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Mouth Cancer for Clinicians Part 1: Cancer

Abstract: A MEDLINE search early in 2015 revealed more than 250,000 papers on head and neck cancer; over 100,000 on oral cancer; and over 60,000 on mouth cancer. Not all publications contain robust evidence. We endeavour to encapsulate the most important of the latest information and advances now employed in practice, in a form comprehensible to healthcare workers, patients and their carers.
This series offers the primary care dental team, in particular, an overview of the aetiopathogenesis, prevention, diagnosis and multidisciplinary care of mouth cancer, the functional and psychosocial implications, and minimization of the impact on the quality of life of patient and family.
Clinical Relevance: This article offers the dental team a simplified overview of carcinogenesis, and a review of cancers that affect the oral region.
Dental Update 2015; 42: 250–260

Cancer is a potentially lethal disease with many other consequences, especially if treated late, and is the most important area in oral healthcare (Figure 1), affecting not only the mouth, but potentially the jaws, neck, face, and distant regions. Estimates suggest that around half of all American men and one third of all American women will develop a cancer of some type during their lifetimes, mainly in older age.

The mouth cancer situation is worsening with an increase in the disease, and it is widely recognized and accepted that there is an urgent need for the rapid development of better:

- Public awareness and knowledge;
- Professional awareness and knowledge; it is important that a high uptake of a good quality continuing professional development (CPD) programme is encouraged;
- Preventive strategies – action on reducing:
  - tobacco use;
  - alcohol consumption to within recommended levels;
  - areca nut (betel) and other similar habit use.
- Diagnosis and referral;
- Care and after-care.

With increasing understanding of the molecular pathology of cancer, it is anticipated that the development of molecular profiling will aid early diagnosis of lesions likely to transform to malignancy, help the development of new targeted therapies and hopefully thereby to reduce treatment adverse effects.

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What is cancer?

The word tumour in Latin means a swelling but a tumour or swelling is not always a cancer. Some tumours may be caused by inflammation, infections, cysts or fluid-filled lesions or be due to new growths (neoplasms). There are several causes of non-cancerous (benign) neoplasms in the mouth and oropharynx. Benign neoplasms do not invade other organs or spread to other parts of the body, although they can grow to a large size and start to press on

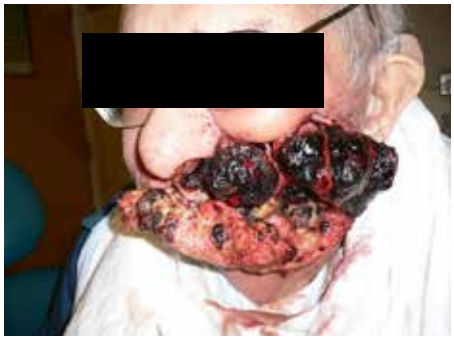

Figure 1. Advanced oral carcinoma in an older gentleman.

surrounding organs and tissues and cause ill-effects. In contrast, those neoplasms that are capable of invading and spreading are referred to as malignant tumours or malignant neoplasms or 'cancer' – they are autonomous and have the capacity to grow rapidly and to metastasize or spread to other tissues.

Cancer, however, is a term that refers to a group of more than a hundred diseases that can originate in many different parts of the body. Most malignant neoplasms grow as solid masses of tissue; some, like leukaemias, grow as cell suspensions but the following features are common to all types of cancer:

- Abnormal cell growth; as cancer cells proliferate, they eventually form tumours;
- Capacity to invade other tissues;
- Capacity to spread to distant organs via blood vessels or lymphatic channels (metastasis).

If cancer is left untreated, it can invade tissues, spread through the body and eventually lead to death.

How does cancer arise?

The trillions of cells that grow, divide and die normally do so in a very organized manner. These processes are tightly controlled by DNA (deoxyribonucleic acid) – the 'blueprint' that instructs cell growth, protein synthesis, function and death. If DNA becomes damaged in a normal cell, that cell then either repairs the damage or the cell dies. DNA damage is often termed a 'mutation'.

DNA damage is often a mutation which leads to altered amino acids and thus changes in cell proteins and activities that may lead to the complex pathophysiology of cancer. Gene changes may be a single point mutation affecting a single DNA nucleotide, or can occur at many levels (eg a gain or loss of entire chromosomes or deletions and insertions, especially at the promoter region of the gene). Disruption of a single gene may also result from integration of genomic material from a DNA virus (eg certain human papillomaviruses, HPV) or retroviruses, which may lead to the formation of oncogenes (cancer-promoting genes). Translocation is yet another process when two separate chromosomal regions become abnormally fused, often at a characteristic location. One example is the Philadelphia chromosome, a translocation of chromosomes 9 and 22, seen in chronic myeloid leukaemia, which results in the production of an oncogenic enzyme called tyrosine kinase (BCR-abl fusion protein). Cancer cell growth differs from normal cell growth. Instead of dying, the cancer cells continue to grow and form new, abnormal cells, which can also invade other tissues.

In order to transform a normal cell into a cancer cell, changes in many genes are typically required (as discussed below), but there are two broad categories of genes which are especially involved:

1. Oncogenes: genes, which may be normal genes inappropriately expressed at high levels in patients with cancers, or may be normal genes altered or changed due to mutation.
2. Tumour suppressor genes (TSGs): genes which normally inhibit cell division and prevent the survival of cells that have damaged DNA. In patients with cancer, TSGs are often disabled.

The genetic abnormalities in mouth cancer are many, and may alter the protein coded for by particular genes and modify cell behaviour. They are mainly at chromosomes 9p21 (TSG is p16), 3p, 17p and 8p, and include mutation of the TSGs p53, retinoblastoma (Rb) and p16 and amplification of cyclin D1. Most mouth cancers have copy number alterations (CNAs), including losses of chromosomal regions 3p and 8p, and gains of 3q, 5p and 8q.

What is responsible for the DNA damage?

DNA mutation is most commonly spontaneous and it is now suggested that the majority are due to 'bad luck', that is, random mutations arising during DNA replication in normal, non-cancerous stem cells. DNA damage may, on rare occasions, be inherited from parents, such as in some breast cancers, or in the head and neck cancers seen in Fanconi anaemia. However, mutations may also be triggered by exposure to certain exogenous factors (mutagens or carcinogens) – termed 'risk factors' – such as those present in tobacco and alcoholic beverages. Malignant transformation may follow an accumulation of exposure to such mutagens and, along with damage from spontaneous mutations, the subsequent accumulation of genetic changes or molecular aberrations. Working in the other direction are factors that may confer some protection (Figure 2).

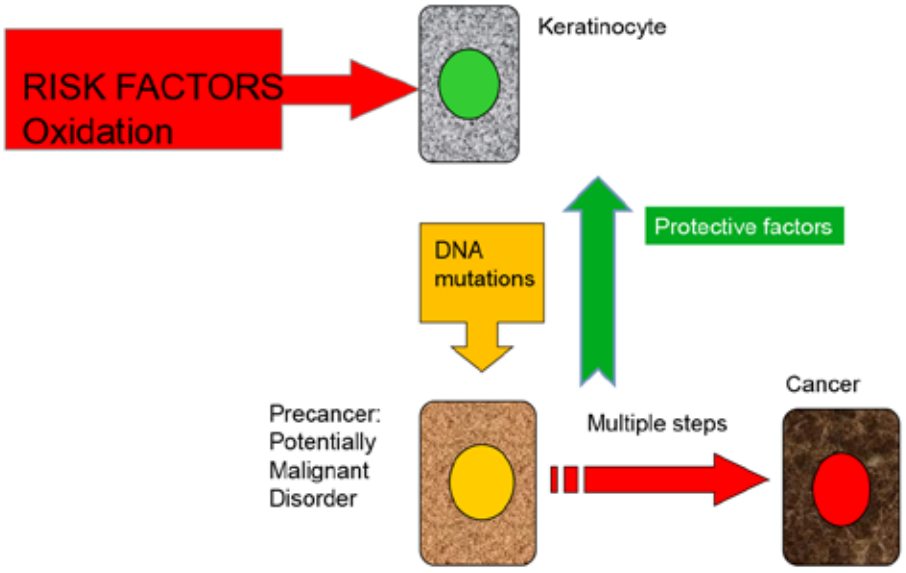

Figure 2. Factors affecting keratinocyte changes.

Mutations may affect cell behaviour:

- In the cell error-correcting machinery; this may cause accumulation of errors rapidly in the cell and its progeny;
- In cell signalling (endocrine) machinery; this leads transmission of the error signals to nearby healthy cells as well;
- By allowing the cells to migrate and disrupt more healthy cells away from the primary site of origin;
- By making the cell immortal so that the abnormal cell refuses to die.

Several DNA mutations are necessary before the affected cells change in appearance and behaviour to a recognizably pre- or potentially malignant cell characterized by an ability to proliferate in a less-controlled fashion than normal (they become autonomous). The progression from a normal cell to a pre-malignant or a potentially malignant cell – oncogenesis (carcinogenesis) – is characterized by an ability of cells to escape normal growth control mechanisms, and to proliferate autonomously. A series of steps lead to the aberrant expression and function of molecules regulating cell signalling, growth, survival, motility, angiogenesis (blood vessel proliferation), and cell cycle control. Crucial genes involved in cell cycle regulation are those encoding proteins in the p53 and Rb pathways (see below). Normally, the body safeguards against cancer in a number of ways, such as: apoptosis (programmmed cell death) or a process by which abnormal cells die of their own accord, helper molecules (some DNA polymerases), possibly senescence or ageing, etc. A key cellular function that is often changed in cancer cells to overcome senescence and to obtain limitless replicative potential is the regulation of the cell cycle. In cancer cells, the damaged DNA is not repaired, and neither does the cell die, but rather it gives rise to more such abnormal cells with abnormal DNA. These new cells all have the same defective DNA of the original cancer cell.

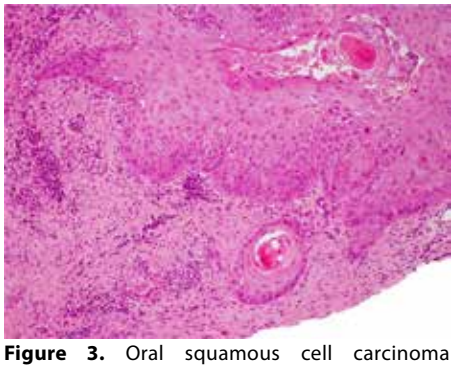

Figure 3. Oral squamous cell carcinoma histopathology.

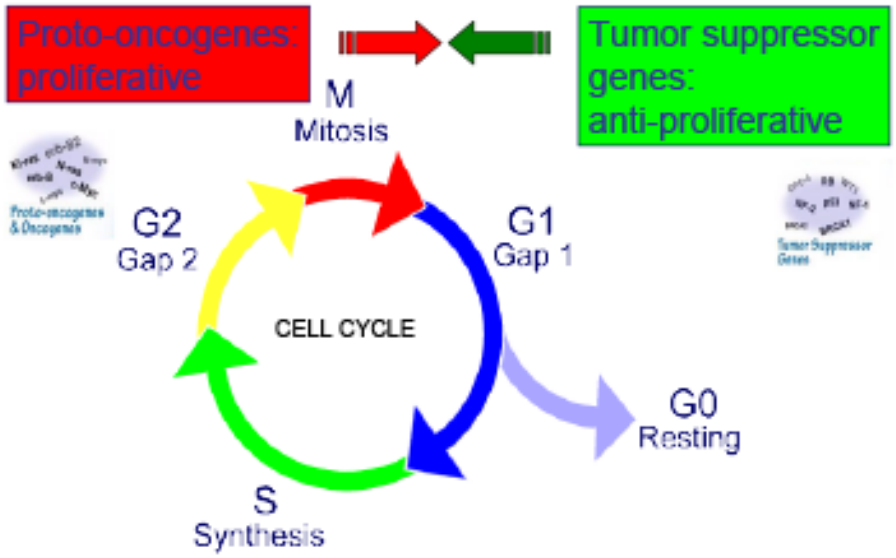

Figure 4. Genes affecting cell cycle control (with acknowledgement to Scully C, Warnakulasuriya S. Cancer of the mouth for the dental team. Comprehending the condition, causes, controversies, control and consequences: 1. General principles. *Dent Update* 2010; **37**; 638–640)

The effects of these changes may *eventually* be visible under the microscope as dysplasia – disordered cell size and arrangement, with abnormal cell divisions (mitoses). Dysplasia can transform to cancer, characterized by malignant epithelial cells (keratinocytes) which proliferate and invade across the epithelial basement membranes as a 'growth' into the underlying tissues. The changes may become evident clinically, usually as a red (erythroplasia) or white lesion (leukoplakia).

Clinical assessment of cancer risk in oral potentially malignant disorders (PMD) (Article 6) is most inaccurate and, although the World Health Organization has criteria to define dysplasia, it may be difficult to make a confident objective categorization of dysplasia owing to a high inter-observer and intra-observer variation in dysplasia assessment. Molecular aberrations have been identified in PMD which increase in number as lesions progress towards malignancy. These cancer-associated genetic changes in PMD are increasingly being used to discriminate leukoplakias with a low risk from those with a high risk of malignant transformation.

Development of technologies like immunohistochemistry, flow cytometry and molecular biologic approaches to cancer diagnosis have profoundly influenced knowledge. Microarray DNA technology has shown that many genes can be involved in oncogenesis. RNA and DNA profiling studies, in particular, have highlighted the molecular heterogeneity of these lesions.

What are the main mouth cancer histological and molecular changes?

Several DNA mutations are necessary before the affected cells change appearance and behaviour to a recognizably pre- or potentially malignant cell characterized by an ability to proliferate in a less-controlled fashion than normal (they become autonomous). The effects of these changes may be seen under the

microscope as dysplasia. This can transform to cancer, characterized by malignant epithelial cells (keratinocytes) which proliferate and invade across the epithelial basement membranes as a 'growth' into the underlying tissues (Figure 3).

Solid malignant neoplasms are composed of a parenchyma that contains cancer tissues and cells, and the other is a stroma that the neoplastic cells induce and in which they are dispersed. All solid tumours require stroma if they are to grow beyond a minimal size; the stroma is essential for malignant neoplasm growth and contains non-malignant supporting tissue such as connective tissue, blood vessels and, very often, inflammatory cells. In addition, cancers also have the property of new blood vessel formation (neoangiogenesis). There are in addition proteoglycans and glycosaminoglycans, interstitial collagens (types I, III and, to a lesser extent, type V), fibrin, fibronectin, fibroblasts, etc. Malignant neoplasms that originate from epithelial cells (carcinomas) have a basal lamina that separates malignant cells from stroma. However, the basal lamina is often incomplete, especially at points of invasion. Indeed, invasion of the epithelial basement membrane is the feature that distinguishes a PMD from cancer. The number of lymph node metastases, the proportion in the neck and extra-nodal spread are important prognostic factors and predictors of distant disease and survival. Metastatic dissemination involves several steps, including degradation of the extracellular matrix as one of the initial steps. Matrix metalloproteinases (MMPs) are known to be involved in the degradation of the extracellular matrix. The CSMD1 gene on chromosome 8p or other genes or micro-RNAs (miRNAs) might be involved in invasion and metastasis. All solid tumours induce neo-angiogenesis, usually by producing angiogenic factors (growth factors that induce sprouting of endothelial cells, and formation of new vessels feeding the tumour). The strongest angiogenesis inducer is Vascular Endothelial Growth Factor (VEGF), the expression of which is a poor prognostic factor.

Loss of heterozygosity (LOH) at chromosomes 3p, 9p and 17p occur in dysplasia, apparently reflecting early carcinogenesis, whereas other alterations at chromosomes 11q, 4q and of chromosome 8 are typically present in carcinomas, probably corresponding to a relatively late phase in carcinogenesis. LOH at chromosome 9p21 is thought to be an early event in mouth carcinogenesis. Cyclin dependent kinase inhibitor 2A (also known as p16Ink4A or p16 [CDKN2A]), which encodes the TSG p16INK4A, is located on chromosome 9p21 and is frequently inactivated in oral squamous cell carcinoma (OSCC). The loss of chromosome 9p21 (which is where CDKN2A is located) and p53 mutations are frequently found in PMD. Mutations in TSGs *p53* and *Rb* are common in tobacco-related malignant oral lesions. CCND1, which encodes cyclin D1, on chromosome 11q13, is amplified or gained in most HPV-negative OSCC. Together with the abrogation of p53, these changes cause cellular immortalization (Figure 4). Tumours with p53 mutation-positive molecular fields have a 10% increase in local recurrence rate compared to those with wild type (non-mutated) p53.

The normal shortening of telomeres (ends of chromosomes) also needs to be overcome to achieve the limitless cell replicative potential in cancers. Abnormal expression of the enzyme telomerase reverse transcriptase (TERT), which controls telomeres, occurs, as well as somatic mutations in p53, overexpression of cyclin D1, or a p16INK4A-insensitive cyclin-dependent kinase 4 (CDK4) mutant. Telomerase or TERT activity is seen in most OSCCs.

Changes in growth factor signalling are also important. One growth factor, the epidermal growth factor receptor (EGFR) contains receptor tyrosine kinases of the Erb-b family which initiate a signalling cascade (through other pathways [Ras–MAPK, PI3K–PTEN–AKT and phospholipase C]). EGFR is also able to translocate to the nucleus and induce CCND1, has been implicated in the malignant transformation of oral keratinocytes, and is often overexpressed in OSCCs. These findings have led to EGFR-specific antibodies (cetuximab) now being used for targeted mouth cancer therapy (Article 12).

Other factors affecting cell signalling include the transforming growth factor-β (TGFβ) pathway – an inhibitory growth factor – and the PI3K–PTEN–AKT pathway, which evades apoptosis and mutations may cause cancer cell increased migration and invasion.

Smoking-related (HPV-unrelated) HNSCCs typically have loss-of-function p53 mutations and CDKN2A inactivation, with amplification of 3q26/28 and 11q13/22. Indeed, TP53 mutations are detected in all HPV-negative mouth cancer cases and there is abrogation of the cell cycle G1/S checkpoint via CDKN2A/B deletion and/or CCND1 amplification in most HPV-tumours, suggesting that, in future, CDK inhibitors may have possible therapeutic uses.

In contrast, most HPV-related oropharyngeal cancers have wild-type TSGs *Rb* and *p53* that are also the first to be inactivated by the viral E6 and E7 oncogenes found in oncogenic HPVs (Article 5). HPV-associated cancers are characterized by amplification of the cell cycle gene E2F1, mutations of PI3 kinase (PIK3-CA), and loss of TRAF3. Mutation and copy number alterations of PI3K pathway components appear particularly common, further supporting a role for HPV inhibiting the p53 and RB pathways. HPV also modulates the epigenome via hypermethylation of genes of the Polycomb repressive complex 2 (eg cadherins), implicated in cancer progression and metastasis.

Recurrences and second primary tumours

There is a high propensity to develop local recurrences after treatment of oral squamous cell carcinoma (OSCC) and a high likelihood that multiple independent tumours will develop in the head and neck mucosa. It is also known that, in at least 35% of OSCCs, the oral carcinoma is surrounded by mucosal epithelium that contains genetic changes which, though macroscopically normal, may be histologically dysplastic. This tumour-adjacent mucosal epithelium characterized by genetic changes has also been termed 'field' changes and, if left after resection, may readily initiate recurrence.

Furthermore, people with one particular cancer are also predisposed to develop another malignant neoplasm – a second *primary* cancer (second primary tumour; SPT). In the case of mouth

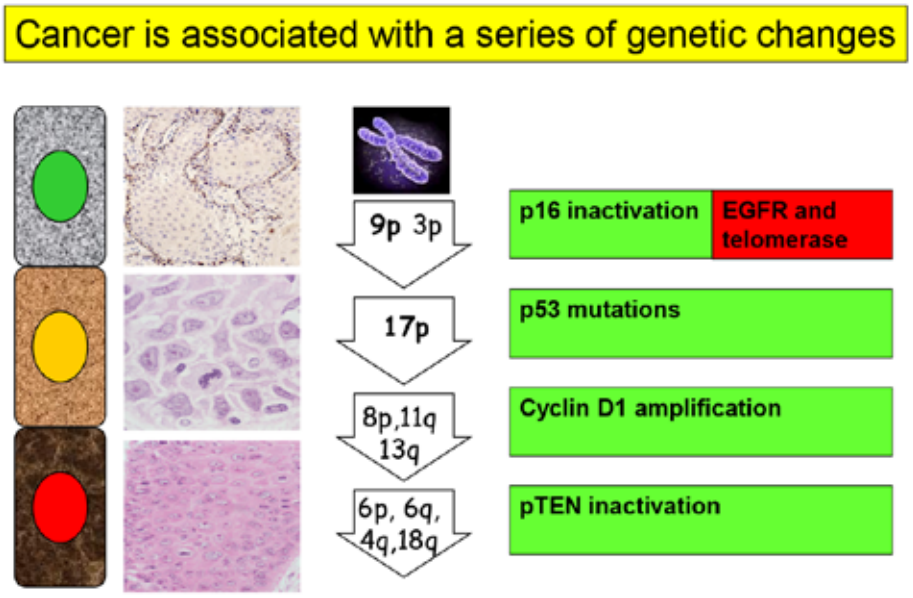

Figure 5. Diagrammatic representation of cellular, histological and molecular changes through dysplasia to carcinoma (see text for terminology).

cancer, SPTs may be in the mouth, or aerodigestive tract (pharynx, larynx, bronchi, oesophagus). This has been termed 'field cancerization' and is linked to the frequent observation of dysplastic changes surrounding these tumours with the occurrence of local recurrences and multiple primary tumours. This concept has now been defined in molecular terms on the basis of the genetic characterization of morphological changes in the squamous epithelium. SPTs are usually in the upper aerodigestive tract: women have a higher risk of a SPT than do men.

Summary

Head and neck squamous cell cancers (HNSCC), the most common of which are mouth cancers, arise as a consequence of DNA mutations caused mainly by free radicals and oxidants, and DNA mutations change various crucial genes and other nucleic acid components involved in cell growth and control (and are potential targets for anti-cancer therapies) (Figure 5) such as:

- Tumour suppressor genes, such as p16, p53 and Rb, control the fate of chromosomally damaged cells and the cell growth cycle;
- Oncogenes, such as the epidermal growth factor receptor (EGFR) gene, PRAD-1, Int-2, hst-1, bcl-1 and H-ras, involved in cell signalling;
- Telomerase genes – control telomerases – enzymes involved in chromosome shortening;
- MicroRNAs, non-coding pieces of RNA which are incorporated into the RNA-induced silencing complex which binds to mRNA to mediate gene expression, with effects similar to tumour suppressors or oncogenes.

Oncogenesis (carcinogenesis) is the progression from a normal cell (eg epithelial cell or keratinocyte) to a pre-malignant or a potentially malignant cell, characterized by a series of steps leading to the aberrant expression and function of molecules regulating cell signalling, cell cycle control, growth, survival, motility and angiogenesis (new blood vessel proliferation).

Cell cycle control is disturbed particularly by over-expression or over-activity of oncogenes, such as the EGFR gene. Cell cycle control is also influenced by TSGs, which help protect cells but, if a TSG is defective, cancer protection is impaired.

DNA technology is making major advances in understanding the pathogenesis and suggesting routes towards developing targeted treatments. Microarray DNA technology has shown that many genes can be involved in oncogenesis and that some genes (eg EGFR) may be potential targets for cancer therapy ('targeted therapy'; Article 12). Single nucleotide polymorphisms (SNPs) – gene areas with altered DNA sequences which may not lead to an amino acid alteration – may, in various genes (TSGs, xenometabolizing enzymes, and DNA repair), also sometimes play a role in cancer. Information from the human genome project together with technological advances has allowed the development of rapid, accurate platforms to carry out comprehensive genetic characterization of germ line and tumour samples, technology referred to as next-generation sequencing (NGS).

What are head and neck cancers?

Cancers can arise in any tissues or organs in the head and neck: there are over 30 different sites that can be affected. Most are squamous cell carcinomas. In the head and neck region the mouth is the most common site. Head and neck cancers can also affect the throat, and there are rarer cancers of the nose, sinuses, salivary glands, skin and middle ear, or arising in other tissues (Figure 6).

What other malignant tumours affect the mouth or oropharynx?

One in 10 mouth and oropharyngeal cancers (10%) are one of the following types:

- Adenoid cystic cancer: a rare cancer that develops from glandular tissue mostly in salivary glands. The parotid gland is most commonly affected but only about 20% of parotid tumours are cancers.
- Lymphoma: cancers in lymph tissue at the base of the tongue and tonsils or the neck lymph nodes.
- Melanoma: develop from pigment-producing cells on the skin or inside the nose or mouth.

WHO International Classification of Diseases

	ICD–9	ICD–10
Lip	140	C00
Tongue	141	C01–02
Gum	143	C03
Floor of mouth	144	C04
Other and unspecified mouth	145	C05–06
Salivary gland	142	C07–08
Oro-, naso-, and hypo-pharynx Other and ill-defined sites of lip, oral cavity and pharynx	146–149	C09–14

Table 1. ICD for mouth cancers.

■ Salivary gland cancer: most tumours are in the parotid glands.
■ Sarcomas: develop from the cells in muscles, cartilage, bone or blood vessels.
■ Sinus tumours: develop from the lining cells.

What are mouth cancers?

Cancers can develop on the lips, tongue, floor of the mouth, the buccal mucosae, the palate, the gingivae or oropharynx. Mouth cancers (Figure 7) are mostly squamous carcinomas and include:
■ Lip cancers: most are seen on the lower lip.
■ Intra-oral cancers: most develop on the lateral tongue and floor of the mouth.
■ Oropharyngeal cancers: most affect the tonsils and base of the tongue.

There are also malignant tumours that arise from other structures including the salivary glands and paranasal air sinuses, or other tissues, but these cancers do not share a similar natural history and are not included here.

What are the types of mouth cancer?

Cancers can be described according to the cell type in which the cancer began. In the oral mucosa the most common arise from keratinocytes and are squamous cell carcinomas (SCC). About 9 out of 10 mouth cancers are squamous and are termed OSCC. About 1 in 20 mouth cancers (5%) are an unusual type of squamous cell carcinoma which rarely spreads to other parts of the body but can grow deeply into local tissues.

OSCC cancers are more common:
■ As with most cancers, in older people and in men;
■ In people who have already had mouth or oropharyngeal cancer or potentially malignant oral disorders;
■ In people who have had some other types of cancer such as:
– cancer of the oesophagus;
– squamous cell skin cancer;
– cervical, anal or genital cancer in women;
– cancer of the rectum in men.

Oral pre-cancerous (potentially malignant) disorders include especially (Article 6):
■ Erythroplakia: over 70% can become cancerous;
■ Leukoplakia: from 3–35% can develop cancerous changes;
■ Submucous fibrosis: from 1–10% can develop cancerous changes;
■ Lichen planus; from 1- 3% can develop cancerous changes;
■ Actinic cheilitis: from 10–30% can develop cancerous changes.

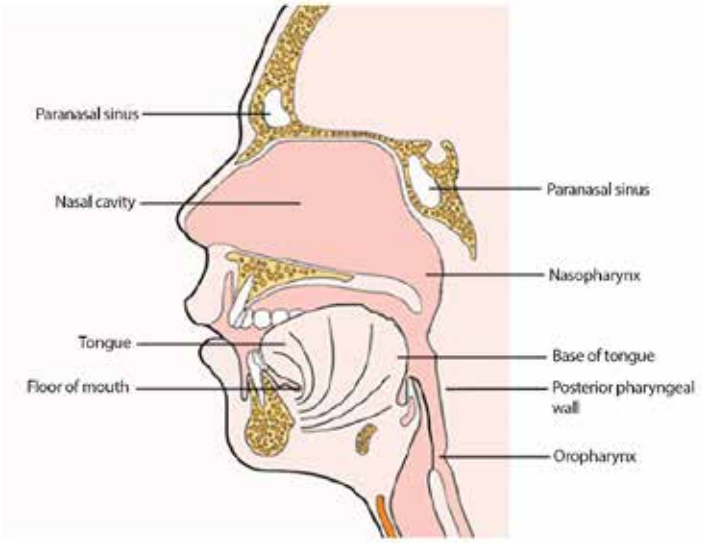

Figure 6. Head and neck cancer sites. (Courtesy of MacMillan Cancer Support.)

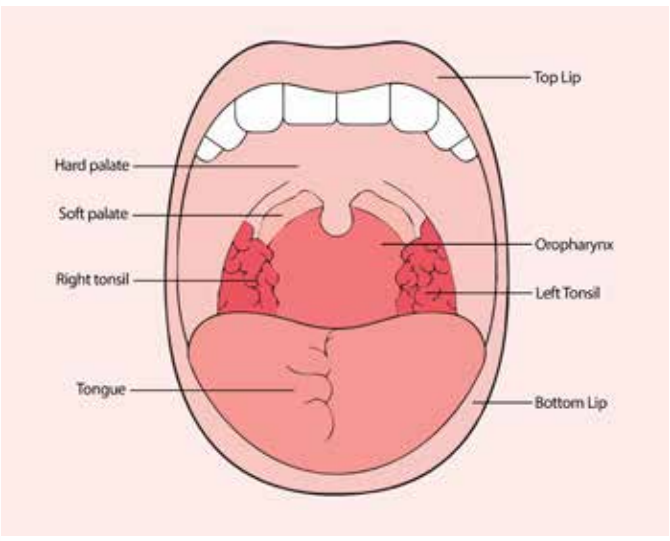

Figure 7. Oral sites. (Courtesy of MacMillan Cancer Support.)

What is the international classification of mouth tumours?

Of the many malignant neoplasms that can affect the mouth, oral squamous cell carcinoma (OSCC) is the most important.

Cancers of the 'oral cavity and oropharynx', as classified in the WHO International Classification of Diseases (ICD), include cancers of the lip, tongue and mouth (oral cavity) [ICD-10: C00-06], and oropharynx [ICD-10: C09-C10], but excludes the salivary glands [C07-08] and other pharyngeal sites [C11-13] (Table 1). ICD-9 is shown for comparison.

What is oropharyngeal cancer (cancer of the oropharynx)?

Oropharyngeal cancers are most commonly SCCs. Oropharyngeal cancer is uncommon: about 1,340 people are diagnosed each year in the UK. Like oral cancer, most oropharyngeal cancer affects mainly people over 50 and is more common in men than in women. However, oropharyngeal cancers are becoming more prevalent, which may be related to an increase in oropharyngeal viral infections (Article 4), notably HPV.

Conclusion

This review gives the dental team a simplified overview of carcinogenesis, and a summary of cancers that affect the oral region, as this is the single most important aspect of oral healthcare.

Further reading

1. **http://oralcancerldv.org/en/home/**
2. **https://entuk.org/sites/default/files/files/head_and_neck_cancer.pdf**
3. Blitzer GC, Smith MA, Harris SL, Kimple RJ. Review of the clinical and biologic aspects of human papillomavirus-positive squamous cell carcinomas of the head and neck. *Int J Radiat Oncol Biol Phys* 2014 Mar 15; **88**(4): 761–770. doi: 10.1016/j.ijrobp.2013.08.029. Review. PubMed PMID: 24606845; PubMed Central PMCID: PMC3990872.
4. Gonzales-Moles MA, Scully C, Ruiz-Avila I, Plaza-Campillo JJ. The cancer stem cell hypothesis applied to oral carcinoma. *J Oral Oncol* 2013 May 1; **49:** 738–746. pii: S1368-8375(13)00552-6. doi: 10.1016/j.oraloncology.2013.04.002. [Epub ahead of print].
5. Gonzalez-Moles M, Scully C, Ruiz-Avila I. Molecular findings in oral premalignant fields: update on their diagnostic and clinical implications. *Oral Dis* 2012; **18**: 40–47.
6. González-Moles MA, Ruiz-Ávila I, Gil-Montoya JA, Plaza-Campillo J, Scully C. β-Catenin in oral cancer: an update on current knowledge. *Oral Oncol* 2014 Jul 3; **50**: 88–825. pii: S1368-8375(14)00164-X. doi: 10.1016/j.oraloncology.2014.06.005. [Epub ahead of print] PubMed PMID: 4998198.
7. Hunter KD, Parkinson EK, Harrison PR. Profiling early head and neck cancer. *Nature Rev* 2005; **5**: 127–35.
8. Hunter KD, Thurlow JK, Fleming J, Drake PJ, Vass JK, Kalna G, Higham DJ, Herzyk P, Macdonald DG, Parkinson EK, Harrison PR. Divergent routes to oral cancer. *Cancer Res* 2006; **66**: 7405–7413.
9. Lechner M, Fenton T, West J, Wilson G, Feber A, Henderson S, Thirlwell C, Dibra HK, Jay A, Butcher L, Chakravarthy AR, Gratrix F, Patel N, Vaz F, O'Flynn P, Kalavrezos N, Teschendorff AE, Boshoff C, Beck S. Identification and functional validation of HPV-mediated hypermethylation in head and neck squamous cell carcinoma. *Genome Med* 2013 Feb 5; **5**(2):15. doi: 10.1186/gm419. eCollection2013. PubMed PMID: 23419152; PubMed Central PMCID: PMC3706778.
10. Lechner M, Frampton GM, Fenton T, Feber A, Palmer G, Jay A, Pillay N, Forster M, Cronin MT, Lipson D, Miller VA, Brennan TA, Henderson S, Vaz F, O'Flynn P, Kalavrezos N, Yelensky R, Beck S, Stephens PJ, Boshoff C. Targeted next-generation sequencing of head and neck squamous cell carcinoma identifies novel genetic alterations in HPV+ and HPV- tumors. *Genome Med* 2013 May 29; **5**(5): 49. doi: 10.1186/gm453. eCollection 2013. PubMed PMID: 23718828; PubMed Central PMCID: PMC4064312.
11. Leemans CR, Braakhuis BJ, Brakenhoff RH. The molecular biology of head and neck cancer. *Nat Rev Cancer* 2011; **11**(1): 9–22.
12. Lucchese A, Serpico R, Guida A, Crincoli V, Scully C, Kanduc D. Interkeratin peptide-protein interactions that promote HPV16 E7 gene expression. *Int J Immunopathol Pharmacol* 2010; **23**: 857–864.
13. Rizzo G, Black M, Mymryk JS, Barrett JW, Nichols AC. Defining the genomic landscape of head and neck cancers through next-generation sequencing. *Oral Dis* 2015; **21**: e11–e24.
14. Scully C, Warnakulasuriya S. Cancer of the mouth for the dental team. Comprehending the condition, causes, controversies, control and consequences: 1. General principles. *Dent Update* 2010; **37**; 638–640.
15. Scully C. Challenges in predicting which oral mucosal potentially malignant disease will progress to neoplasia. *Oral Dis* 2014; **20**: 1–5.
16. The Cancer Genome Atlas Network. Comprehensive genomic characterization of head and neck squamous cell carcinomas. *Nature* 2015; **517**: 576–582.
17. Tomasetti C, Vogelstein B. Cancer etiology. Variation in cancer risk among tissues can be explained by the number of stem cell divisions. *Science* 2015 Jan 2; **347**(6217): 78–81. doi: 10.1126/science.1260825. PubMed PMID: 25554788.
18. Werning J. *Oral Cancer: Diagnosis, Management, and Rehabilitation*. Stuttgart, Germany: Thieme Publishing, 2007.
19. Zahran F, Ghalwash D, Shaker O, Al-Johani K, Scully C. Salivary micro-RNAs in oral cancer: a case control study. *Oral Dis* 2015 Mar 17. Epub 2015 Mar 17.