesions seem to be greater in patients over 50 years of age, in severely dysplastic lesions of the tongue, erythro-leukoplakic or leukoplakic lesions following surgical excision, and when antifungal therapy had been used.

Oral epithelial dysplasia may occur in a significant population of non-users of tobacco and/or alcohol. In this group the location of the primary disease may involve sites not

commonly associated with exposure to specific agents. The development of second dysplastic lesions among non-users is a common event.

Therefore the following recommendations are suggested:

1. Identify reliable markers for risk assessment of malignant transformation of OED to augment histopathological evaluation.
2. Research on chemopreventive drugs which may protect high risk patients from malignant transformation, recurrence and development of second dysplastic lesions.
3. More effort to inform the population and general practitioners of the risk of lesions showing OED, emphasising that tobacco and alcohol habits do correlate with malignant transformation, recurrence and development of second dysplastic lesions.

In summary this study has examined a number of pertinent aspects of OED. Clearly much has been learnt, however there is considerable scope to increase the knowledge regarding the risk factors, detection and management of OED in the UK and other countries.

CHAPTER 8

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CHAPTER 9

Tables

Table 1.1. Ultrastructural characteristics of dysplastic lesions and carcinomas of squamous epithelium.

Alterations of nuclei and nucleoli
Enlargement
Multiple nucleoli
Alterations of cellular organelles
Reduction in numbers of specific and highly organized organelles (mitochondria, keratinosomes)
Numerical reduction of mitochondrial lamellae
Myelin-like figures, multiple membrane complexes and mitochondrial inclusions
Increase in numbers of polyribosomes
Aggregation of cisternae of the endoplasmic reticulum
Increase in numbers of microfilaments
Loss or decrease in numbers of keratohyaline granules
Alterations of tonofibrils
Decrease/increase in numbers
Premature keratosis
Formation of coarse clumps
Perinuclear spirals
Dyskeratotic necrosis
Fibrillar dyskeratosis
Formation of immature keratin with remnants of organelles
Lack of stable membrane formation
Organoid keratosis (keratin pearls)
Alterations of the cellular membrane
Spongiosis
Formation of cytoplasmic protrusions (microvilli)
Pinching-off of cytoplasmic processes
Reduction in the number of junctions
Alterations of desmosomes
Reduction in numbers
Reduction in length
Predominance of simple forms
Lack of intercellular contact layers
Reduction of attachment plaque formation
Lack of insertion of tonofilaments
Loss or extraction of desmosomes
Intracytoplasmic desmosomes
Alterations of the basement membrane
Multilamellar
Presence of atypical material incomplete hemi-desmosomes
Thinning
Gaps
Cytoplasmic protrusions

(After Meyer-Breiting & Burkhardt, 1987).

Table 1.2. Prevalence of oral leukoplakia

Author(s)	Country	Number of persons examined	Prevalence %
Gerry et al, 1952	Guam	2004	0.4
Mehta et al, 1961	India	4734	3.5
Bruszt, 1962	Hungary	5613	3.6
Atkinson et al, 1964	New Guinea	3996	8.1
Pindborg et al, 1965a,b ,1966	India	30,000	1.6-3.3
Manghi et al, 1965	India	2004	6.5
Zachariah et al, 1966	India	5000	2.4
Pindborg et al, 1968	New Guinea	1226	4.6
Wahi et al, 1970	India	7286	5.2
Mehta et al, 1971	India	50,915	0.2-4.9
Mehta et al, 1972a	India	101,761	0.7
Smith et al, 1975	India	57,518	11.7
Bhonsle et al, 1976	India	5449	1.6
Axel, 1976	Sweden	20,333	3.6
Wilsch et al, 1978	Germany	4000	2.2
Lay et al, 1982	Burma	6000	1.7
Rodriguez et al, 1983	Cuba	749	2.1
Mani, 1985	Saudi Arabia	674	1.9
Bouquot and Gorlin, 1986	USA	23,616	2.9
Zaridze et al, 1986	USSR	1569	8.0
Axell et al, 1987	Sweden	20,333	0.7-24.8
Hogewind and van der Waal, 1988	The Netherlands	1,000	1.4
Axell, 1990	Thailand	234	1.3
Axill, 1990	Malaysia	233	1.3
Banoczy and Rigo, 1991	Hungary	7,820	1.3
Ikeda et al, 1995	Cambodia	1319	1.1
Reichart et al, 1996	Germany	1000	0.9

Table 1.3. Frequency of oral leukoplakia in different sites of the mouth in three countries

Site	Percentage of cases *India	⊕Denmark	♦Hungary
Lips	5.4	7	6.8
Commissures	24.9	24.5	37.5
Cheek	47.2	32	25.3
Gingiva	0.4	ϕ13	ϕ6.7
Alveolar mucosa	0.6		
Palate	19.4	4.5	9.8
Tongue	1.9	7.3	8.2
Floor of mouth	0.04	6.4	5.7

* Data extracted from Mehta et al, (1971) and Mehta et al, (1972a)
⊕ Roed-Petersen and Renstrup (1969).
♦ Banoczy (1977).
ϕ Gingiva and alveolar mucosa combined.

Table 1.4. Rates of regression of oral leukoplakia

Authors (years)	Country	Observation period (years)	Rate of regression percentage
Pindborg et al, (1968)	Denmark	1-9	37.4 Partial or total
Mehta et al, (1972b)	India	10	45.3 Total
Mehta and Pindborg (1974)	India	5	26.1-43.7 Partial or total
Banoczy and Sugar (1975) *	Hungary	Mean 8.5	25.3
Silverman et al, (1976)	India	2	31.6

* This group of patients were treated but the other investigations show rates of spontaneous regression

Table 1.5. Malignant transformation rates of oral leukoplakia

Authors	Year	Country	Patients No.	Observation period (in years)	Malignant transformation
Sturgis and Lund,	1934	USA	143		13.0%
Leonardelli and Talamazzi,	1950	Italy	268		19.8%
Sugar,	1957	Hungary	86	1-11	5.8%
Skach,	1960	Czecho-slovakia	71	3-6	1.4%
Mela,	1966	Italy	141	3-11	3.5%
Pindborg et al,	1968	Denmark	248	3-9	4.4%
Einhorn and Wersall,	1967	Sweden	782	1-20	4.0%
Silverman,	1969	USA	117	1-11	6.0%
Roed-Peterson,	1971	Denmark	331	>1	3.6%
Banoczy,	1972	Hungary	500		13.2%
Pindborg,	1975	India	170	7	3.5%
Silverman,	1976	India	4762	2	0.12%
Banoczy et al,	1977	Hungary	670	30 mean 9.8	5.9%
Maerker and Burkhardt,	1978	Germany	200		12.0%
Kramer et al,	1978	UK	63	1-19 mean 4.2	38.1%
Gupta et al, Bhavnagar district Ernakulam district	1980	India	 360 410	 1-10 mean 7 1-10 mean 7	 0.3% 2.2%
Silverman et al,	1984	USA	257	1-39	17.5%
Bouquot et al,	1988	USA	463		10.3%

Table 1.6. Prevalence of erythroplakia

Author (s)	Country	Number of persons examined	Prevalence %
Metha et al, 1971	India	50,915	0.02
Lay et al, 1982	Burma	6,000	0.1
Shafer and Waldron,1975	USA	65,000*	0.09

* Surgical specimens in two dental schools over a 15 and 24 years

Table 1.7. Prevalence of oral lichen planus

Author (s)	Country	Number of persons examined	Prevalence (%)
Bouquot and Gorlin, 1986	USA	23,616	0.12
Lay et al, 1982	Burma	6,000	0.4
Pindborg et al, 1965a,b, 1966	India	35,000	0.02-0.4
Metha et al, 1971	India	7,639	0.1-1.5
Pindborg et al, 1972	India	7,639	0.1-1.5
Axel and Rundquist, 1987	Sweden	20,333	1.9
Axell et al, 1990	Thailand	234	3.8
Axell et al, 1990	Malaysia	233	2.1
Banoczy and Rigo,1991	Hungary	7,820	0.08
Ikeda et al, 1995	Cambodia	1,319	1.8

Table 1.8. Development of squamous cell carcinoma in patients with oral lichen planus (LP): literature review

Authors	Year	Country	Patients with LP No	Patients develop oral cancer	Frequency of malignant change (%)	Observation period (years)
Willinger,	1924	Germany	20	2	10.0	-
Montgomer & Culver,	1929	UK	17	1	6.0	1-9
Schuermann,	1939	Germany	310	2	0.6	-
Deschaume et al,	1957	France	50	5	10.0	-
Sugar & Banoczy,	1959	Hungary	36	1	3.0	11
Warin,	1960	UK	53	5	9.0	1-10
Altman & Perry,	1961	USA	128	1	0.8	6-10
Andreasen & Pindborg,	1963	Denmark	115	0	0	2-5
Rhode,	1966	Germany	207	6	3	-
Janner et al,	1967	Germany	585	9	1.7	1-24
Abramova,	1968	Russia	436	5	1.1	5-8
Cawson,	1968	UK	138	1	0.7	-
Shklar,	1972	USA	600	3	0.5	1-15
Fulling,	1973	Denmark	225	1	0.4	3.6
Kovesi & Banoczy,	1973	Hungary	274	1	0.4	1 > 10
Silverman et al,	1974	USA	200		2.5	1-8
Holmstrup & Pindborg	1979	Denmark	8	1	12.5	0.4-6.5 Median 3.6
Gupta et al,	1980	India	-	-	0.3	-
Vaskovskaya & Abramov,	1981	Russia	725	29	4.0	-
Silverman et al,	1985	USA	570	7	1.2	6 months- 10 years
Murti et al,	1986	India	722	3	0.4	5.1
Holmstrup et al,	1988	Denmark	611	9	1.5	1-26 Mean: 3.6
Salem,	1989	Saudi Arabia	72	4	5.6	3.2
Sigurgeirsson & Lindelof,	1991	Sweden	2071	8	0.4	9.9
Silverman et al,	1991	USA	214	5	2.3	0.5-> 10 Mean: 7.5
Voute et al,	1992	Holland	113	3	2.7	0.5-22 Mean7.8
Barnard et al,	1993	UK	241	8	3.3	1-10
Vescovi and Gennari,	1996	Italy	71	3	4.22	5.0
Markopoulos et al,	1997	Greece	326	4	1.3	6 months-10 years
Lo Muzio et al,	1998	Italy	263	14	5.3	1-10 years

(Modified from Barnard et al, 1993)

Table 1.9. Prevalence of oral submucous fibrosis

Author(s)	Country	Number of cases examined	Prevalence %
	South Africa		
Shear et al, 1967	Pretoria and Johannesburg	1,000	0.5
Dockrat and Shear, 1969	Durban	1,200	0.6
van Wyk et al, 1977	Capetown	-	1.3
Seedat and van Wyk, 1988	Durban	2,058	3.4
	India		
Pindborg et al, 1965b	Bombay	10,000	0.5
Pindborg et al, 1965a	Lucknow	10,000	0.5
Pindborg et al, 1966	Bangalore	10,000	0.18
Zachariah et al, 1966	Trivandrum	5,000	1.2
Pindborg et al, 1968	Ernakulam	10,287	0.4
Pindborg et al, 1968	Srikakulam	10,169	0.04
Pindborg et al, 1968	Bhavnagar	10,071	0.2
Pindborg et al, 1968	Darbhanga	10,340	0.07
Pindborg et al, 1968	Singhbhum	10,048	0
Wahi et al, 1970	Agra	7,286	0.6
Metha et al, 1972	Pune	101,761	0.03
Bhonsle et al, 1976	Goa	5,449	0.04
Gupta et al, 1980	Ernakulam	-	0.65
Gupta et al, 1980	Bhavnagar	-	0.06
Lay et al, 1982	Burma	6,000	0.1
Ikeda et al, 1995	Cambodia	1,319	0.2

(After Johnson et al, 1993)

Table 1.10. Malignant transformation in oral submucous fibrosis

Authors & year	Country	No of patients	Observation period (years)	Transformation rate
Pindborg et al, 1984	India	66	1-15	4.5
Murti et al, 1985	India	66	1-17	7.6

Table 1.11. Odds ratio and 95% confidence limits for studies on oral cancer where exposure to tobacco/betel quid is clearly defined

Principal Author	S		BQ+T+S		BQ+T		BQ+S		BQ	
	OR	95%.C.I.	OR	95% C.I.	OR	95% C.I.	OR	95% C.I.	OR	95% C.I.
Sankaranarayanan, 1989, 1990	4.1	2.6, 6.3	13.0	8.6, 19.5	8.8	6.0, 12.7				
Jafarey, 1976	5.4	4.2, 7.2	20.2	15.3, 27.3	13.7	10.6, 17.8	21.0	15.8, 28.3	3.6	2.3, 5.2
Hirayama 1966	3.4	2.0, 6.1	14.1	8.8, 24.1	9.1	5.8, 15.0	3.2	1.6, 6.8	1.2	0.5, 2.5

[Reproduced from Thomas and Wilson (1993)]

S	Smokers only
BQ+T+S	Betel quid+Tobacco+Smoking
BQ+T	Betel quid +Tobacco
BQ+S	Betel quid + Smoking
BQ	Betel quid

Table 1.12. Odds ratio and 95% confidence limits for studies where exposure to tobacco/betel quid and sub-site is clearly defined

Principal Author	Site	BQ+T+S		BQ +T		S		BQ	
		OR	95% C.I.	OR	95% C.I.	OR	95% C.I.	OR	95% C.I.
Chandra, 1962	Buccal	4.2	2.7, 6.7	4.3	2.9, 6.2	0.8	0.5, 1.4	1.2	0.8, 1.9
Hirayama, 1966	Buccal	31.9	16.1, 84.3	21.9	11.3, 56.9	5.2	2.4, 14.5	2.1	0.6, 6.9
Hirayama, 1966	Buccal + Lip	33.4	16.9, 88.3	23.7	12.2, 61.4	5.4	2.5, 15.1	2.8	0.9, 8.8
Sankaranarayanan, 1989	Buccal + Lip	20.5	12.1, 38.5	13.9	8.4, 25.3	4.0	2.0, 8.1		
Hirayama, 1966	Tongue ant 2/3	7.1	3.8, 15.4	2.6	1.4, 5.7	1.7	0.7, 4.0	0.5	0.06, 1.6
Sankaranarayanan, 1990	Tongue ant 2/3 + floor of mouth	6.6	3.7, 12.2	5.4	3.2, 9.8	4.7	2.5, 9.0		
Hirayama, 1966	Gingiva	13.8	4.8, 107.8	12.7	4.6, 97.8	8.0	2.5, 63.6	3.5	0.6, 29.8
Sankaranarayanan, 1989	Gingiva	17.1	8.6, 41.3	11.9	6.1, 27.9	4.2	1.6, 11.1		

BQ+T+S Betel quid+Tobacco+Smoking S Smokers only
BQ+T Betel quid +Tobacco BQ Betel quid
[Reproduced from Thomas and Wilson (1993)]

Table 1.13. Relative risks (RR) associated with alcohol intake in cohort studies

Authors	Cohort	No	Site	Exposure category	RR[¢]
Hirayama, 1989	General population				
		59	Mouth	Non-drinker	1.0
				Occasional	0.7
				Daily	2.3
		28	Pharynx	Non-drinker	1.0
				Occasional	0.9
				Daily	2.4
		438	Oesphagus	Non-drinker	1.0
				Occasional	1.0
				Daily	2.3
Kono et al, 1986	Physicians	18	All subsites combined*	Occasional	1.0
				< 54 ml/day	1.5
				> 54 ml/day	8.6
Kato et al, 1992	General population	75	All subsites combined*	Non-drinker	1.0
				< 30 ml/day	1.2
				> 30 ml/day	5.4
Klatsky et al, 1981	Participants in multiphasic health check-up programme	15	Mouth, pharynx, oesphagus	Non-drinker	1.0
				1-2 drinks/day	0.0
				3-5 drinks/day	2.5
				> 6 drinks/day	4.0

*Cancer of mouth, pharynx, oesphagus combined.
[¢]RR Relative risks

Table 1.14. Relative risks (RR) of oral cancer associated with alcohol intake from the selected case- control studies

Authors	Type of control	Exposure category	Cancer site and (No.) Relative risk			
Franceschi et al, 1990	Hospital	Drink[#]	Mouth (157)	Pharynx (134)	Larynx (162)	Oesphagus(288)
		<19	1.0	1.0	1.0	1.0
		20-34	1.1	0.9	0.8	1.0
		35-59	3.2	1.5	1.3	3.1
		>60	3.4	3.6	2.1	6.0
Choi & Kahyo, 1991[*]	Hospital	ml	(113)	(133)	(94)	-
		None	1.0	1.0	1.0	-
		< 5.5	0.6	1.2	0.3	-
		5.5-11.1	3.6	2.2	1.2	-
		11.1-22.2	4.2	4.1	2.4	-
		> 22.2	14.8	11.2	11.2	-
Martinez, 1969[*]	Hospital and neighbour-hood	Unit[ϕ]	(108)	(39)		(111)
		None	1.0	1.0	-	1.0
		<1	0.5	4.1	-	0.6
		2-4	1.7	1.4	-	2.1
		>5	2.8	14.7	-	7.7
Boffetta et al, 1992	Hospital	WE°	Floor of mouth (153)	Oral tongue (50)	Soft palate (44)	Tonsillar pillar (49)
		<1	1.0	1.0	1.0	1.0
		2-5	5.2	2.5	1.2	5.0
		6-10	9.6	7.6	4.3	20.3
		11-21	12.1	10.9	4.0	17.7
		>22	10.2	8.1	3.7	16.6

Cont **Table 1.14. Relative risks (RR) of oral cancer associated with alcohol intake from the selected case-control studies**

Authors	Type of control	Exposure category	Cancer site and (No.) Relative risk			
Merletti et al, 1989*	Population	Gram/day	Mouth/Oropharynx (86)			
		1-20	1.0			
		21-40	0.7			
		41-80	1.3			
		81-120	0.6			
		>120	2.1			
Brugere et al, 1986	Hospital	Gram/day	Mouth[Φ] (759)	Oropharynx (634)	Epilarynx (217)	hypopharynx (366)
		0-39	1.0	1.0	1.0	1.0
		40-99	2.7	2.6	1.9	3.3
		100-159	13.1	15.2	18.7	28.6
		> 160	70.3	70.3	101.4	143.1
Tuyns et al, 1988	Population	Gram/day	Supraglottic (426)	Other glottic (270)	Epilarynx (118)	Hypolarynx (281)
		<20	1.0	1.0	1.0	1.0
		21-40	0.9	0.8	0.9	1.6
		41-80	1.1	1.1	1.5	3.2
		81-120	1.7	1.7	5.1	5.6
		>121	2.0	3.4	10.6	12.5

*Results are presented only for males.
#1 drink corresponds to around 15 ml of ethanol.
ϕ1 unit is nearly equal to 20 ml of ethanol.
°Whisky-equivalent: 10.24g of ethanol.
ΦMouth cancer includes tongue (397), floor of mouth (206), gingiva (48), buccal mucosa (101).
Figures between parenthesis indicate total number of patients

Table 1.15. A summary of relative risks (RR) of oropharyngeal cancer for joint exposure to alcohol and smoking

Authors	Cancer site	No	Relative risk for combination AH[1]/SL[2]	AL[3]/SH[4]	AH/SH
Franco et al, 1989	Mouth	232	23.1	15.2	141.6
Rothman et al, 1972	Mouth-pharynx	598	2.3	2.4	15.5
Blot et al, 1988#	Pharynx	762	5.8	7.4	37.7
Francesch et al, 1990	Mouth-pharynx Oesphagus	291 288	2.3 7.9	17.6 6.4	79.6 17.5
Vassallo et al, 1985	Oesphagus	226	17.0	5.9	43.1
Brownson 1987	Larynx	63	2.4	3.4	7.7
Guenel et al, 1988	Glottis Supraglottis	197 214	5.1 50.6	19.2 46.8	289.4 109.2
Tuyns et al, 1988	Endolarynx Hypopharynx-epilarynx	727 399	3.8 14.7	11.5 4.9	43.2 135.5
Kato et al, 1992	All subsites combined	75	8.6	3.3	17.3
Zheng et al, 1990	Oral cancer	404	2.42	2.49	17.44
Zheng et al, 1997	Tongue cancer	111	2.41	7.60	4.07

[1] AH Highest exposure category for alcohol intake in each study.

[2] SL Lowest exposure category for smoking in each study.

[3] AL Lowest exposure category for alcohol intake in each study.

[4] SH Highest exposure category for smoking in each study.

The results are presented only for males.

Table 1.16. Natural history of epithelial dysplasia in uterine cervix

Degree of dysplasia	Regress %	Persist %	Progress to CIS %	Progress to SCC %
Mild	57	32	11	1
Moderate	43	35	22	5
Severe	32	<56	-	>12

(After Ostor, 1993)
CIS, Carcinoma-in-situ
SCC, Squamous cell carcinoma

Table 1.17. Summary of published cases of oral epithelial dysplasia that transformed to invasive squamous cell carcinoma

Authors	Number of cases	Number of invasive SCC	Transformation time (years)	Transformation rate (%)
Mincer et al, 1972	45	5	up to 8	11
Banoczy and Csiba 1976	68	9	1-20 Mean 6.3	13.2
Pindborg et al, 1977	61	4	up to 7	6.6
Gupta et al, 1980	73	6	10 mean 8.5	8.2
Silverman et al, 1984	22	8	Mean 8.1	36.4
Vedtofte et al, 1987	47	3	mean 3.9	6.3
Lumerman et al, 1995	44	7	up to 6.5	16
Total	360	42		11.6

Table 2.1. Mean patient age at time of diagnosis of oral epithelial dysplasia

Variables	No	%	Mean age (years)	SD°	Range (years)
Gender					
Male	351	55.7	54.8	13.7	20-93
Female	279	44.2	55.1	15.3	13-90
Total	630	100.0	55.0	14.4	13-93
Race and gender§					
White male	229/404	56.6	55.3	13.6	20-85
White female	175/404	43.3	55.3	16.2	13-90
Indian male	28/43	65.1	52.8	11.4	23-70
Indian female	15/43	34.8	52.1	13.1	30-73
P & B male•	43/88	48.8	52.8	13.5	28-81
P & B female•	45/88	51.1	53.3	12.5	30-79
Black male	14/28	50.0	52.2	16.2	23-93
Black female	14/28	50.0	58.8	12.3	37-75
Others male*	8/17	47.0	55.6	17.7	29-79
Others female*	9/17	52.9	52.0	17.1	37-82
Total Race	580				
Histology					
Mild dysplasia	297	47.1	54.1	14.6	13-93
Moderate dysplasia	183	29.0	55.6	14.0	20-86
Severe dysplasia	138	21.9	55.9	14.9	13-85
Carcinoma-in-situ	12	1.9	56.3	9.4	38-73
Total	630	100.0			
Overall	630	100	55.0	14.4	13-93

° Standard deviation

§ Race recorded for 580 patients only

• P&B: Pakistani and Bangladeshi

* Include Chinese, Middle eastern, Latin America

Table 2.2. Distribution of oral epithelial dysplasia lesions according to clinical presentation and ethnic-background of patients

Ethnic-background*	White patch		Red patch		Mixed		Ulcer		Lump		Total	
	No	%	No	%	No	%	No	%	No	%	No	%
Caucasian	201	49.7	6	1.4	186	46.0	10	2.4	1	0.2	404	69.6
Indian	18	41.8	-	-	21	48.8	3	7.0	1	2.3	43	7.4
Pakistani	15	38.4	2	5.1	21	53.8	1	2.5	-	-	39	6.7
Bangladeshi	33	67.3	2	4.0	13	26.5	-	-	1	2.0	49	8.4
Africans	11	39.2	1	3.5	16	57.1	-	-	-	-	28	4.8
Others	12	70.5	1	5.8	4	23.5	-	-	-	-	17	2.9
Total	290	50.0	12	2.0	261	45.0	14	2.4	3	0.5	580	100

*Clinical presentation of only 580 patients with known ethnic-background

Table 2.3. Clinical type of lesion and histological grading of oral epithelial dysplasia

Clinical presentation of lesion	Dysplasia				Total
	Mild	Moderate	Severe	CIS*	
White patch	140 (43.8)	96 (30.0)	79 (24.7)	4 (1.2)	319 (50.6)
Mixed (white and red)	138 (49.6)	77 (27.6)	57 (20.5)	6 (2.1)	278 (44.1)
Red patch	4 (33.3)	5 (41.6)	1 (8.3)	2 (16.6)	12 (1.9)
Ulcer	14 (77.7)	4 (22.2)	-	-	18 (2.8)
Lump	1 (33.3)	1 (33.3)	1 (33.3)	-	3 (0.4)
Total	297 (47.1)	183 (29)	138 (21.9)	12 (1.9)	630 (100.0)

* CIS=carcinoma-in-situ

Table 2.4. Comparison between patients with oral epithelial dysplasia aged 35 years or under, and those over 35 years of age

Variables	< 35 years of age No	%	> 35 years of age No	%	*P value*
Gender					
Male	25	42.3	326	57.0	0.03
Female	34	57.6	245	42.9	0.03
Total	59	100.0	571	100.0	
Ethnic-background*					
White	40	75.4	364	69.0	0.3
Indian	4	7.5	39	7.4	0.5
Pakistani and Bangladeshi	7	13.2	81	15.3	0.6
Black	1	1.8	27	5.1	0.2
Others	1	1.8	16	3.0	0.5
Total	53	100.0	527	100.0	
Site of dysplasia					
Buccal mucosa	11	18.6	122	21.3	0.6
Tongue	19	32.2	148	25.9	0.2
Floor of mouth	12	20.3	107	18.7	0.7
Labial mucosa	5	8.4	59	10.3	0.6
Gingiva	7	11.8	60	10.5	0.7
Soft palate	2	3.3	38	6.6	0.2
Retro-molar area	2	3.3	36	6.3	0.2
Commissure	1	1.6	1	0.1	0.1
Total	59	100.0	571	100.0	
Histology					
Mild dysplasia	37	62.7	260	45.5	0.01
Moderate dysplasia	12	20.3	171	29.9	0.1
Severe dysplasia	10	16.9	128	22.4	0.3
Carcinoma in-situ	-	-	12	2.1	-
Total	59	100.0	571	100.0	
Clinical type of lesion					
White patch	35	59.3	284	49.7	0.1
Red patch	1	1.6	11	1.9	0.6
Mixed (white and red)	22	37.2	256	44.8	0.2
Ulcer	1	1.6	17	2.9	0.4
Lump	-	-	3	0.5	-
Total	59	100.0	571	100.0	
Follow-up					
Recurrence	5•	12.8	58°	18.1	0.4
Second dysplasia	3∉	8.5	34•	10.4	0.7
Malignant transformation	3Ψ	5.0	17ϕ	5.6	0.8

* Ethnic-background of patients recorded for 53 patients of (<35 years group) and 527 patients of (>35 years group).

• Out of 39 cases having sufficient information regarding recurrence.

° Out of 320 cases having sufficient information regarding recurrence.

∉ Out of 35 cases having sufficient information regarding second dysplasia.

• Out of 324 cases having sufficient information regarding second dysplasia.

Ψ Out of 59 cases having sufficient information regarding malignant transformation.

ϕ Out of 300 cases having sufficient information regarding malignant transformation.

Table 2.5. Comparison between results of present study of oral epithelial dysplasia and previously reported similar investigations

Authors	Country	No of patients	Gender (%) M	F	Mean Age (years)	Dysplasia Mild	Moderate	Severe	Common site	(%)	Malignant change%
Mincer et al, 1972	USA	67	65.7	34.3	61.2	-	40	16	Lower lip Mandibular ridge Floor of mouth	19.4 17.9 16.4	11.1
Banoczy & Csiba, 1976	Hungary	120	86.7	13.3	-	20.6	60.0	19.5	Commissure Buccal mucosa Lips	39.1 20.8 20.0	13.2
Pindborg et al, 1977	India	61	52.5	47.5	-	-	-	-	Palate Buccal mucosa	50.8 42.6	6.6
Katz et al, 1985	USA	207	67	33	-	41	29	30	Tongue LT Floor of mouth	30.0 19.0 16.0	-
Kaugars et al, 1988	USA	1651	54.6	45.4	57.5	54.1*	19.5	8.1	Buccal mucosa Palate Floor of mouth	21.8 13.7 12.3	-
Lumerman et al, 1995	USA	308	52.8	47.2	59.3	59.4#	27.6	11.0ϕ	Floor of mouth Tongue Buccal mucosa	26.5 25.2 17.1	16.0
Present 1997	UK	630	55.7	44.2	55.0	47.1	29.0	21.9	Buccal mucosa Floor of mouth LT	21.1 18.8 15.3	5.5¶

* 18.3% of cases were diagnosed as focal mild dysplasia
# 3.6% of cases were verrucous hyperplasia with mild dysplasia
ϕ 3.9% of cases were diagnosd as carcinoma-in-situ LT: Lateral border of tongue
¶ 20/359 patients having sufficient follow-up data between two to 274 months mean 3.3 years

Table 3.1. Details to be included in an ideal referral letter or document

1. **Details of referring clinician:**
   Name
   Address
   Contact telephone numbers
   Facsimile number

2. **Patient detail:**
   Name
   Address
   Date of Birth
   Contact telephone numbers

3. **Patient Social History**:
   Occupation
   Ethnic Group
   Language spoken
   Alcohol/tobacco consumption
   Other habits
   Marital status/children

4. **Patient's medical history:**
   Including: Allergies
   Current medication

5. **Reason for Referral:**
   Symptoms
   Nature of lesion - site, size, colour, consistency, induration
   Associated lymphadenopathy
   Duration
   Previous treatment
   Possible diagnosis

6. **Requested Management:**
   Examine
   Treatment by specialists
   Advice regarding future treatment

7. **Degree of Urgency:**
   Urgent (1-7 days)
   Early (7-14 days)
   Routine

(After Zakrzewska, 1995)

Table 3.2. Summary of details of past medical history of patients omitted in letters of referral

Category of medical history	Patients omissions	%
Cardiovascular	25	17.0
Haematological	1	0.6
Respiratory	8	5.4
Endocrine	18	12.2
Neurological and psychological	18	12.2
Genitourinary	1	0.6
Musculoskeletal	18	12.2
Dermatological	10	6.8
Allergies	10	6.8
Previous surgery/hospital admissions	30	20.4
Previous malignancy	8	5.4
Total	147	100.0

Table 3.3. Demographics of 182 responding UK dental practitioners

Factors	Respondents number	%
Gender (Total 182)		
Male	127	69.7
Female	55	30.2
Dental qualifications (Total 182)		
BDS	109	59.8
LDS RCS	13	7.1
BDS & LDS	40	21.9
Others	20	10.9
Postgraduate qualifications (Total 56)		
MSc	9	16.0
FDS	17	30.3
FDS/MSc	5	8.9
DGDP	10	17.8
PhD	4	7.1
Others	11	19.6
Attendance at postgraduate meetings (Total 182)		
BDA	98	58.8
Section 63	124	68.1
Royal College of Surgeons	73	40.1
University-organised	79	43.4
Postgraduate courses in oral malignancy and premalignancy	82	45.0
Methods of continuous education by dental surgeons (Total 45)		
Computer assisted learning	8	17.7
Videotapes	32	71.1
CAL and Videotapes	4	8.8
Audiotapes	1	2.2
Use of local postgraduate medical and dental centers (Total 113)		
Once a year	21	18.5
2-5 times per year	52	46.0
>5 times per year	40	35.3
Professional Journals commonly read (Total 182)		
BDJ	136	74.7
Dental Practice	58	31.8
Dental Update	37	20.3
Probe	34	18.6
BMJ	8	4.3
Other	3	1.6

BDS: Bachelor of Dental Surgery
LDS: Licentiate of Dental Surgery
MSc: Master of Science
FDS: Fellow Dental Surgery
DGDP: Diploma in General Dental Practice
CAL: Computer-assisted-learning
BDJ: British Dental Journal
BMJ: British Medical Journal
PhD: Doctor of Philosophy

Table 3.4. Undergraduate clinical experience of responding dental practitioners of potentially malignant and malignant lesions

Number and type of malignant and potentially malignant lesions	No of respondents who witnessed lesions	%
Number of malignant lesions		
<10	40	31.2
11-21	53	41.4
>21	35	27.3
Total	128	100.0
Type of malignancy		
Squamous cell carcinoma	72	56.2
Ameloblastoma	16	12.5
Lymphoma	7	5.4
Kaposi's sarcoma	7	5.4
Melanoma	11	8.5
Salivary gland tumours	15	11.7
Total	128	100.0
Number of potentially malignant lesions		
<10	22	13.8
11-21	92	57.8
>21	45	28.3
Total	159	100.0
Type of potentially malignant lesions		
Leukoplakia	80	50.3
Erythroplakia	20	12.5
Lichen planus	36	22.6
Atrophic glossitis	6	3.7
Submucous fibrosis	6	3.7
Sideropenic anaemia	7	4.4
Others	4	2.5
Total	159	100.0

Table 3.5. The likely characteristics of oral squamous cell carcinoma as detailed by responding dental practitioners

Characteristics	Number	%
Likely sites : (182 respondents)		
Floor of mouth	65	35.7
Tongue	43	23.6
Cheek	25	13.7
Lateral border of tongue	23	12.6
Ventral surface of tongue	15	8.2
Upper lip	27	14.8
Lower lip	17	9.3
Hard palate	17	9.3
Soft palate	6	3.2
Retro-molar area	6	3.2
Oropharynx	4	2.1
Dorsum of the tongue	3	1.6
Cervical Lymph node	1	0.5
Commisure	-	-
Likely size: (129 respondents)		
5-10 mm	84	65.1
10-20 mm	25	19.3
20-30 mm	20	15.5
Likely colour: (175 respondents)		
White	44	25.1
Red	35	20.0
Speckled	32	18.2
White and red	64	36.5
Additional features: (182 respondents)		
Paraesthesia	78	42.8
Tooth mobility	44	24.1
Pathological fracture	29	15.9
Pain	28	15.3
Anaesthesia	3	1.6

Table 3.6. Reported aetiological features of oral squamous cell carcinoma by 182 responding dental practitioners

Aetiological features	Number	%[#]
Tobacco and alcohol	110	60.4
Previous oral cancer	75	41.2
HIV infection	36	19.7
Poor oral hygiene	30	16.4
Candidal infection	21	11.5
Syphilis	13	7.1
Tobacco alone	11	6.0
Malnutrition	8	4.3
Leukaemia	5	2.7
Epstein Barr virus	4	2.1
Herpes simplex virus	3	1.6
Human papilloma virus	3	1.6

[#]Percentages out of 182 responding dental practitioners

Table 3.7. Suggested efficacy and recommended duration of oral screening programmes by responding dental practitioners

Screening	Number	%
Effectiveness: (182 respondents)		
Effective	151	82.9
Ineffective	6	3.2
Un-decided	25	13.7
Duration of screening: (151 respondents)		
Every 3 months	63*	41.7
Every 6 months	70*	46.3
Every year	9	5.9
Every 2 years	6	3.9
Every 3 years	1	0.6
> 3 years	2	1.3

*133/151 (88.0%) respondents suggested screening at an interval of 3-6 months an effective means of reducing the frequency of premalignant and malignant lesions.

Table 4.1. Demographic characteristics of oral epithelial dysplasia cases and control subjects

Variable	Cases		Controls	
	No	%	No	%
Age (years)				
<45	148	23.4	163	25.3
45-54	132	20.9	123	19.1
55-64	175	27.7	178	27.6
65-74	123	19.5	123	19.1
75+	52	8.2	56	8.7
Total	630	100.0	643	100.0
Gender				
Male	351	55.7	354	55.0
Female	279	44.2	289	44.9
Total	630	100.0	643	100.0
Race#				
Caucasians	404	69.6	403	69.7
Indians	43	7.4	43	7.4
Pakistani & Bangladeshi	88	15.1	89	15.3
Afrocaribbeans	28	4.8	15	2.5
Others	17	2.9	28	4.8
Total	580	100.0	578	100.0
Marital status#				
Married	204	57.4	233	67.3
Single	60	16.9	16	4.6
Divorced	66	18.5	97	28.0
Widowed	25	7.0	-	-
Total	355	100.0	346	100.0

#= Number do not equal the total shown due to missing values.

Table 4.2. Odds ratios of oral epithelial dysplasia for tobacco smoking alcohol consumption: results of univariate analysis

Risk factors	Cases	Controls	OR# (95% C.I.) ϕ	*P*
Smoking				
Never*	119	132	1	
Ex-smoker	123	73	1.20 (0.96, 1.50)	
Current	285	157	1.29 (1.08, 1.50)	
Smoking period (years)				
Never *	119	131	1	
1-19	158	83	0.84 (0.63,1.12)	
20-39	212	68	1.38 (1.03,1.08)	
> 40	39	8	2.15 (1.20, 3.85)	<0.001
Cigarette (per day)				
None*	119	134	1	
1-9	81	54	0.55 (0.37, 0.82)	
10-19	139	133	0.38 (0.27, 0.55)	
20-29	109	39	1.02 (0.67, 1.55)	
30+	79	2	14.43 (4.66, 44.6)	<0.001
Years stopped smoking				
None*	384	438	1	
1-10	52	6	3.32 (1.87, 5.92)	
10 +	75	67	0.43 (0.30, 0.62)	
Type of tobacco				
None*	122	190	1	
Filter cigarette	296	158	1.03 (0.76, 1.39)	
Non filter cigarette	76	13	3.22 (1.90, 5.44)	
Pipe	18	12	0.83 (0.44, 1.54)	
Roll-up	15	8	1.03 (0.51, 2.11)	
Alcohol				
Never*	131	104	1	
Current	302	182	0.53 (0.41, 0.69)	
Ex-drinkers	45	81	1.20 (0.96, 1.49)	
Type of alcohol				
None *	131	104	1	
Beer	87	74	0.83 (0.62, 1.12)	
Wine	72	48	1.06 (0.76, 1.48)	
Fortified wine	39	29	0.95 (0.63, 1.44)	
Spirits	47	25	1.33 (0.88, 2.01)	
Units (per week)				
None*	129	104	1	
1-19	222	183	0.84 (0.65, 1.09)	
20-39	108	69	1.09 (0.80, 1.47)	
40 +	20	11	1.26 (0.72, 2.22)	0.1

#=Crude Odds ratio
ϕC.I.=Confidence interval
*= Reference category
P value for linear trends

Table 4.3. Logistic regression analysis of smoking and alcohol status and risk of oral epithelial dysplasia

Variable	Cases	Controls	OR#	95% C.I. ϕ
Smoking				
Never*	99	109	1	-
Current	240	117	1.20	(0.97, 1.49)
Previous	98	46	1.48	(1.12, 1.95)
Alcohol				
Never*	115	88	1	-
Current	279	139	1.46	(1.16, 1.82)
Previous	43	45	0.64	(0.46, 0.87)

*Reference category
#OR=Odds ratio
ϕC.I.=Confidence intervals

Table 4.4. Interaction between alcohol drinking and tobacco smoking (logistic regression) and risk of oral epithelial dysplasia

Variable	Cases	Controls	OR#	95% C.Iϕ
Smoking				
Never*	99	109	1	-
Current	240	117	0.67	(0.39, 1.14)
Previous	98	46	2.07	(1.44, 2.98)
Smokers (Non-drinkers)				
No *	209	155	1	-
Yes	228	117	1.22	(1.01, 1.47)
Drinkers (Non-smokers)				
No*	208	101	1	-
Yes	229	272	1.57	(1.03, 2.40)

*Reference category
#OR=Odds ratio
ϕC.I.=Confidence intervals

Table 4.5. Interaction between alcohol, alcohol type, and daily number of cigarettes smoked (logistic regression) and risk of oral epithelial dysplasia

Variables	Cases	Controls	OR#	95% C.I.ϕ
Alcohol drinking				
None*	115	88	1	-
Current	279	139	1.59	(1.19, 2.12)
Previous	43	45	0.57	(0.36, 0.89)
Alcohol type				
None*	115	88	1	-
Beer	81	61	0.57	(0.34, 0.94)
Wine	70	47	1.94	(0.94, 4.03)
Fortified wine	38	3	0.89	(0.38, 2.10)
Spirits	47	13	1.24	(0.33, 4.68)
Cigarettes (day)				
None*	83	87	1	-
1-9	45	26	0.81	(0.51, 1.28)
10-19	91	76	0.60	(0.41, 0.87)
20+	132	23	4.38	(2.64, 7.27)

*Reference category
#OR=Odds ratio
ϕC.I.=Confidence intervals

Table 4.6. Interaction between alcohol use, daily number of cigarettes smoked and number of years smoking tobacco (logistic regression) and risk of oral epithelial dysplasia

Variables	Cases	Controls	OR[#]	95% C.I.[ϕ]
Smoking				
Never*	83	86	1	-
Current	194	89	0.35	(0.22, 0.57)
Previous	74	37	0.90	(0.61, 1.34)
Tobacco type				
None*	83	110	1	-
Filter	200	87	1.04	(0.64, 1.70)
Non-filter	49	4	6.60	(2.60, 16.76)
Pipe	9	7	1.07	(0.40, 2.82)
Roll-up	10	4	1.21	(0.42, 3.49)
Alcohol type				
None*	133	88	1	-
Beer	81	61	0.61	(0.39, 0.93)
Wine	70	47	0.66	(0.42, 1.02)
Fortified wine	38	3	3.84	(1.44, 10.2)
Spirits	47	13	1.31	(0.72, 2.40)
Interaction 1*	60	10	1.76	(1.18, 2.62)
Interaction 2[#]	21	1	5.61	(2.01, 15.61)
Interaction 3[δ]	25	1	2.96	(1.06, 8.28)

*Reference category
*Interaction 1= 'smoking 20+ cigarettes per day and a current drinkers'
[#] Interaction 2= 'smoking 20+ cigarettes per day and a non-drinkers'
[δ]Interaction 3= 'smoking for 20 years and a current drinker'
[#]OR=Odds ratio
[ϕ]C.I.=Confidence intervals

Table 4.7. Interaction between tobacco and alcohol related behaviours (logistic regression) and risk of oral epithelial dysplasia

Variable	Cases	Controls	OR#	95% C.I.ϕ
Alcohol				
None*	79	93	1	-
Current	74	169	2.39	(0.76, 7.45)
Previous	22	15	0.77	(0.25, 2.35)
Alcohol type				
None*	79	93	1	-
Beer	47	68	0.46	(0.20, 1.05)
Wine	34	55	0.49	(0.21, 1.16)
Fortified wine	3	29	2.79	(0.67, 11.53)
Spirits	12	32	0.44	(0.20, 0.96)
Interaction 4#	12	93	6.28	(3.28, 12.02)

*Reference category
Interaction 4 = current smoker and smoking 20+ cigarettes per day
#OR=Odds ratio
ϕC.I.=Confidence intervals

Table 4.8. Effect of tobacco cessation upon oral epithelial dysplasia (logistic regression)

Variables	Cases	Controls	OR#	95% C.I.ϕ
Stop smoking				
None*	285	157	1	-
1-9	52	6	3.21	(1.69, 6.12)
10+	71	67	0.61	(0.39, 0.97)
Tobacco type				
None*	4	71	1	-
Filter	295	130	1.71	(1.12, 2.60)
Non-filter	76	12	5.54	(3.00, 10.23)
Pipe	18	11	1.77	(0.84, 3.75)
Roll-up	15	6	1.72	(0.74, 4.02)
Cigarettes (day)				
None*	1	2	1	-
1-9	80	54	0.97	(0.46, 2.08)
10-19	139	133	0.59	(0.28, 1.28)
20 +	188	41	3.20	(1.44, 7.11)

*Reference category
#OR=Odds ratio
ϕC.I.=Confidence intervals

Table 4.9. Interaction between alcohol-related behaviours (logistic regression)

Variables	Cases	Controls	OR#	95% C.I.Ψ
Alcohol				
Non-drinker*	115	88	1	-
Current	279	139	1.23	(0.99, 1.52)
Previous	43	45	1.35	(1.03, 1.77)
Interaction 5#	35	3	7.06	(2.13, 23.33)
Interaction 6ϕ	41	9	2.74	(1.29, 5.78)
Interaction 7Θ	9	21	0.28	(0.12, 0.63)

*Reference category
#Interaction 5= 'current drinkers and drinks fortified wine'
ϕ Interaction 6= 'current drinkers and drinks spirits'
ΘInteraction 7= 'previous drinkers and used to drink beer'
#OR=Odds ratio
ΨC.I.=Confidence intervals

Table 4.10 Multivariate adjusted odds ratio and 95% confidence interval of oral epithelial dysplasia associated with alcohol and tobacco type, daily number of cigarettes

Variables	Cases	Controls	OR#	95% C.I.ϕ
Alcohol				
None*	93	79	1	-
Current	228	117	1.52	(1.14, 2.03)
Previous	18	30	0.58	(0.37, 0.91)
Cigarettes (day)				
None*	99	110	1	-
1-9	46	27	0.83	(0.52, 1.31)
10-19	81	74	0.60	(0.41, 0.88)
20 +	113	15	4.38	(2.60, 7.21)
Tobacco type				
None*	108	99	1	-
Filter cigarettes	99	173	0.58	(0.34, 0.96)
Non-filter cigarettes	9	46	1.83	(0.88, 3.80)
Cigar	8	13	0.86	(3.67, 2.03)
Roll-up	2	8	1.29	(0.34, 4.89)
Interaction 5#	26	3	3.73	(1.05, 13.23)

*Reference category
Interaction 5= 'current drinker and drinks fortified wine'
#OR=Odds ratio
ϕC.I.=Confidence intervals

Table 4.11. Odds ratio for non-drinking oral dysplasia cases and controls by smoking habit

Tobacco consumption	Non-drinkers Cases	Controls	OR# (95% C.I.) ϕ
Smoking status			
Non-smokers*	38	47	1*
Current smokers	55	32	1.14 (0.75, 1.73)
Previous smokers	22	9	1.62 (0.93, 2.83)
Total	115	88	
Cigarettes (day)			
None*	37	47	1*
1-9	14	10	0.66 (0.29, 1.49)
10-19	13	21	0.29 (0.13, 0.62)
20+	29	1	13.74 (2.99, 33.02)
Total	93	79	
X^2 P for linear trend			13.78 $P< 0.001$
Tobacco type			
None*	39	48	1*
Filter cigarettes	57	34	1.31 (0.63, 2.69)
Non-filter cigarettes	15	3	3.91 (1.23, 12.40)
Pipe	1	3	0.26 (0.04, 1.68)
Roll-up	3	2	1.17 (0.25, 5.40)
Total	115	90	
Stop smoking (years)			
None*	71	48	1*
< 10	11	1	3.90 (0.95, 15.88)
10 +	11	8	0.48 (0.19, 1.22)
Total	93	57	

*Reference category
#OR=Odds ratio
ϕC.I.=Confidence intervals

Table 4.12. Odds ratio for non-smoking oral dysplasia cases and controls by alcohol consumption

Alcohol consumption	Non-smokers Cases	Controls	OR# (95% C.I.) ϕ
Alcohol status			
Non-drinkers	38	47	1*
Current drinkers	34	48	1.46 (0.95, 2.23)
Previous drinkers	7	14	0.65 (0.34, 1.22)
Total	79	109	
Units (week)			
None*	36	47	1*
1-19	46	47	0.73 (0.37, 1.40)
20-39	12	14	0.64 (0.28, 1.42)
40 +	5	1	3.73 (0.73, 19.03)
Total	99	109	
X^2 P for linear trend			1.78 $P<$ 0.18
Alcohol type			
None*	38	47	1*
Beer	19	19	0.65 (0.31, 1.37)
Wine	14	16	0.57 (0.26, 1.25)
Fortified wine	5	1	3.27 (0.56, 18.88)
Spirits	7	3	1.52 (0.46, 4.99)
Total	83	86	

*Reference Category
#OR=Odds ratio
ϕC.I.=Confidence intervals

Table 4.13. Distribution of oral epithelial dysplasia by site, and patient gender, and age

Site	Gender			Age (years)		
	Male	Female	M/F Ratio	Mean	SD	Range
Upper labial mucosa	19	12	1.5	51.97	14.95	13-77
Lower labial mucosa	17	16	1	59.12	14.1	29-84
Lateral tongue	56	41	1.3	55.91	15.2	24-93
Ventral tongue	27	20	1.3	54.53	15.0	21-85
Dorsum of tongue	13	11	1.1	55.82	15.0	26-61
Gingiva	10	12	0.8	50.41	17.5	15-86
Alveolus	23	21	1	55.75	16.0	13-82
FOM (Ant.)	45	43	1	54.49	12.7	20-82
FOM (post.)	16	15	1	50.13	12.2	20-71
Buccal mucosa	78	55	1.4	54.69	13.7	23-86
Soft palate	21	17	1.2	57.65	14.6	23-85
RMA	25	14	1.7	57.95	14.0	21-90
Commissure	1	2	0.5	42.50	16.2	31-54
Total	351	279	1.25	55.0	14.4	13-93

FOM= Floor of mouth
RMA= Retro-molar area
S.D= Standard deviation

Table 4.14. Tobacco smoking and site of oral epithelial dysplasia

Site	No. of patients	Non-smokers	Cigarette per week ≤ 70	> 70
Males		OR* 95% C.I. ϕ	OR* 95% C.I. ϕ	OR* 95% C.I.ϕ
Labial mucosa	37	1	27.0 (3.20,597.8)	9.56 (1.26,199.95)
Tongue	97	1	2.32 (0.99,5.43)	1.50 (0.80,2.84)
Gingiva	33	1	2.50 (0.60,10.38)	1.72 (0.58,5.28)
Floor of mouth	62	1	10.80 (3.31,37.49)	4.56 (1.59,14.05)
Buccal mucosa	78	1	3.50 (1.31,9.42)	2.27 (1.05,4.95)
Soft palate	21	-	- -	- -
Non-specific site	23	1	2.25 (0.37,13.10)	2.02 (0.56,7.87)
Females				
Labial mucosa	27	1	0.52 (0.10,2.26)	0.49 (0.15,1.58)
Tongue	70	1	0.74 (0.23,2.33)	2.38 (1.12,5.11)
Gingiva	34	1	0.94 (0.28,3.10)	0.67 (0.23,1.88)
Floor of mouth	57	1	0.86 (0.20,3.51)	3.16 (1.26,8.12)
Buccal mucosa	55	1	1.08 (0.40,2.89)	0.71(0.30,1.70)
Soft palate	19	1	4.32 (0.68,34.31)	3.66 (0.70,25.65)
Non-specific site	36	1	1.73 (0.26,1157)	1.36 (0.27,7.54)

*OR= Odds ratio ϕC.I.=Confidence intervals

No Odds ratio or confidence limits were calculated for the soft palate in males because as no male patient with dysplastic lesion of soft palate did not smoke.

Table 4.15. Alcohol consumption and site of oral epithelial dysplasia

Site	No. of patients	Non-drinkers	Unit of alcohol per week ≤ 21	> 21
Males		OR*95% C.I.ϕ	OR* 95% C.I. ϕ	OR* 95% C.I.ϕ
Labial mucosa	37	1	0.69 (0.21,2.32)	0.90 (0.20,3.94)
Tongue	97	1	1.58 (0.73,3.46)	2.25 (0.92,5.57)
Gingiva	33	1	0.56 (0.20,1.59)	0.45 (0.09,1.99)
Floor of mouth	62	1	1.54 (0.64,3.76)	1.35 (0.44,4.14)
Buccal mucosa	78	1	1.02 (0.48,2.21)	0.96 (0.35,2.61)
Soft palate	21	1	1.08 (0.24,5.49)	2.25 (0.43,12.76)
Non-specific sites	23	1	0.46 (0.12,1.71)	1.35 (0.35,5.19)
Females				
Labial mucosa	27	1	1.15 (0.34,4.12)	1.58 (0.27,8.73)
Tongue	70	1	1.03 (0.46,2.30)	2.07 (0.72,5.94)
Gingiva	34	1	0.57 (0.19,1.71)	2.34 (0.69,7.93)
Floor of mouth	57	1	0.94 (0.42,2.14)	0.94 (0.25,3.34)
Buccal mucosa	55	1	0.82 (0.36,1.89)	0.56 (0.11,2.45)
Soft palate	19	1	1.15 (0.34,4.12)	0.53 (0.02,5.24)
Non-specific sites	36	1	1.72 (0.30,12.88)	- -

*OR=Odds ratio ϕC.I.=Confidence intervals

Table 4.16. Demographic characteristics of patients with oral epithelial dysplasia and control subjects

Variables	Cases		Control	
	No	%	No	%
Age (yrs)				
< 30	1	0.8	7	2.8
30-50	43	35.8	79	31.6
51-70	60	50.0	125	50.0
> 70	16	13.3	39	15.6
Total	120	100.0	250	100.0
Gender				
Male	64	53.3	143	57.2
Female	56	46.7	107	42.8
Total	120	100.0	250	100.0
Ethnic-background				
White	98	81.6	175	70.0
Indian	9	7.5	19	7.6
Pakistani	10	8.3	26	10.4
African	3	2.5	30	12.0
Total	120	100.0	250	100.0

Table 4.17. Serum and Red Blood cell folate and serum vitamin B12 in patients with oral epithelial dysplasia and control subjects

Groups	No	Serum Folate µg/l Mean	± SD	RBC folate µg/l Mean	± SD	B12 ng/l Mean	± SD
(A) **Oral dysplasia**	120	3.1	2.9	228.8	138.7	277.6	201.3
Controls							
(B) Smokers	118	13.7	2.6	587.0	189.7	272.3	161.3
(C) Non-smokers	132	10.1	2.8	504.6	197.2	326.9	193.2
(D) Drinkers	111	3.4	3.1	350.9	167.3	308.7	177.6
(E) Non-drinkers	139	3.7	3.1	392.6	186.2	317.0	204.1

S.D.= Standard deviation

Significance:

Serum Folate	**RBC folate**	**Vitamin B12**
A vs B $P<0.05$	A vs B $P<0.05$	A vs B $P=0.9$
A vs C $P=0.07$	A vs C $P=0.09$	A *vs* C $P=0.5$
A vs D $P=0.9$	A vs D $P=0.7$	A vs D $P=0.8$
A vs E $P=0.9$	A vs E $P=0.6$	A vs E $P=0.7$

Table 4.18. Serum ferritin, iron and total iron binding capacity in patients with oral epithelial dysplasia and control subjects

Groups	No	Serum Ferritin mg/l Mean	± SD	Iron umol/l Mean	± SD	TIBC umol/l Mean	± SD
(A) **Oral dysplasia**	120	118	113.3	18.8	7.2	58.2	14.9
Controls							
(B) Smokers	118	107	118	12.0	6.7	58.6	10.3
(C) Non-smokers	132	112	147	14.1	7.5	75.3	10.2
(D) Drinkers	111	103	125	12.3	6.9	62.9	12.1
(E) Non-drinkers	139	114	147	13.5	7.1	67.9	14.1

S.D.= Standard deviation

TIBC=Total iron binding capacity

Significance:

Serum Ferritin	**Iron**	**TIBC**
A vs B $P=0.4$	A vs B $P=0.4$	A vs B $P=0.9$
A vs C $P=0.9$	A vs C $P=0.9$	A vs C $P=0.7$
A vs D $P=0.03$	A vs D $P=0.6$	A vs D $P=0.9$
A vs E $P= 0.8$	A vs E $P=0.8$	A vs E $P=0.8$

Table 4.19. Association between levels of folate, vitamin B12 and oral epithelial dysplasia (multiple logistic regression)

Factors	Cases	Controls	OR #	95% C.I.ϕ
Serum folate (μg/l)				
1-3*	89	46	1*	
4-6	24	71	0.17	0.09, 0.33
> 6	7	64	0.06	0.02, 0.14
Total	120	181		
Chi-square test			68.9	
P trend			<0.001	
RBC folate (μg/l)				
< 150*	54	10	1*	
150-300	32	52	0.11	0.05, 0.27
> 300	34	121	0.05	0.02, 0.12
Total	120	183		
Chi-square test			68.2	
P trend			<0.001	
B12 (ng/l)				
< 165*	54	38	1*	
165-684	61	130	0.33	0.19, 0.57
> 684	5	12	0.29	0.08, 1.00
Total	120	180		
Chi-square test			16.6	
P trend			<0.001	

*Reference category
#OR=Odds ratio
ϕC.I.=Confidence intervals

Table 4.20. Tobacco and alcohol consumption and risk of oral epithelial dysplasia results of multiple logistic regression

	Cases	Controls	OR#	95%C.I. ϕ
Smoking status				
None*	24	132	1*	
Current	60	88	3.75	2.10, 6.72
Ex-smokers	36	30	6.60	3.28, 13.37
Cigarette (per day)				
None*	24	132	1*	
1-9	7	25	1.54	0.54, 4.29
10-19	35	80	2.41	1.28, 4.53
20-29	28	13	11.85	5.0, 28.28
> 29	26	-	-	
Total	96	118		
P trend			<0.001	
Type of cigarette				
None*	24	132	1*	
Filter	40	96	2.29	1.25, 4.2
Non-filter	26	14	10.21	4.3, 24.1
Cigar	30	2	82.5	18.3, 731.
Roll-up	-	6	-	
Total	96	118		
Years smoking				
None*	24	132	1*	
1-19	44	61	3.97	2.1, 7.4
20-39	34	53	3.53	1.8, 6.8
> 39	18	4	24.75	7.1, 106.3
Total	96	118		
P trend			<0.001	
Alcohol consumption				
None*	4	139	1*	
Current	92	89	35.92	12.1, 138.0
Previous	24	22	37.91	11.2, 159.5
Unit (per week)				
None*	4	139	1*	
1-7	56	28	69.50	22.4, 277.3
8-15	37	33	38.96	12.4, 157.2
> 15	23	50	15.99	5.0, 65.7
Total	116	111		
P trend			<0.001	
Type of alcohol				
None*	4	139	1*	
Beer	46	49	35.62	10.9, 129.0
Wine	34	43	27.48	8.9, 110.5
Fortified wine	19	7	94.32	22.0, 453.8
Spirits	17	12	49.23	12.7, 222.5
Total	116	111		

*Reference category
#OR=Odds ratio
ϕC.I.=Confidence intervals

Table 4.21. Interactive effect between red blood cell folate and tobacco smoking, vitamin B12 upon risk of oral epithelial dysplasia

Factors	red blood cell folate μg/l <300μg/l Case/control	OR #	95%C.I. ϕ	>300μg/l Case/control	OR #	95%C.I. ϕ
Smoking						
Never*	6/67	1	-	15/65	1	-
Current	25/36	5.1	2.0, 13.4	35/52	2.9	1.3,6.2
Ex-smoker	17/13	9.7	3.2, 30.3	19/17	4.8	1.8,12.6
Cigarette/day						
None*	9/67	1	-	15/65	1	-
1-9	3/12	1.8	0.3, 8.9	4/13	1.3	0.2,5.1
10-19	15/45	2.4	0.9,6.7	20/35	2.4	1.0,5.8
20-29	17/7	18.0	5.2,66.2	11/6	7.6	2.2,9.3
>29	16/-	-	-	10/-	-	-
Years smoking						
None*	9/67	1	-	15/65	1	-
1-19	21/20	7.8	2.8,22.0	23/41	2.4	1.0,5.5
20-39	23/22	7.7	2.8,21.5	11/31	1.5	0.5,4.0
>39	12/3	29	6.0,18.3	6/1	26	2.7,12.3
Vitamin B12						
<165*	31/28	1	-	23/10	1	-
165-684	20/45	0.4	0.1,0.8	41/85	0.2	0.08,0.5
>684	2/5	0.3	0.03,2.4	3/7	0.1	0.03,1.0

*Reference category
#OR=Odds ratio
ϕC.I.=Confidence intervals

Table 4.22. Studies of Candida associated with oral leukoplakia

Author(s)	Year	Size of material	No and percentage with Candida in biopsy
Jepsen & Winther,	1965	24	13 (54.2%)
Cawson,	1966	138	15 (10.8%)
Renstrup,	1970	235	55 (23.4%)
Roed-Petersen et al,	1970	98	30 (30.6%)
Daftary et al,	1971	723	49 (6.7%)

Table 4.23. Salivary Candida albicans counts in patients with oral epithelial dysplasia (cases) and control subjects

Candida count	Cases		Controls		*P* value
	No	%	No	%	
< 1000	59	35.3	15	35.7	0.9
1000-10,000	75	44.9	24	57.1	0.1
> 10,000	33	19.7	3	7.1	0.05
Total	167	100	42	100	<0.001

P for chi-squared test

Table 4.24. Correlation of salivary Candida albicans counts and presence of Candida in oral epithelial dysplasia tissue

Candida count	Number	%	Candida in lesional tissue Number	(%) of patients with Candida in tissue
< 1000	59	35.3	3*	20.0
1000-10,000	75	44.9	5	33.3
> 10,000	33	19.7	7	46.6
Total	167	100.0	15	8.9

* (3/15) 20.0% of patients with Candida in tissue were found among patients who have Candida count of less than 1000

Table 4.25. Clinical presentation of oral epithelial dysplasia and presence of Candidal hyphae and subsequent malignant transformation

Clinical presentation	Candidal hyphae No	%	Malignant transformation of lesion with Candida No	%
White patch	6/90	6.6	3/6	50.0
Mixed red/white patch	8/75	10.6	5/8	62.5
Red patch	1/2	50.0	1/1	100.0
Total	15/167	8.9	9/15*	60.0

*9 of 15 lesions with Candidal hyphae transform to malignancy within a follow-up period range between 27 to 274 months.

Table 4.26. Anatomical distribution of oral epithelial dysplasia with intralesional Candida

Location	Oral epithelial dysplasia Number	%	Candida hyphae Number	%
Lateral border of tongue	11	6.5	1	6.6
Ventral border of tongue	28	16.7	3	20.0
Dorsal surface of tongue	8	4.7	1	6.6
Floor of mouth	66	39.5	4	26.6
Buccal mucosa	43	25.7	3	20.0
Gingiva	5	2.9	2	13.3
Retromolar area	6	3.5	1	6.6
Total	167	100.0	15	100.0

Table 4.27. HCV antibodies in 75 patients with oral epithelial dysplasia and healthy control subjects

Patient group	No of patients	Gender Male	Female	Age (yrs.) Median	Range	HCV-antibodies No	%
Oral epithelial dysplasia	75[#]	41	34	51	32-70	2	2.6
Control subjects	110	86	24	39	25-65	-	0
Total	185	127	58	90	25-70	2	1.0

[#] Results available for 75 patients out of total of 120 patients

Table 4.28. Frequency of serum IgG antibodes to Helicobacter pylori in patients with oral epithelial dysplasia and healthy control subjects

Patient group	No of patients	Gender M	F	Age (yrs.) Median	Range	IgG antibodies to H. pylori No	%	*P* value
Oral epithelial dysplasia	120	64	56	54	29-80	15	12.5	>0.05
Healthy control subjects	25	6	19	26	20-50	6	24.0	>0.05
RAS[ϕ]	75	24	51	34	9-72	23	30.6	<0.05
Oral dysaesthesia	27	10	17	60	27-74	11	40.7	<0.05
Total controls	127	40	87	40	9-74	40	31.4	<0.05

P for chi-squared test

ϕ Recurrent aphthous stomatitis

Table 5.1. Outcome of long-term follow-up of 359 patients with oral epithelial dysplasia

Status	No	%
No lesions	237	66.0
Recurrence of dysplastic lesion	63	17.5
Second dysplastic lesion	37	10.3
Malignant transformation	20	5.5
Dead-oral cancer	1	0.2
Dead-other causes	1	0.2
Total	359	100.0

Table 5.2. Demographic characteristics of 20 patients who developed a squamous cell carcinoma subsequent to oral epithelial dysplasia compared with patients who did not develop malignancy

Variables	OED with later SCC		OED with no SCC	
	No	%	No	%
Age (years)				
< 40	2	10.0	50	14.7
40-50	7	35.0	79	23.3
> 50	11	55.0	210	61.9
Total	20	100.0	339	100.0
Gender				
Male	9	45.0	181	53.3
Female	11	55.0	158	46.6
Total	20	100.0	339	100.0
Ethnic-background				
Caucasian	15	75.0	204	60.1
Indian	2	10.0	21	6.1
Pakistani & Bangladeshi	3	15.0	47	13.8
Afro-caribbians	-	-	4	1.1
Others	-	-	63	18.5
Total	20	100.0	339	100.0
Marital status				
Married	8	40.0	142	53.9
Single	4	20.0	48	18.2
Widowed	4	20.0	32	12.1
Divorced	4	20.0	41	15.5
Total	20	100.0	263	100.0

OED= Oral epithelial dysplasia
SCC= Squamous cell carcinoma

Table 5.3. Clinical type of oral epithelial dysplasia (OED) and subsequent development of squamous cell carcinoma (SCC)

Clinical type of the lesion	OED with later SCC		OED with no SCC		
	No	%	No	%	*P* value
White patch	4	20.0	166	48.9	0.09
Mixed (white and red)	9	45.0	150	44.2	0.9
Red patch	6	30.0	3	0.8	0.001
Ulcer	1	5.0	17	5.0	0.7
Lump	-	-	3	0.8	-
Total	20	100.0	339	100.0	

P for chi-squared test
OED=Oral epithelial dysplasia
SCC=Squamous cell carcinoma

Table 5.4. Distribution according to site of oral epithelial dysplasia (OED) and subsequent development of squamous cell carcinoma (SCC)

Site	OED with later SCC No	%	OED with no SCC No	%	*P* value
Floor of mouth	8	40.0	59	17.4	0.01
Gingiva	3	15.0	8	2.3	0.003
Soft palate	3	15.0	20	5.8	0.1
Buccal mucosa	2	10.0	68	20.0	0.2
Lateral border of tongue	2	10.0	21	6.1	0.5
Ventral border of tongue	1	5.0	56	16.5	0.1
Labial mucosa	1	5.0	38	11.2	0.3
Dorsal surface of tongue	-	-	23	6.7	-
Alveolar ridge	-	-	19	5.6	-
Retro-molar area	-	-	25	7.3	-
Commissure	-	-	2	0.5	-
Total	20	100.0	339	100.0	

P for chi-squared test
OED=Oral epithelial dysplasia
SCC=Squamous cell carcinoma

Table 5.5. Distribution according to size of oral epithelial dysplasia (OED)and subsequent development of squamous cell carcinoma (SCC)

Size in (cm)	OED with later SCC No	%	OED with no SCC No	%	*P* value
1	9	45.0	156	46.0	0.9
2	10	50.0	137	40.4	0.3
3	1	5.0	36	10.6	0.4
4	-	-	8	2.3	-
5	-	-	1	0.2	-
6	-	-	1	0.2	-
Total	20	100.0	339	100.0	

P for chi-squared test
OED=Oral epithelial dysplasia
SCC=Squamous cell carcinoma

Table 5.6. Histology of oral epithelial dysplasia (OED) and subsequent development of squamous cell carcinoma (SCC)

Degree of dysplasia	OED with later SCC No	%	OED with no SCC No	%	*P* value
Mild dysplasia*	3	15.0	164	48.3	0.004
Moderate dysplasia$^{\phi}$	4	20.0	100	29.4	0.3
Severe dysplasia#	11	55.0	75	22.1	0.001
Carcinoma in-situ	2	10.0	-	-	-
Total	20	100.0	339	100.0	

OED=Oral epithelial dysplasia
SCC=Squamous cell carcinoma
* chi-square= 3.88
ϕ chi-square= 0.44
chi-square= 5.57

Table 5.7. Treatment method of oral epithelial dysplasia (OED) and subsequent development of squamous cell carcinoma (SCC)

Treatment methods	OED with later SCC		OED with no SCC		
	No	%	No	%	*P* value
Surgical excision*	3	15.0	207	61.0	0.0001
Antifungal drugs$^{\phi}$	12	60.0	52	15.3	0.001
Advice to moderate alcohol and tobacco habits#	5	25.0	80	23.5	0.8
Total	20	100.0	339	100.0	

OED=Oral epithelial dysplasia
SCC=Squamous cell carcinoma
* chi-square = 5.85
ϕ chi-square = 13.4
chi-square = 4.32

Table 5.8. Smoking habits in patients with oral epithelial dysplasia (OED) with and without subsequent malignant transformation

Smoking Habits	OED with later SCC No	%	OED no SCC No	%	*P* value
Status					
None	6	37.5	70	21.4	0.1
Current	7	43.7	180	55.2	0.3
Ex-smokers	3	18.7	76	23.3	0.6
Total	16	100.0	326	100.0	
Cigarette/ week					
1-77	5	50	74	28.9	0.15
> 77	5	50	182	71.0	0.15
Total	10	100.0	256	100.0	
Tobacco type					
Filter cigarettes	6	60	186	73.2	0.3
Non-filter cigarettes	4	40	45	17.7	0.07
Pipe	-	-	11	4.3	-
Rolled-up	-	-	12	4.7	-
Total	10	100.0	254	100.0	
Period of smoking (years)					
1-20	3	30	111	43.3	0.4
> 20	7	70	145	56.6	0.4
Total	10	100.0	256	100.0	
Alcohol					
None	3	20.0	78	27.3	0.5
Current	12	80.0	183	64.2	0.2
Ex-drinkers	-	-	24	8.4	-
Total	15	100.0	285	100.0	
Units/ week					
1-20	5	41.6	145	70.0	0.04
> 20	7	58.3	62	30.0	0.04
Total	12	100.0	207	100.	
Alcohol type					
Beer	3	25.0	54	26.0	0.9
Wine	4	33.3	59	28.5	0.7
Spirits	2	16.6	39	18.8	0.8
Fortified wine	-	-	32	15.4	-
Alcohol	3	25.0	23	11.1	0.1
Total	12	100.0	207	100.0	

P for chi-squared test
OED=Oral epithelial dysplasia
SCC=Squamous cell carcinoma

Table 5.9. Demographic characteristics of patients showing recurrence or additional oral epithelial dysplastic lesions

Variables	Recurrence (no=63)		Second dysplastic lesions (no=37)	
	No	%	No	%
Age (years)				
< 40	4	6.3	2	5.4
40-50	16	25.3	10	27.0
> 50	43	68.2	25	67.5
Total	63	100.0	37	100.0
Gender				
Male	37	58.7	17	46.0
Female	26	41.2	20	54.0
Total	63	100.0	37	100.0
Ethnic-background				
Caucasian	33	71.7	24	68.5
Indian	3	6.5	6	17.1
Pakistani & Bangladeshi	6	13.0	1	2.8
Afrocaribbeans	-	-	-	-
Others	4	8.6	4	11.4
Total	46	100.0	35	100.0

Table 5.10. Clinical and histological aspects of oral epithelial dysplasia lesions that recurred or developed second lesions

Variables	Recurrence of OED (no=63)		Second OED lesion (no= 37)	
	No	%	No	%
Clinical type				
White patch	18	28.5	21	56.7
Red patch	9	14.2	5	13.5
Mixed	36	57.1	11	29.7
Site of lesion				
Labial mucosa	6	9.5	2	5.4
Tongue	20	31.7	11	29.7
Gingiva[#]	2	3.1	2	5.4
Floor of mouth	11	17.4	9	24.3
Buccal mucosa	13	20.6	7	18.9
Other sites[*]	11	17.4	6	16.2
Histology[φ]				
Mild	10	16.9	7	18.9
Moderate	16	27.1	14	37.8
Severe	33	55.9	16	43.2
Treatment method				
Surgical excision	16	25.3	20	54.0
Cryosurgery	12	19.0	5	13.5
Laser excision	8	12.6	3	8.1
PDT[ϕ]	2	3.1	-	-
Antifungal therapy	16	25.3	8	21.6
No active treatment[°]	9	14.2	1	2.7

[#] gingiva and alveolar ridge combined
[*] other sites include soft palate, retromolar area
[φ] histology only known for 59 cases
[ϕ] photodynamic therapy
[°] patients advised to stop smoking and drinking
OED=oral epithelial dysplasia

Table 5.11. Smoking and alcohol consumption of the subjects showing recurrence or/ and second oral epithelial dysplasia lesions

	Recurrence of OED (no=63)		Second OED lesion (no=37)	
	No	%	No	%
Smoking				
None	15	25.8	10	27.7
Current	31	53.4	17	47.2
Ex-smokers	12	20.6	9	25.0
Total	58	100.0	36	100.0
Tobacco type				
Filter cigarette	36	83.7	15	57.6
Non-filter	6	13.9	8	30.7
Pipe	1	2.3	-	-
Roll-up	-	-	3	11.5
Total	43	100.0	26	100.0
Cigarette per day				
<21	2	4.6	1	3.8
>21	41	95.3	25	96.1
Total	43	100.0	26	100.0
Smoking period (years)				
1-19	17	39.5	6	23.0
20-39	22	51.1	8	30.7
>40	4	9.3	12	46.1
Total	43	100.0	26	100.0
Alcohol				
None	17	34.0	13	44.8
Current	26	52.0	15	51.7
Ex-drinkers	7	14.0	1	3.4
Total	50	100.0	29	100.0
Alcohol type				
Beer	8	25.0	5	31.2
Wine	13	40.6	2	12.5
Fortified wine	4	12.5	-	-
Spirits	7	21.8	9	56.2
Total	32	100.0	16	100.0
Units per week				
1-19	2	6.2	1	6.2
20-39	5	15.6	3	18.7
40-59	8	25.0	5	31.2
>59	17	53.1	7	43.7
Total	32	100.0	16	100.0

OED= Oral epithelial dysplasia

Table 5.12. Tobacco and alcohol habits of patient with recurrence or second oral epithelial dysplasia lesions (OED) at last clinical appointment

	Recurrence of OED		Second OED lesion	
	No	%	No	%
Stop smoking and drinking	9[#]	15.2	6[ϕ]	16.2
Reduce smoking and drinking	22	37.2	12	32.4
No change of habits	28[#]	47.4	19[ϕ]	51.3
Total	59	100.0	37	100.0

[#] $P< 0.006$

[ϕ] $P< 0.02$

OED= Oral epithelial dysplasia

Table 5.13. Summary of published cases of oral epithelial dysplasia (OED) that transformed to invasive squamous cell carcinoma

Authors (year)	Country	No	Number of invasive SCC	Transformation time (yr.)	Transformation rate %
Mincer et al, 1972	USA	45	5	up to 8	11
Banoczy and Csiba, 1976	Hungary	68	9	1-20 Mean 6.3	13.2
Pindborg et al, 1977	India	61	4	up to 7	6.6
Gupta et al, 1980	India	73	6	10 Mean 8.5	8.2
Silverman et al, 1984	USA	22	8	Mean 8.1	36.4
Vedtofte et al, 1987	Denmark	47	3	Mean 3.9	6.3
Cregg & Cowan, 1992	N. Ireland	135	24	15 years	17.7
Lumerman et al, 1995	USA	44	7	up to 6.5	16
Present study 1997	UK	359	20	Mean 3.3	5.5
Total		854	86		10.0

(Modified from Lumerman et al, 1995)

Table 5.14. Follow-up studies on patients with epithelial dysplasia who received no further surgical treatment after diagnostic biopsy

Authors	No of cases	Unchanged	Improved, regressed, or disappeared	Increased in size	Developed SCC
Mincer et al, 1972	22	10 (45.4%)	5 (22.7%)	5 (22.7%)	2 (9.0%)
Banoczy et al, 1976	22	10 (45.4%)	2 (9.0 %)	2 (9.0%)	8 (36.3%)
Pindborg et al, 1977	47	33 (70.2%)	9 (19.1%)	1 (2.1)	4 (8.5%)
Total	91	53 (58.2%)	16 (17.6%)	8 (8.7%)	14 (15.3%)

(After Lumerman et al, 1995)
SCC=Squamous cell carcinoma

Table 5.15. Follow-up studies on patients with epithelial dysplasia whose lesions were excised

Authors	No of cases	Cured	Recurred	Developed SCC
Mincer et al, 1972	20	10 (50.0%)	7 (35.0%)	3 (15.0%)
Banoczy et al, 1976	45	43 (95.5%)	1 (2.2%)	1 (2.2%)
Vedtofte et al, 1987	61	46 (75.4%)	12 (19.6%)	3 (5.0%)
Total	126	99 (78.5%)	20 (15.8%)	7 (5.5%)

(Modified from Lumerman et al, 1995)
SCC=Squamous cell carcinoma

Table 6.1. Demographic of patients with oral epithelial dysplasia among users and non-users of tobacco or alcohol

	Non-users		Users		*P* value
	No	%	No	%	
Age (years)					
< 45	9/37	24.3	95/419	22.6	0.8
45-54	4/37	10.8	95/419	22.6	0.09
55-64	12/37	32.4	118/419	28.1	0.5
65-74	5/37	13.5	82/419	19.5	0.3
75+	7/37	18.9	29/419	6.9	0.01
Gender					
Male	18/37	48.6	251/419	59.9	0.1
Female	19/37	51.3	168/419	40.0	0.1
Ethnic-background					
Caucasians	24/37	64.8	260/383	67.8	0.7
Asians	10/37	27.0	93/383	24.2	0.7
Afrocaribbeans	3/37	8.1	20/383	5.2	0.4
Others	-	-	10/383	2.6	

P value for chi-square test

Table 6.2. Sites of the oral epithelial dysplastic lesions in users and non-users of tobacco plus alcohol

Site	Non-users		Users		*P* value
	No	%	No	%	
Labial mucosa	3	8.1	41	9.7	0.7
Tongue	11	29.7	113	26.9	0.7
Gingiva	6	16.2	37	8.8	0.1
Floor of mouth	6	16.2	86	20.5	0.5
Buccal mucosa	7	18.9	84	20.0	0.8
Soft palate	4	10.8	31	7.3	0.4
Retro-molar area	-	-	24	5.7	-
Commissure	-	-	3	0.7	-
Total	37	100.0	419	100	

P for chi-squared test

Table 6.3. Clinical type and histology of the primary oral epithelial dysplastic lesion in users and non-users of tobacco plus alcohol

	Non-users		Users		*P* value
	No	%	No	%	
Clinical type					
White patch	16/37	43.2	216/419	51.5	0.3
Red patch	2/37	5.4	7/419	1.6	0.1
Mixed	17/37	45.9	182/419	43.4	0.7
Ulcer	1/37	2.7	14/419	3.3	0.6
Lump	1/37	2.7	-	-	-
Histology					
Mild dysplasia	19/37	51.3	148/419	35.3	0.05
Moderate dysplasia	7/37	18.9	128/419	30.5	0.1
Severe dysplasia	10/37	27.0	96/419	22.9	0.5
Carcinoma in-situ	1/37	2.7	11/419	2.6	1.0*

* Fisher's exact test

P for chi-squared test

Table 6.4. Treatment and clinical behavior of the primary dysplastic lesions in users and non-users of tobacco plus alcohol

	Non-users		Users		
	No	%	No	%	*P* value
Treatment					
Surgery	20/37	54.0	173/419	41.2	0.8
Use of drug therapy	7/37	18.9	42/419	10.0	0.09
Others	10/37	27.0	204/419	48.6	0.01
Recurrence after initial therapy	6/37	16.2	39/233	16.7	0.9
Second dysplastic lesions	5/37	13.5	18/232	7.7	0.06
Transformation to malignancy	4/37	10.8	13/322	4.0	0.08*
Status when last reviewed					
Alive disease free	13/23	56.5	160/235	68.1	0.2*
Alive with dysplastic lesions	9/23	39.1	74/235	31.5	0.4
Dead-oral cancer	-	-	1/235	0.4	
Dead-other causes	1/23	4.3	-	-	

* Fisher's exact test

P for chi-squared test

Table 6.5. Features of squamous cell carcinoma in users and no-users of alcohol plus tobacco

SCC	Non-users		Users	
	No	%	No	%
Site				
Labial mucosa	1/4	25.0	-	-
Tongue	-	-	2/13	15.3
Gingiva	2/4	50.0	-	-
Floor of mouth	1/4	25.0	6/13	46.1
Buccal mucosa	-	-	2/13	15.3
Soft palate	-	-	3/13	23.0
Degree of dysplasia				
Mild	-	-	2/13	15.3
Moderate	1/4	25.0	2/13	15.3
Severe	3/4	75.0	7/13	53.8
Carcinoma in-situ	-	-	2/13	15.3
Clinical type of primary lesion				
White patch	-	-	4/13	30.7
Red patch	1/4	25.0	4/13	30.7
Mixed	3/4	75.0	4/13	30.7
Ulcer	-	-	1/13	7.6
Lump	-	-	-	-

CHAPTER 10

FIGURES

Figure 2.1. Distribution of patients with oral epithelial dysplasia according to age and gender

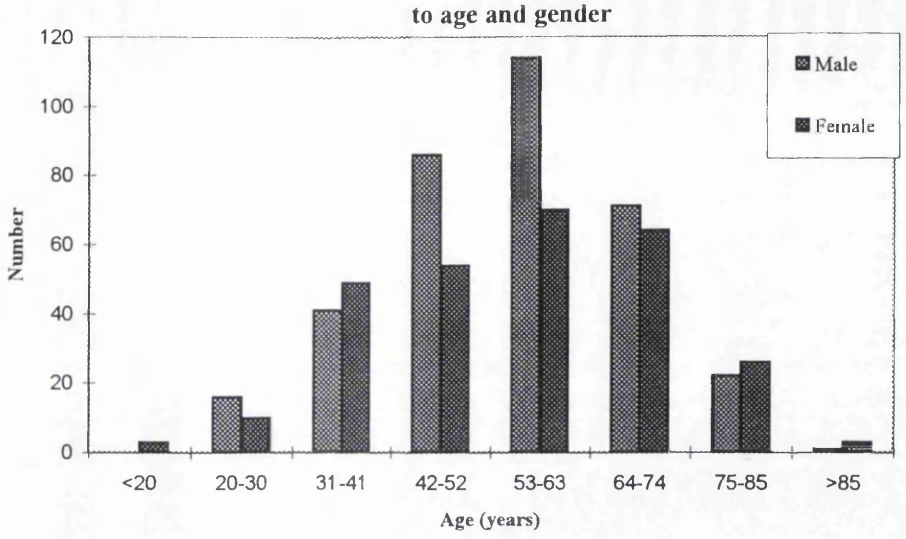


Figure 2.2. Distribution of patients according to gender and histological grading of oral epithelial dysplasia

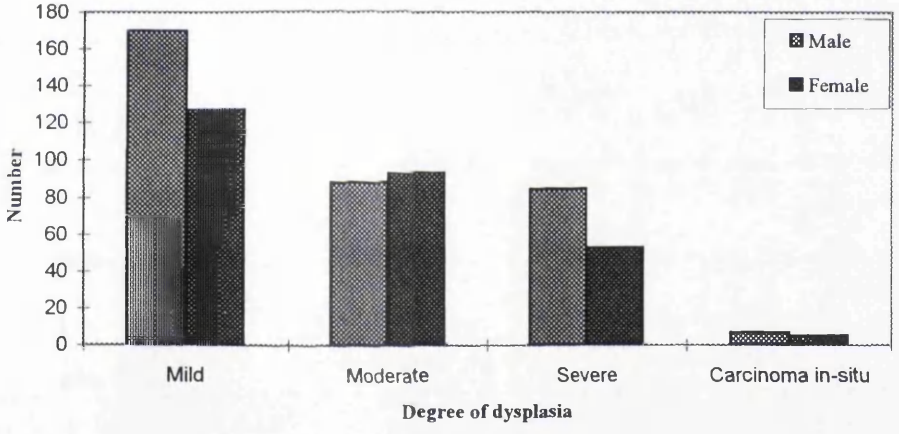


Figure 2.3. Clinical site of oral epithelial dysplasia

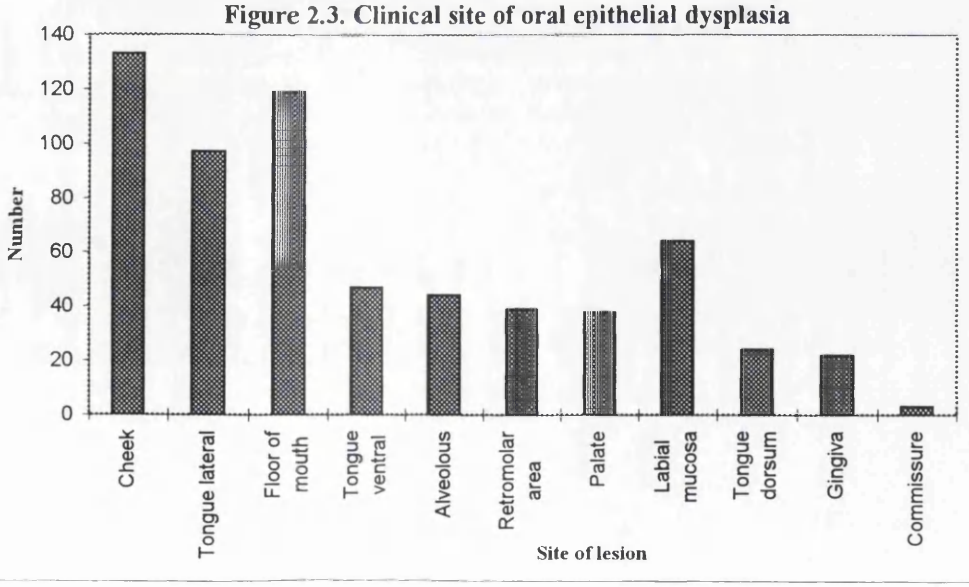


Figure 2.4. Oral site of lesions according to gender

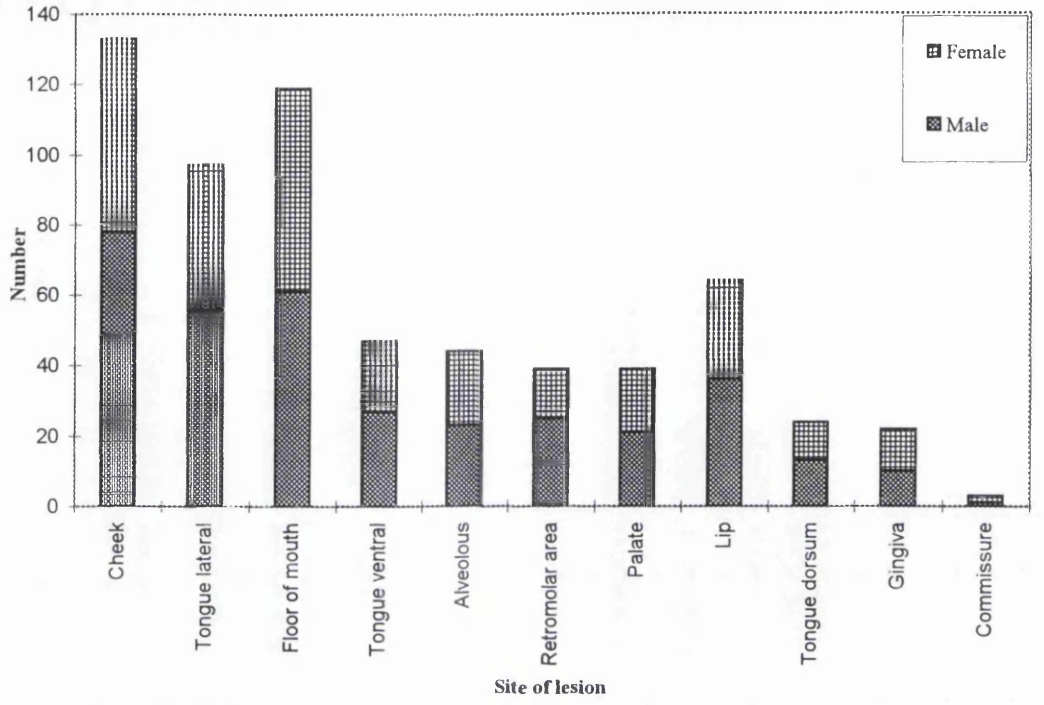


Figure 2.5. Distribution of site of oral epithelial dysplasia according to ethnic-background

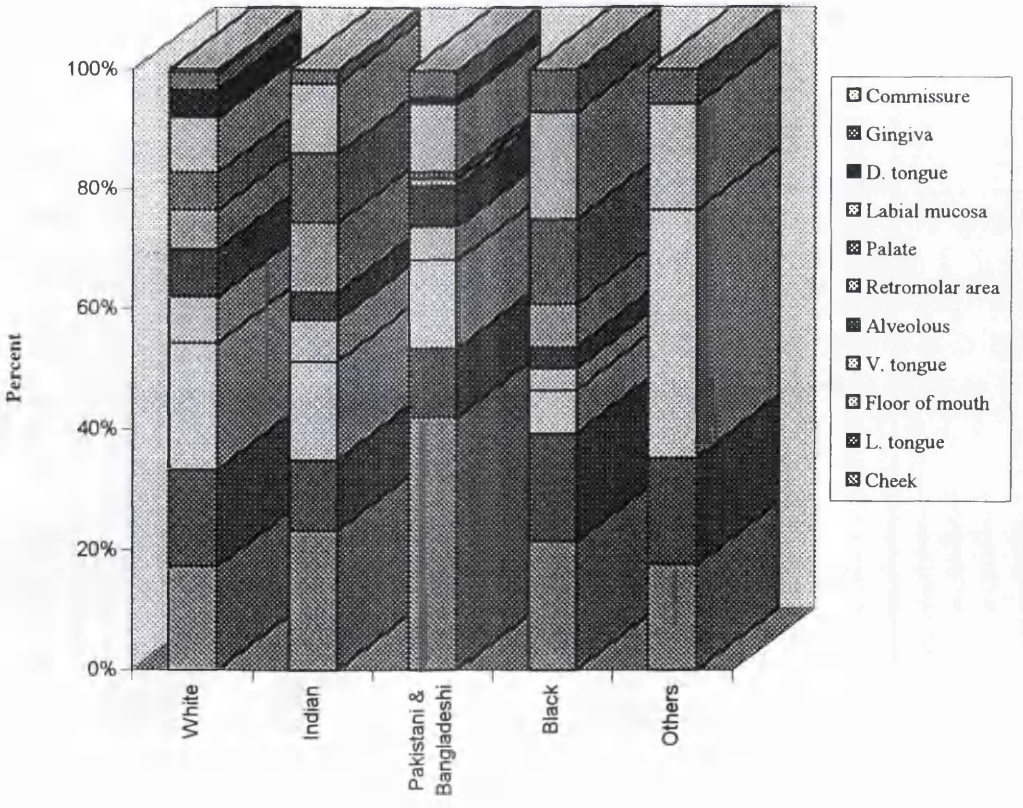


Figure 2.6 Distribution of oral epithelial dysplasia lesions according to site and histology

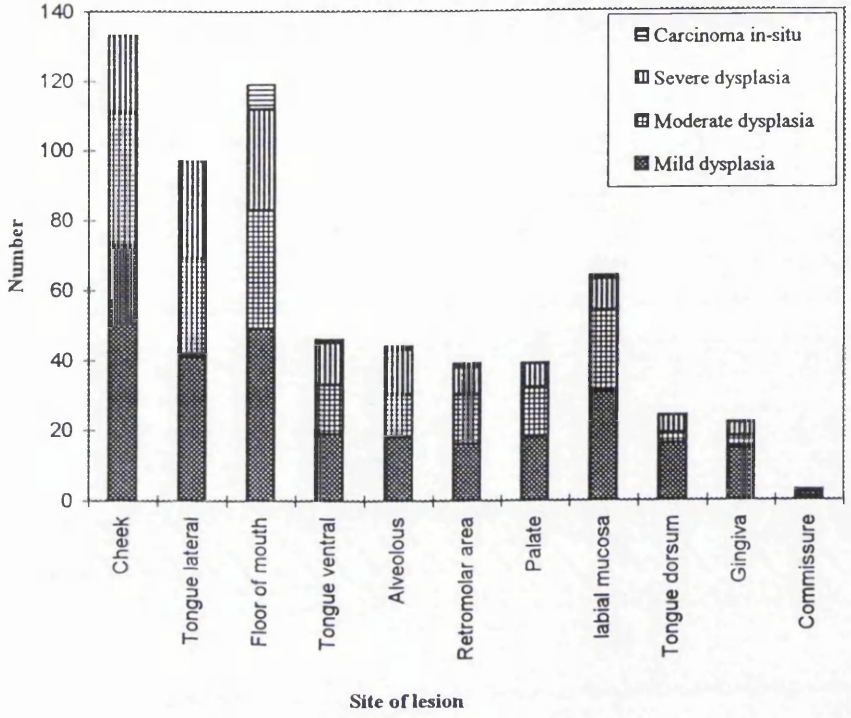


Figure 2.7. Distribution of oral epithelial dysplasia according to clinical presentation and patient gender

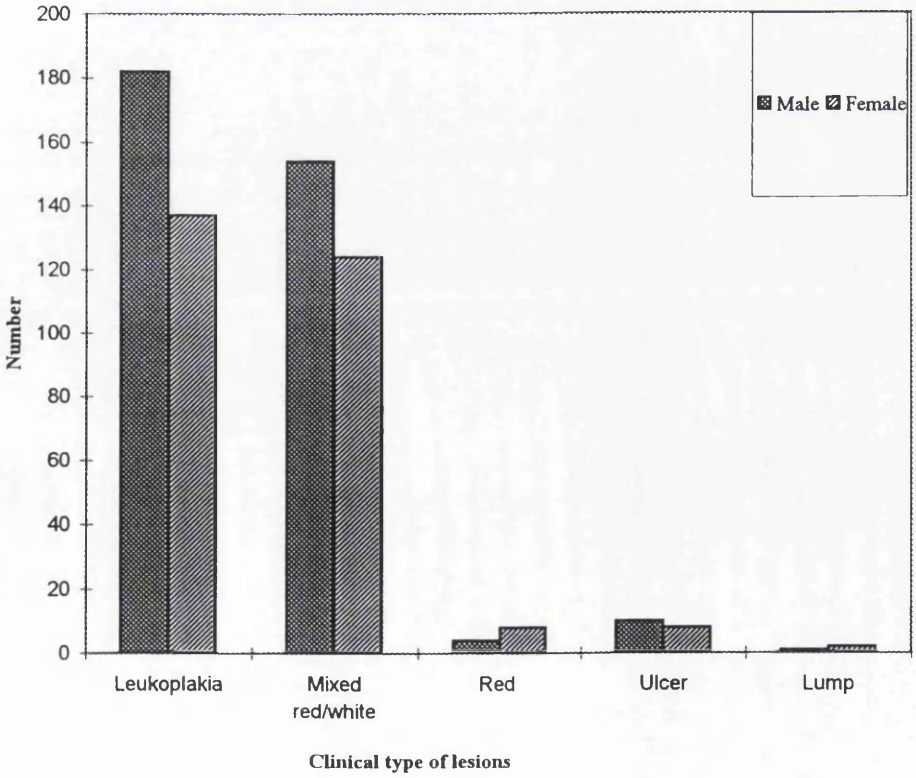


Figure 2.8. Distribution according to self-reported duration of dysplastic lesion and patient gender (324 patients)

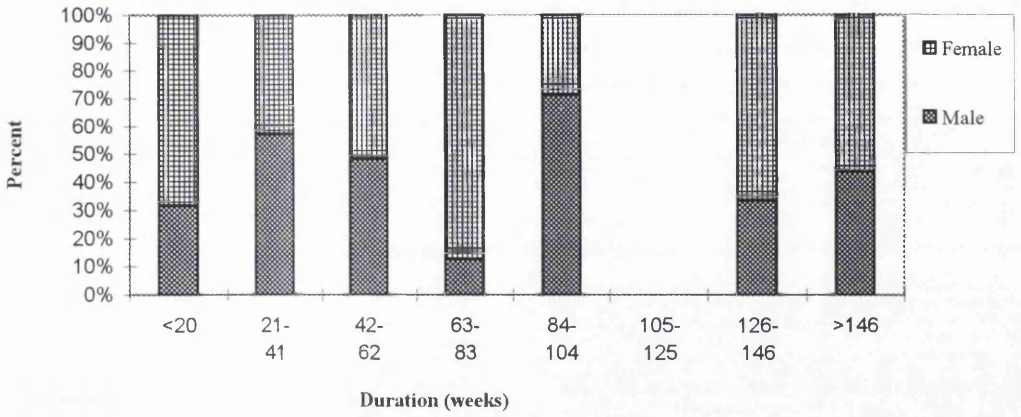


Figure 2.9. Distribution according to treatment methods (427 patients)

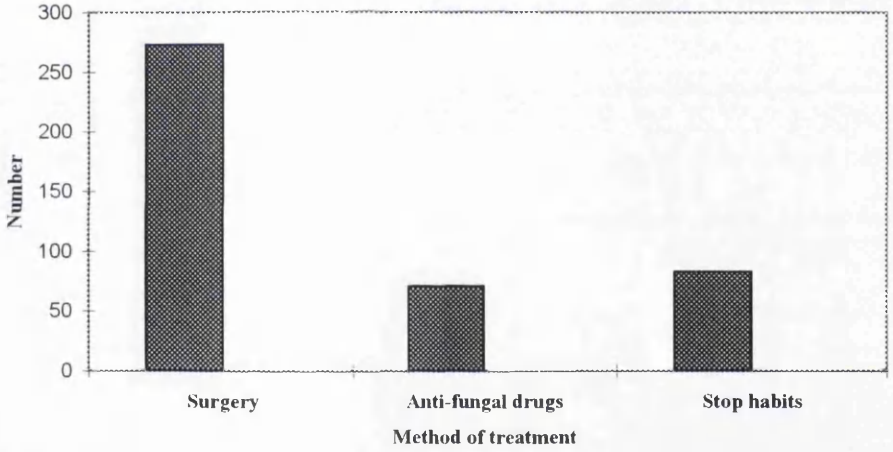


Figure 3.1. Patient details provided in referral letter.

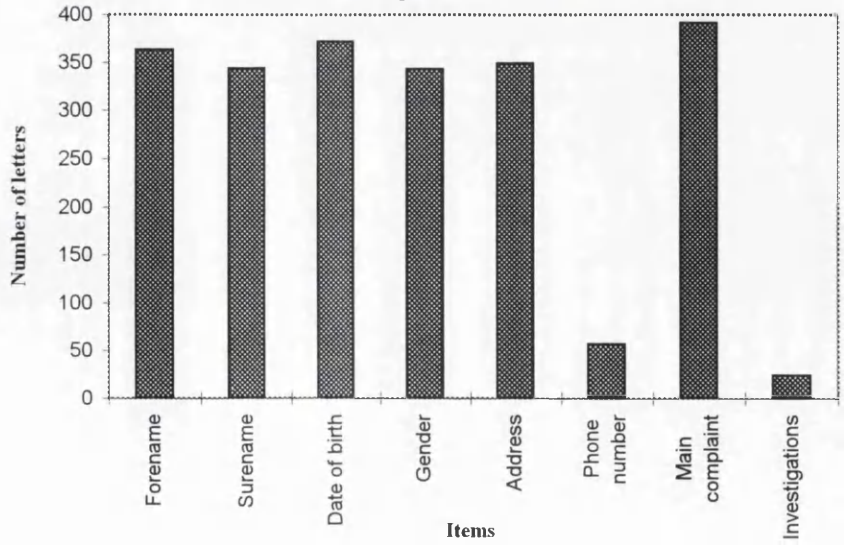


Figure 3.2 Degree of urgency reported in referral letters

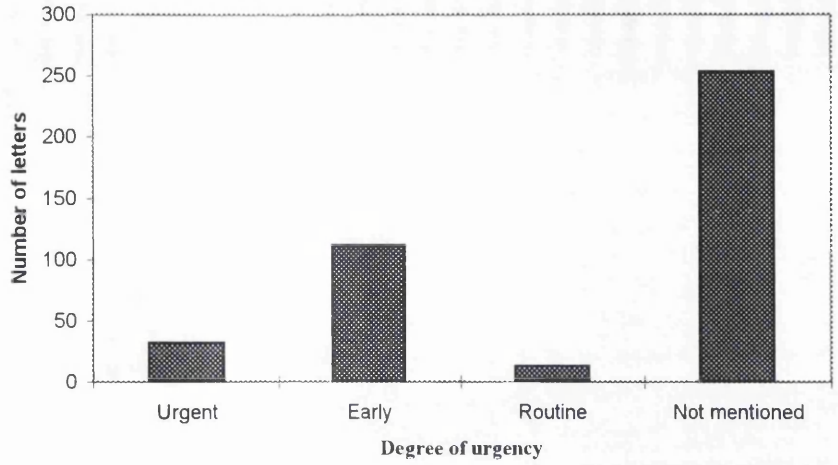


Figure 3.3 Relevant social habits, occupation and ethnic origin of patients mentioned in referral letters

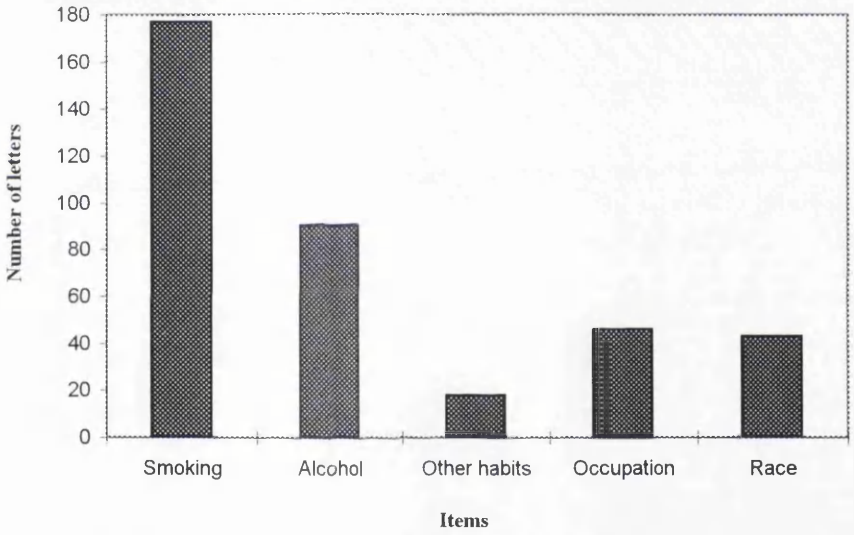


Figure 3.4 Clinical details of the presenting complaint given in referral letters

Figure 3.5 Accuracy of diagnosis by referring clinicians

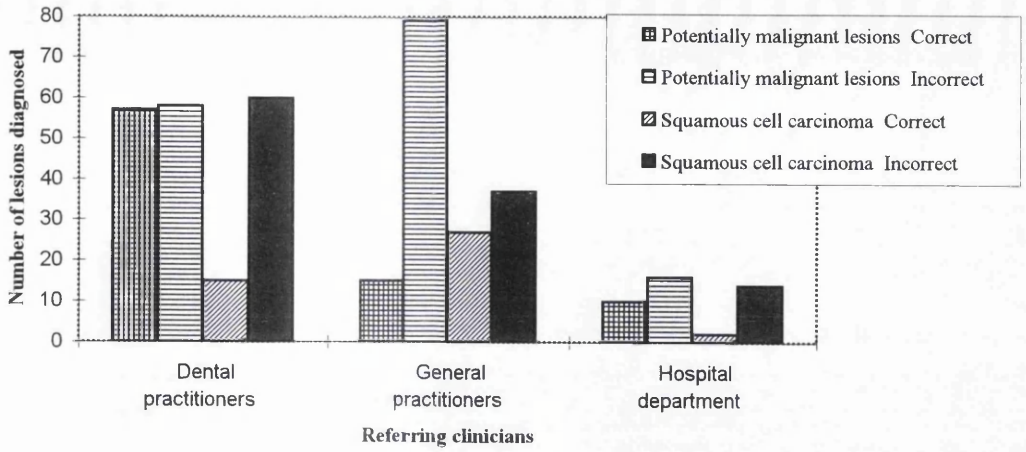


Figure 3.6 Delay between the patients first admitted being aware of symptoms till the first consultation with the practitioners (143 referral letters)

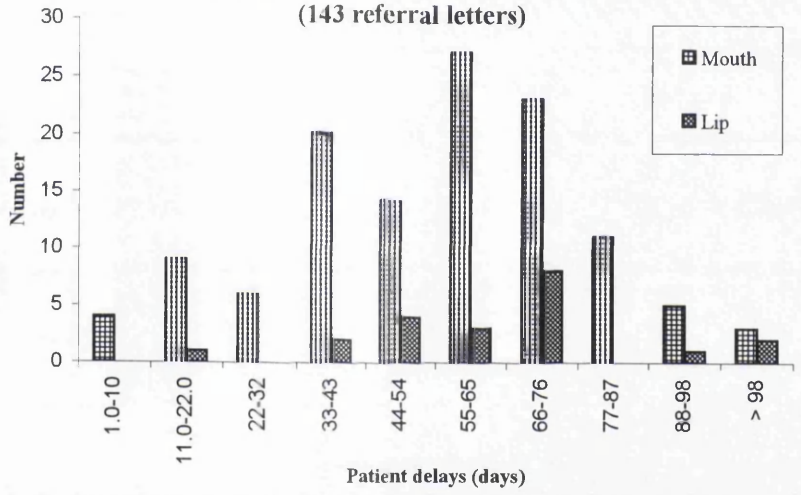


Figure 3.7 Delays between first consultation with GDP and GMP till the referral to hospital (131 referral letters)

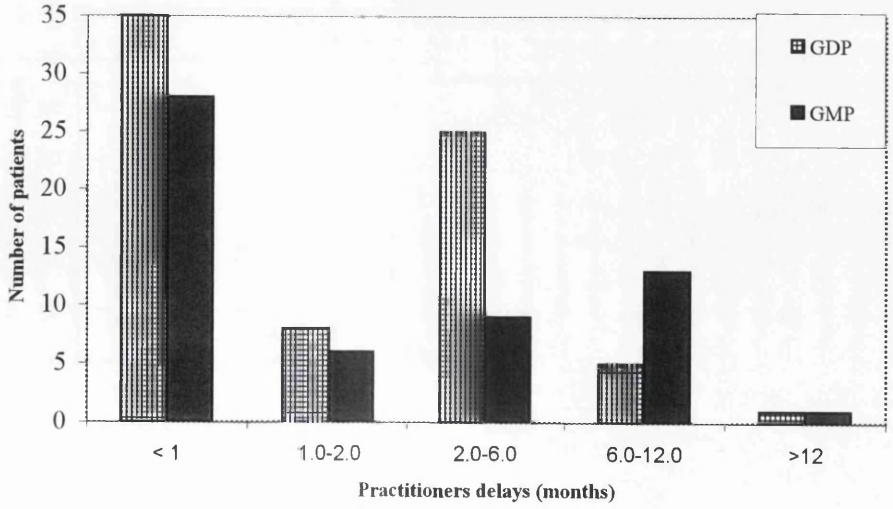


Figure 3.8 Delays between firstt visit to the hospital till the start of the treatment (182 patients)

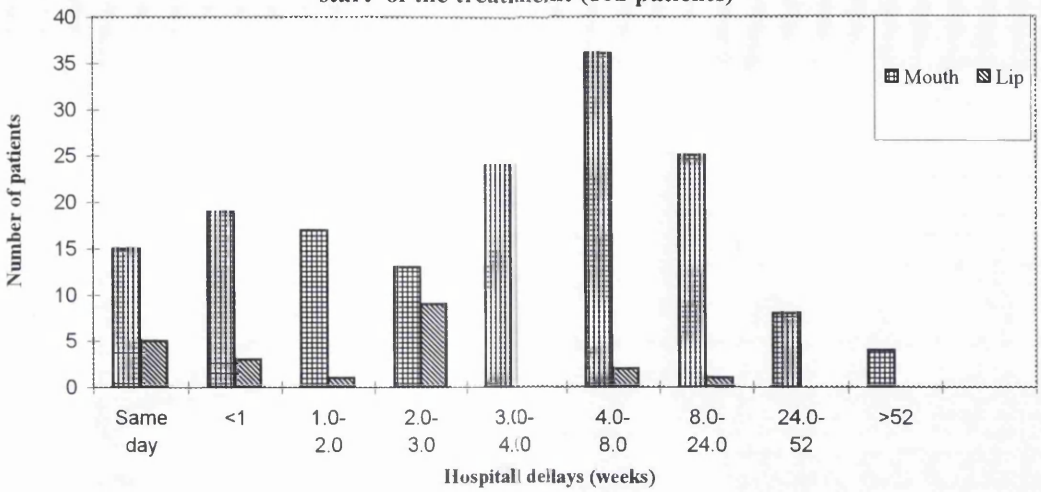


Figure 3.9 Potentially malignant lesions witnessed each year by responding dental practitioners

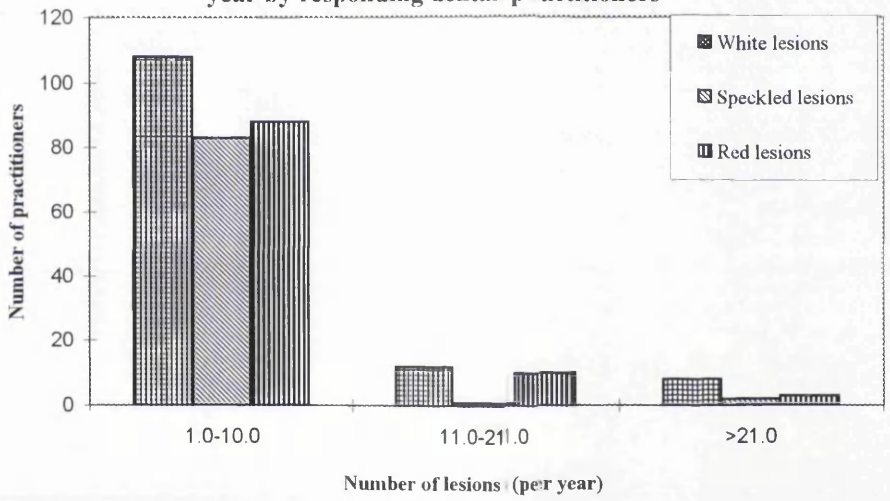


Figure 3.10 Chosen method of examination of potentially malignant or malignant oral lesions by responding dental practitioners (149 respondents)

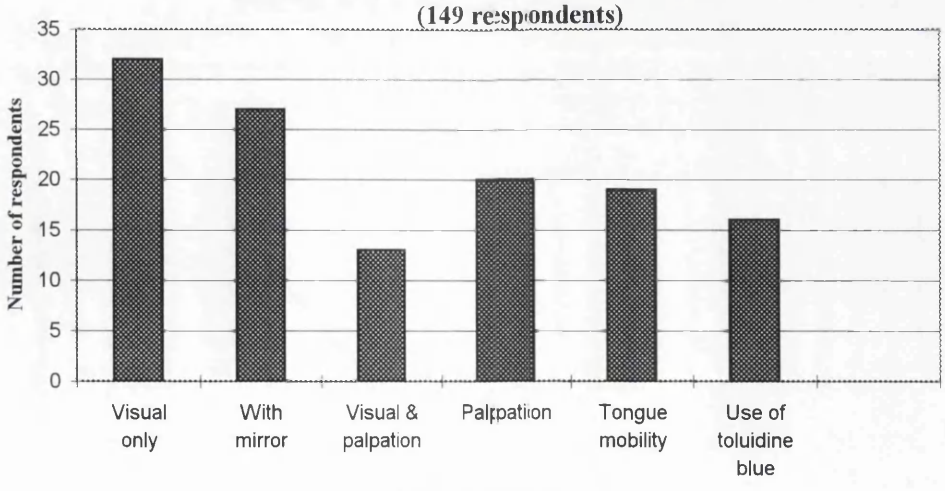


Figure 3.11 Managment of patients with potentially malignant oral lesions by responding dental practitioners

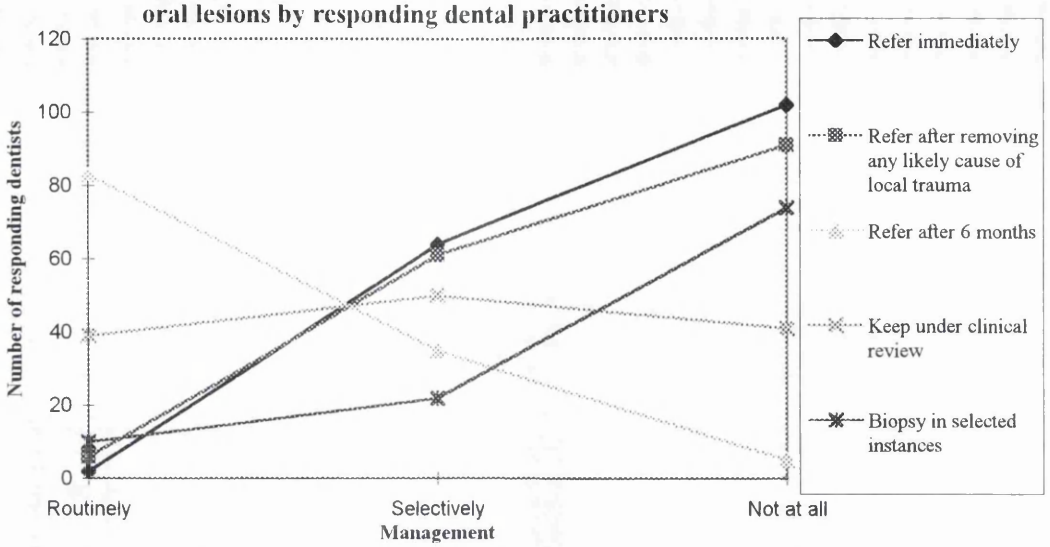


Figure 3.12. Likely method of referral of patients with potentially malignant or malignant lesions by responding dental practitioners

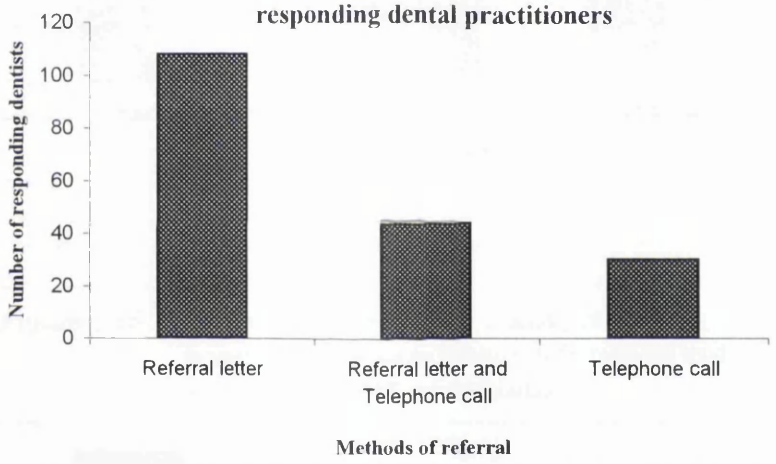


Figure 3.13 Details likely to be included in a referral letter by responding dental practitioners of a patients with a potentially malignant or malignant lesions

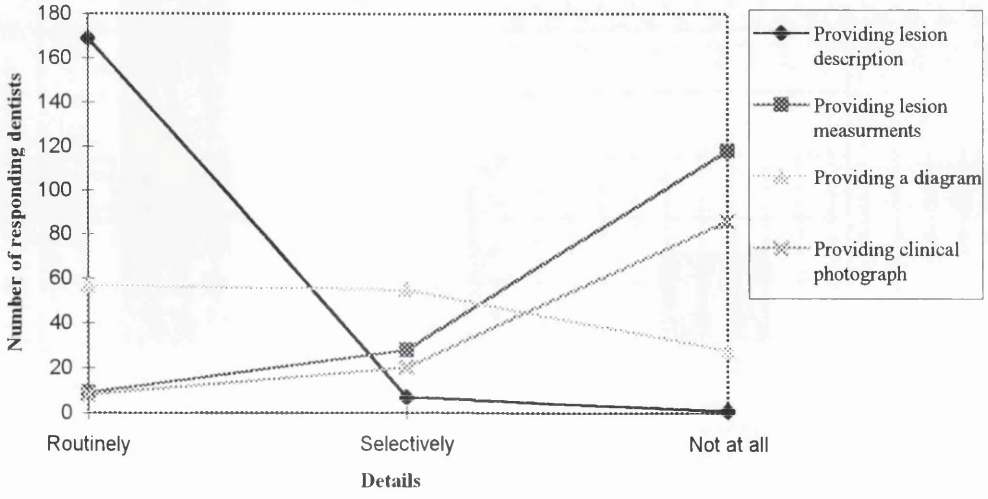


Figure 3.14 Recording tobacco and alcohol consumption and providing of relevant advice by responding dental practitioners (182 respondents)

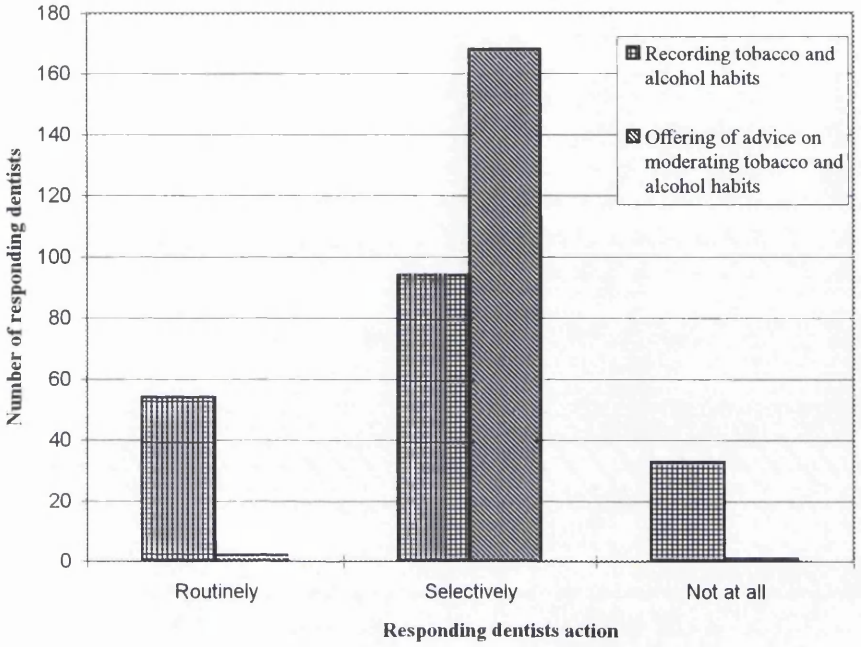
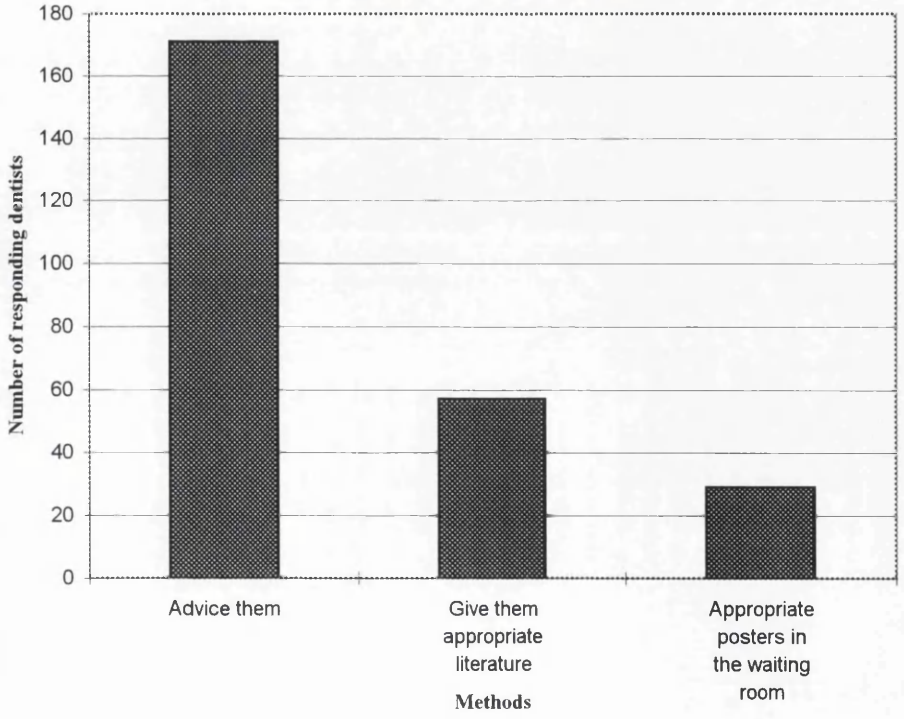


Figure 3.15 Methods used by responding dental practitioners to motivate patients to change their tobacco and alcohol habits (182 respondents)



CHAPTER 11

Appendices

APPENDIX 1

EASTMAN DENTAL INSTITUTE FOR ORAL HEALTH CARE SCIENCES

Department of Oral Medicine
256 Gray's Inn Road
London WC1X 8LD

RETROSPECTIVE FOLLOW-UP DATA ON ORAL EPITHELIAL DYSPLASIA

A Patient details

1. Study number _____

2. Centre:

   Eastman Dental Institute ____
   Royal London Hospital ____
   University College Hospital ____

3. Sex

   Male ____
   Female ____

4. Date of birth: __/__/____

5 a) Country of Birth_____
   b) Ethnicity:

   White ____
   Black Caribbean ____
   Black African ____
   Black others ____
   Indian ____
   Pakistani ____
   Bangladeshi ____
   Chinese ____
   Others ____
   Not known ____

   c) Area of residence in the UK:_____ (postcode)

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B1: Oral Epithelial Dysplasia

1. Study number _____

2. Date oral epithelial dysplasia diagnosed: __/__/____

3. Site of oral epithelial dysplasia: _____
(*Use codings of ICD version 10)

4. Type of the lesion:
   - White patch (leukoplakia) _____
   - Mixed white and red _____
   - Red lesion _____
   - Ulcer _____
   - Lump _____

5. Histology:
   - Mild ___
   - Moderate ___
   - Severe ___
   - Carcinoma in situ ____

6. Size of lesion (cm) ____

7. Treatment:
   - Surgery ___
   - Drug therapy ___
   - No active treatment ___
   - Others ___

8. Prior lesions

   a) Site of prior lesion: _____
   Site from which OED arisen _____
   Size (cm) _____
   Type of lesion _____
   Date of lesion __/__/____

   b) Site of prior lesion: _____
   Site from which OED arisen _____
   Size (cm) _____
   Type of lesion _____
   Date of lesion __/__/____

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C History and referral patterns or disease/medical, dental and social history

1. Study number ____

2. Duration of symptoms _____ (weeks)

3. Date of first consultation __/__/____

4. Type of health specialist:
   - Dentist
   - General Practitioner
   - Hospital department
   - Oral Medicine
   - Oral Surgery
   - Pharmacist
   - Community Dentist

5. a) Date patient referred: __/__/____
   b) Source of referral:

Dentist	____
General Practitioner	____
Hospital department	____
Self-referral	____

6. Symptoms and signs:

Pain duration	____ (weeks)
Dysartheria duration	____ (weeks)
Weight loss duration	____ (weeks)
Dysphagia duration	____ (weeks)
White patch duration	____ (weeks)
Red patch duration	____ (weeks)
Lump duration	____ (weeks)

   Other _____

7. Social class: _____

8. Social history:

Tobacco smoking	____
Tobacco chewing/snuff	____

   1 = current use (currently using or stopped less than one year before diagnosis)

2 = previous use (stopped more than one year before diagnosis)
3 = never used
4 = don't know

Tobacco type
1 = filter cigarette
2 = non-filter cigarette
3 = cigar
4 = roll-up

Cigarette per day _____

Years smoking
a-1-19
b-20-29
c-30-39
d > 39

Years since stopping
a-1-10
b-> 10

Alcohol ____
1 = current use (currently using or stopped less than one year before diagnosis)
2 = previous use (stopped more than one year before diagnosis)
3 = never used
4 = don't know

Alcohol type
1 = beer
2 = wine
3 = fortified wine
4 = spirits

1 Unit = Half pint of beer or lager or, 1 glass of wine or,
1 glass of fortified wine or, 1 measure of spirits.

1.Less than 5 units □ 2.Between 4 and 14 units □
3.Between 15 and 29 units□ 4.Over 30 units □

9. Dental health and history

Patient has dentist?	Yes __	No __
Patient has hygienist?	Yes __	No __

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London WC1X 8LD

D Disease follow-up

1. Study number _____

2. Date of last follow-up: __/__/____

3. Status at last follow-up
   a- Alive disease free
   b- Alive oral dysplasia
   c- Dead other oral cancer
   d- Dead other causes

4. Recurrence of oral epithelial dysplasia: a-Yes
   b-No

   Date of recurrence ___/__/____

   Treatment of recurrence_____

5. Second dysplastic lesions: a-Yes
   b- No

   Site of second dysplastic lesion

   Treatment of second dysplastic lesions

6. Malignant transformation of oral epithelial dysplasia: a-Yes
   b-No

   Date of malignant transformation ___/__/____

   Site of malignant transformation_____

   Treatment of malignant transformation_____

7. Patient habits
   a- still smoking and drinking
   b- reduced smoking and drinking
   c- stopped smoking and drinking

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E Haematological analyses, immunoglobulins and thyroid function tests

1. Study number _____

2. Date of analysis: __/__/____

3. Haemoglobin and indices:

White cells	____
Red blood cells	____
Haemoglobin	____
Packed cell volume	____
Mean cell volume	____
Mean cell haemoglobin	____
Mean cell haemoglobin concentration	____
Platelets	____
Erythrocyte sedimentation rate	____
Neutrophils	____
Lymphocytes	____
Monocytes	____
Eosinophils	____
Basophils	____

4. Haematinic studies:

Serum Ferritin	____
Serum vitamin B12	____
Red cell folate	____
Serum folate	____

5. Serum immunoglobulins:

IgA	____
IgG	____
IgM	____

6. Autoimmune profile:

Parietal cell	____
Reticular cell	____
Mitochondria	____
Smooth muscle	____

E2 Biochemistry and liver function tests

7. Date of analysis: __/__/____

8. Urea and electrolytes:

Urea	____
Sodium	____
Potassium	____
Chloride	____
Bicarbonate	____
Anion Gap	____
Zinc	____

9. Liver function tests

Bilirubin	____
Alkaline phosphatase	____
Aspartate aminotranferase	____
Total protein	____
Albumin	____
Globulin	____
Gamma-glutamyl transferase	____

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London WC1X 8LD

F Radiographic results

1. Study number: _____
2. Date of analysis: __/__/____
3. Number of decayed teeth: _____
4. Number of missing teeth: _____
5. Number of filled teeth: _____
6. Other radiographic findings:

_____
_____
_____
_____

APPENDIX 2

Eastman Dental Hospital for Oral Health Care Sciences
Oral Medicine Department

General Dental Practitioners questionnaire

Please indicate your:

Age:_____

Gender: F □ M □

City/town of practice: _____

Year of graduation in dentistry:_____

Dental School: _____

Dental qualification: 1. BDS □ 2. LDS RCS □

Postgraduate qualifications: 1. Msc □ / 2. Ph.D. □ / 3.VT □ / 4. Primary FDS □ / 5.FDS □
6. DGDP □ / 7. DDCH □ / 8. Other □

Currently are you a: **General dental practitioner-**
1. Trainee□/ 2. associate □/ 3. partner □/ 4. principal □
Hospital based clinician-
1. HO □/ 2. SHO □ / 3. Reg. □/ 4. SenReg □/ 5. Consultant □
6. Other Please State_____.
University based-
1. Lecturer □/ 2. Sen Lect □/ 3. Reader □/ 4. Prof. □
Community DO □ / SDO □ / DDO □.
Other please state

Do you attend BDA meetings ?: 1. Yes □ 2. No □
If yes, how many times per year do you attend:

(please tick)

1. Once □
2. Two - Five □
3. Six - Eleven □
4. Twelve - Seventeen □
5. Eighteen - Twenty-three □
6. More than twenty-three □

Do you attend section 63 events ?: 1. Yes □ 2. No □
If yes, how many times per year do you attend:

(please tick)

1. Once □
2. Two - Five □
3. Six - Eleven □
4. Twelve - Seventeen □

5. Eighteen - Twenty three ☐
6. More than Twenty three ☐

Do you attend University-organised graduate courses (including distance learning)
1. Yes ☐ 2. No ☐

If yes, how many times per year do you attend:

(please tick)

1. Once ☐
2. Two - five ☐
3. Six - Eleven ☐
4. Twelve - Seventeen ☐
5. Eighteen - Twenty three ☐
6. More than Twenty three ☐

Do you attend Royal College of Surgeons courses ?: 1. Yes ☐ 2. No ☐
If yes, how many times per year do you attend:

(please tick)

1. Once ☐
2. Two - Five ☐
3. Six - Eleven ☐
4. Twelve - Seventeen ☐
5. Eighteen - Twenty three ☐
6. More than Twenty three ☐

Have you ever attended a postgraduate course on oral malignancy and/or premalignancy ?
Y ☐ / N ☐
If so please state details:
Date/location/subject/organisers:

Have you ever used the following ?: (please tick)
Department of Health (DoH):
1.Computer assisted Learning (CAL) packages ☐
2.Videotapes ☐
3.Audiotapes ☐
If so please state details:

Do you use your local postgraduate medical/dental center ?: 1. Yes ☐ 2. No ☐
If yes, how many times per year

1. Once ☐
2. Two - Five ☐
3. Six - Eleven ☐
4. Twelve - Seventeen ☐
5. Eighteen - Twenty three ☐
6. More than Twenty three ☐

Which professional journals do you read?: (please tick)

1. BDJ ☐
2. BMJ ☐
3. Probe ☐
11. Int. Dental Journal ☐
12. Journal of Dental Research ☐
13. British Journal of Oral/Maxillofacial Surgery ☐

4. Dental Update □
5. Dental News □
6. Dental Practice □
7. Oral Surg/Oral Med/Oral Path
8. Dentist □
9. Lancet □
10. Quintessence □
14. Journal of the Royal Society of Medicine □
Any others please state:
_____

How many patients with oral cancer did you see when you were a dental undergraduate ?
please state figure:□

Please tick what type:
1. Squamous cell carcinoma □
2. Salivary gland □
3. Kaposi's Sarcoma □
4. Lymphoma □
5. Melanoma □
6. Ameloblastoma □
7. Haemangioma □
8. Others please state:

How many potentially malignant lesions did you see as a dental undergraduate?
Please state figure: □

Please tick what type:
1. Leukoplakia □
2. Eythroplakia □
3. Submucous Fibrosis □
4. Lichen Planus □
5. Atrophic glossitis □
6. Sideropenic anaemia □
7. Other □

How many patients with oral cancer have you seen since graduation in dentistry? please state figure: □

Please tick what type:
1. Squamous cell carcinoma □
2. Salivary gland □
3.Kaposi's sarcoma □
4. Lymphoma □
5. Melanoma □
6. Ameloblastoma □
7. Haemangioma □
8. Others please state □_____

How many potentially malignant lesions have you seen since graduation in dentistry? please state figure: □

Please tick what type:
1. Leukoplakia □
2. Erythroplakia □
3. Submucous fibrosis □
4. Lichen planus □
5. Atrophic glossitis □
6. Candidal leukoplakia □
7. Sideropenic dysphagia □
8. Other □ please state _____

Have you referred patients to hospital/dental school with possible malignant or <u>potentially</u> premalignant lesions ?: Yes □ / No □.
If so, how do/did you do this?: (please tick)

1. By referral letter □ 2. By telephone □ 3. Both □
4. Other please state: _____

Have you ever carried out the biopsy of possible malignant or potentially malignant lesions?: Yes □ / No □

If so why was the biopsy carried out ? Please tick:
1. Lesion looked suspicious □ 2. You were told to do so □
3. Other please state □ _____

What were the final diagnoses ? please tick:
1. Squamous cell carcinoma □
2. Lichen planus □
3. Leukoplakia □
4. Erythroplakia □
5. Melanoma □
6. Salivary gland tumour □
7. Kaposis sarcoma □
8. Candidosis □
9. Other □ _____

(B)-SECTION TWO:
EACH QUESTION CAN HAVE MORE THAN ONE ANSWER. PLEASE TICK THE MOST APPROPRIATE ANSWERS.

Please tick the most common features of an oral tumour?

Site: most likely sites: (please tick)
1. Floor of mouth □
2. Side of tongue □
3. Ventral surface of tongue □
4. Dorsum of tongue □
5. Tongue □
6. Cheek □
7. Commisures of mouth □
8. Upper lip □
8a Lower lip □
9. Soft palate □
10. Hard palate □
11. Oropharynx □
12. Cervical lymph nodes □
13. Retromolar areas of mouth □
14. Other □ (please state):

Size: commonly: (a)5-10mm □ (b)10-20mm □ (c)20-30mm □ (d)Other □ _____mm.

Colour: 1. White □/ 2. Red □/ 3. Speckled □

1. Pain □ / 2. No Pain □ / 3. Variable pain □

4. Paraesthesia □ / 5. Anaesthesia □ / 6. Unlikely □

Tooth mobility: 7. Yes □ 8. No □ 9.Unlikely □

Pathological fracture: 10. Yes □ 11. No □ 12.Unlikely □

Dysphagia yes☐ no☐ unlikely☐/ **Dysartheria** yes☐ no☐ unlikely☐/ **Dysphonia** yes☐ no☐ unlikely☐

Suggest the <u>most common</u> causes of oral squamous cell carcinoma?:

1. Tobacco ☐
2. Alcohol ☐
3. HIV disease☐
4. Leukaemia☐
5. Syphilis ☐
6. Candidal infection ☐
7. Malnutrition☐
8. Poor oral hygiene / dental neglect ☐
9. Previous oral cancer ☐
10. Epstein-Barr virus ☐
11. Herpes Simplex virus ☐
12. Human Papilloma virus ☐

Do you examine the oral mucosa of your patients regularly?: Yes ☐ No ☐

If yes, how? please tick any of the following that apply:

1. Visually ☐ 2. Need good light ☐ 3. Need mirrors ☐ 4. Tongue mobility ☐
5. Palpation ☐ 6. Toluidine Blue staining ☐ 6. Others ☐ _____

Does your examination of the oral soft tissues include :(please tick)

Floor of mouth	1.Yes ☐	10. No ☐
Side of tongue	2.Yes ☐	11. No ☐
Ventral of tongue	3.Yes ☐	12. No ☐
Dorsum of tongue	4.Yes ☐	13. No ☐
Cheeks	5.Yes ☐	14. No ☐
Lips	6.Yes ☐	15. No ☐
Palate	7.Yes ☐	16. No ☐
Oropharynx	8.Yes ☐	17. No ☐
Cervical lymph nodes	9.Yes ☐	18. No ☐

How many <u>potentially</u> malignant lesions do you see each year ?, (please state figure)

White lesions☐
Red lesions☐
Red and white lesions☐
Lumps ☐
Ulcers☐
Others☐

How would you manage a patient with a <u>potentially</u> malignant oral lesion? please tick

	Not at all	Selectively	Routinely
Refer to specialist immediately	1☐	2☐	3☐
Refer after removing any likely cause of local trauma	1☐	2☐	3☐
Refer after six months	1☐	2☐	3☐
Keep patients under clinical review	1☐	2☐	3☐
Offer biopsy on selected cases	1☐	2☐	3☐

How would you detail suspicious lesions in referral letters:

	Not at all	Selectively	Routinely
By writing a description	1☐	2☐	3☐
Providing lesion measurements	1☐	2☐	3☐
Providing a diagram	1☐	2☐	3☐
Providing a clinical photograph	1☐	2☐	3☐

Other-please state_____

Approximately How many suspicious lesions have you referred each year ? (please state figure)

White lesions ☐
Red lesions ☐
Red and white lesions ☐
Lumps ☐
Ulcers ☐
Others ☐

Do you think routine screening for oral cancer and precancer at a dental check-up is likely to be effective?

1.Yes☐ 2. No☐ 3. Undecided☐

In an optimally resourced oral screening programme how regularly do you believe patients be invited for oral examination:(please tick)

1. Every 3 months ☐
2. Every 6 months ☐
3. Every year ☐
4. Every 2 years ☐
5. Every 3 years ☐
6. Every 4 years ☐
7. Others ☐

Please indicate if you believe that oral cancer mortality might be better reduced by supporting more vigorous health:

Promotion 1.Yes ☐2.No ☐ 3.Undecided ☐
Screening process 1.Yes ☐2.No ☐ 3.Undecided ☐

Do you regularly record information about patient's smoking and alcohol consumption:

1.Not at all ☐ 2.Selectively ☐ 3.Routinely ☐

Do you or any of your staff regularly offer patients who smoke or drink, health promotion advice on moderating these habits:

1.Not at all ☐ 2.Opportunistically ☐ 3.Routinely ☐

How do you motivate your patients to changing their habits or life style:

1. Advising them ☐
2. Giving them appropriate literature ☐
3. Placing appropriate posters in your waiting room ☐

Are you aware of the possible options for referral of patients with suspected oral malignancy and premalignancy within reasonable distance from your practice:

1. Yes ☐ 2. No ☐

Please state these:

Do you know the oral surgeons/physicians in your health region or district, where their clinics are held and when:

1. Yes ☐ 2.No ☐ Please give details:

Appendix 3

Eastman Dental Hospital For Health Care Sciences
Oral Medicine Department

Oral Epithelial Dysplasia Study

PATIENT DETAILS: Name_____

Hospital number_____ D.O.B: _____ Gender: M / F

ETHNIC BACKGROUND
1.White □ 2. Black Caribbean □ 3. Black African □ 4. Black others □ 5. Indian □
6. Pakistani □ 7. Bangladeshi □ 8.chinese □ 9.Others □

ADDRESS
POSTCODE

OCCUPATION

MARITAL STATUS:
1.Married □ 2.Single □ 3.Divorced □ 4.Widowed □ 5.Separated □
6.Live with partner □ 7.Unknown □

CHILDREN YES □ NO □ ARE THEY LIVING AT HOME : YES □ NO □
MALE AGE
FEMALE AGE

EDUCATION
1. Did you leave school at 16 years of age 1.YES □ 2.NO □ 3.At an earlier date □
2. Did you leave school with qualifications 1.YES □ 2.NO □
3. GCSE □
4. CSE □
5. O'Level □
6. A'Level □
7. CPVE (certificate of prevocational education) □
8. University Degree □
9. Postgraduate Degree □
10. Others □ please state _____
11. No formal qualifications or subsequent educational courses or apprenticeships □

PREVIOUS HISTORY OF CANCER ?
YES □ NO □
Treatment received?_____

HISTORY OF PRECANCER PROBLEMS ?
YES □ NO □
Treatment received?_____

FAMILY HISTORY OF CANCER ?

Please tick YES ☐ NO ☐
Details:_____

FAMILY HISTORY OF PRECANCER?
YES ☐ NO ☐
Details:_____
MEDICAL HISTORY

REFERRED BY
1.Dentist ☐ 2.General medical practitioner ☐ 3.Hospital Department ☐
4.Yourself ☐ 5.Others ☐ please state_____

WHO FIRST NOTICED THE LESION
1.Dentist ☐ 2.General medical practitioner ☐
3.Hygienist ☐ 4.Other ☐ please state_____

TREATMENT RECEIVED BEFORE REFERRAL TO THIS CLINIC:
1. None ☐ 2.Received Drugs ☐ (please name):

3. Other treatment ☐ (please specify):

DURATION OF LESION(S) before you were referred to this clinic _____months.

SITE OF LESION(S)_____

TYPE OF LESION(S) please tick:
1.White patch ☐ 2.Red patch ☐ 3.Mixed red and white patch ☐
4.Ulcer ☐ 5.Others ☐ please state_____

SIZE OF THE LESION(S):
A.1cm ☐ B.2cm ☐ C.3cm ☐ D.4cm ☐ E. Other ☐_____

Histopathological diagnosis:

date ___/___/___. Report No:_____ **Other information e.g. Candida count?**

1. Mild dysplasia ☐
2. Moderate dysplasia ☐
3. Severe dysplasia☐
4. Carcinoma in situ ☐
5. Early invasive SCC ☐

HAVE YOU EVER SMOKED CIGARETTES ?
1. Never ☐ 2. Not for 10 years ☐ 3. Within the last 10 years ☐ 4. Current user ☐
CIGARETTES SMOKED PER DAY DURING THE LAST TEN YEARS:_____

HOW LONG HAVE YOU BEEN SMOKING?_____years/months

Type of cigarettes smoked
1. Filter □ 2. Non-filter □

HAVE YOU EVER SMOKED CIGARS ?
1.Never □ 2.Not for 10 years □ 3.Within the last ten years □ 4.Current user □
CIGARS SMOKED PER DAY DURING THE LAST TEN YEARS:_____

DO YOU OR DID YOU
1.Roll own □ 2.Use a pipe □ 3.Chew tobacco □ 4.Take snuff □ 5. Betel nut □
OUNCES OF TOBACCO (PER WEEK):_____

CHEW TOBACCO?
1.Never □ 2.Not for 10 years □ 3.Within the last 10 years □ 4.Current user □
OUNCES OF TOBACCO CHEWS (PER WEEK):_____

DO YOU DRINK ALCOHOL: YES □(INDICATE BELOW) NO □
1.Alcoholic beer □ 2.Non-alcoholic beer (including lager and cider) □
3.Wine □ 4.Fortified Wine (Sherry, Port, Martini etc.) □
5.Spirits □ please state_____

UNITS (PER WEEK)OF ALCOHOL:

1 UNIT = Half pint of beer or lager or, 1 glass of wine or,
1 glass of fortified wine or, 1 measure of spirits.

1.Less than 5 units □ **2.**Between 4 and 14 units □
3.Between 15 and 29 units□ **4.**Over 30 units □

DENTITION AND ORAL HYGIENE

CURRENTLY REGISTERED WITH A DENTIST: YES □ NO □
VISIT THE DENTIST REGULARLY: YES □ NO □
HOW OFTEN
1.Every 6 months □ 2.Every 12 months □
3.Every 18 months □ 4.Other □ please state_____
HOW OFTEN DO YOU CLEAN YOUR TEETH
1.Once a day □ 2.Twice a day □ 3.Three times per day □
4.More than three times per day □ 5.Do not clean teeth □

USE of DENTAL FLOSS OR OTHER FORM OF INTERDENTAL CLEANING
YES □ NO □ type/brand_____

Teeth missing_____ Decayed_____ Filled_____

DO YOU WEAR A DENTURE YES □ NO □
1.Lower partial denture □ 2.Lower complete denture □
3.Upper partial denture □ 4.Upper complete denture □ 5.Bridge □

YEARS WEARING DENTURE _____.

HOW MANY MONTHS / YEARS BETWEEN HAVING TEETH EXTRACTED AND DENTURE(S) FITTED:_____

USE OF DENTURE CLEANING AGENTS YES □ NO □

TYPE _____**HOW OFTEN USED**_____

USE OF MOUTH WASH YES □ NO □ Type/brand_____

HOW MANY TIMES PER DAY DO YOU USE MOUTHWASH_____

HOW LONG HAVE YOU BEEN USING A MOUTHWASH FOR
1. Between 1 and 10 years □ 2. Between 10 and 20 years □
3. Between 20 and 30 years □ 4. Between 30 and 40 years □
5. Over 40 years □

BLOOD REQUESTED: Date requested [_____]

FBC	Ferritin	Iron	B12	Serum Folate	RBC Folate	Hepatitis C	H.Pylori

Liver function tests							
Bilirubin	APH	AST	Total protein	Albumin	Globulin	GGT	ALT

APH =Alkaline phosphatase
AST= Aspartate aminotransferase
GGT= Gamma-glutamyl transpeptidase
ALT= Alanine transaminase

Publications arising from this thesis

Jaber MA, Porter SR, Scully C, Gilthrope M, Bedi R. (1998). The role of alcohol in non-smokers and tobacco in non-drinkers in the aetiology of oral epithelial dysplasia. Int. J. Cancer 77:333-336.

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