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Oral Cancer Chemoprevention - A Review

Abstract

Oral cancer is a serious and growing problem in many parts of the globe. Oral and pharyngeal cancers, grouped together, are the sixth most common cancer in the world. Cancer chemoprevention is defined as the use of natural, synthetic, or biologic chemical agents to reverse, suppress, or prevent carcinogenic progression to invasive cancer. The success of several recent clinical trials in preventing cancer in high-risk populations suggests that chemoprevention is a rational and appealing strategy. This review will highlight current clinical research in chemoprevention, the biologic effects of chemopreventive agents on epithelial carcinogenesis, and the usefulness of intermediate biomarkers as markers of premalignancy.

Key Words

Oral Cancer; chemoprevention; carcinogenesis

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INTRODUCTION

The world is heading towards various types of non-communicable diseases, also known as "modern epidemics". Among these modern epidemics cancer is the second commonest cause of mortality in developed countries. Oral cancer is a serious and growing problem in many parts of the globe. Oral and pharyngeal cancers, grouped together, are the sixth most common cancer in the world. The predominant type of cancer found in the oral cavity is squamous cell carcinoma. Oral carcinogenesis is a multifactorial and complex process related to the sequential occurrence of alterations in genetic structures, promoting inhibitory or excitatory effects of the tumor oncogenes and gene suppressors, compromising the histophysiology of the division, differentiation and cell death. Biomarkers help in the evaluation of prevention or use of therapies and the detection of the earliest stages of oral mucosal malignant transformation. Biomarkers reveal the genetic and molecular changes related to early, intermediate and late endpoints in the process of oral carcinogenesis. Genetic and molecular biomarkers will also determine the effectiveness and safety of chemopreventives. Biomarkers will also reduce the number of patients and the time for long-term follow-up required to define a significant clinical response to a chemopreventive agent, thereby, clarifying the types, doses, frequencies and regimens to achieve the maximum level of benefit from chemopreventives.[1]

Role of Cellular Biomarkers

Indicators of deoxyribonucleic acid (DNA) repair mechanisms. Indicators of programmed cell death (PCD). Indicators of tumor development and growth. Indicators of genetic markers of Oral Cancer.[2]

Indicators of DNA Repair mechanisms

PCD functions not by itself, but in concert with systems that facilitates DNA repair. Present evidence indicates that cancer cells require a high level of DNA repair. In general, these include repair of the telomeric ends of chromosomes produced through the action of telomerase and repair of nucleotide sequences, exemplified by mismatch repair and nucleotide excision repair (NER). In human oral carcinomas, telomerase is elevated in the proliferative areas of the carcinoma. Defective repair processes and checkpoints are also linked to cancer genomic instability. Examples of the more

common DNA repair sites are approximately 20 genes known to be involved in the process of NER or the repair/transcription factors, such as Transcription factor II Human (TFIIH), that are required to orchestrate the function of incisional proteins, that is, DNA polymerases and ligases. These changes may become additional markers for the aggressive and metastatic characteristics of oral carcinoma. DNA repair influences the progression of oral carcinogenesis through the regulation of various growth factors, for example, transforming growth factor (TGF)-β3.[2]

Indicators of PCD

There is a considerable body of work that has identified several hundred cellular changes or biomarkers associated with the growth of oral carcinoma and carcinomas of the aero digestive tract. Many of these indicators are also markers for PCD. PCD or gene-directed apoptosis is characterized by the lack of an inflammation-driven necrosis of the tissue and the histologic appearance of apoptotic cells. PCD is also an important feature of oral keratinocytes undergoing differentiation or transformation during oral cancer development. The surviving transforming cells appear to have suppressed PCD and they have a high rate of proliferation, enhanced levels of resistance to different antitumor therapies and elevated levels of DNA repair.[3]

Oxidation and the effect on PCD

Oral malignant transformation could be a product of oxidative state change. The oxidative state change would then result in changes in DNA repair and PCD. The observed cellular manifestations of these processes are losses of cell growth control and modifications in cell-cell interactions, which could enhance the potential for tumor metastases. Nutrients that act as chemo preventives alter the oxidative state of the oral transforming cell by acting as reducing agents (e.g., antioxidant) and/or oxidizing agents (e.g. pro-oxidant).[4]

Chemopreventives and PCD

The treatment with chemopreventives, during malignant transformation or in fully transformed malignant oral mucosal cells, results in the induction of PCD and the observed inhibition of oral carcinogenesis and malignant tumor growth. Chemopreventives induce PCD because of their oxygen-responsive characteristics, which trigger inducers such as the tumor suppressor gene p53, modifiers of PCD such as the bcl-2 family and immune-derived cytokines, for example, tumor necrosis factor (TNF). Chemopreventives, as exemplified by retinoids, carotenoids, tocopherols, bioflavonoids, Isothiocynates, indoles and polyphenols induce PCD.[5,6]

Indicators of tumor development and growth

Biomarkers establish the level of risk for individuals in a target group of patients and they may provide information concerning the etiology and the process of carcinogenesis. The primary goal for the use of early, intermediate and late biomarkers is to identify individuals at risk of developing malignancy and indicate their level of risk.[1,7]

Indicators of genetic markers of Oral Cancer

A developing solid clone of transforming cells found in an oral carcinoma arrives at the state of malignancy by proceeding through stages of transformation. The transformation rate is dependent on the location of the clone in the spherical tumor mass and the oxygen states of the cells.[8]

Chemopreventives

Chemoprevention is an appealing strategy and its success has been demonstrated in breast cancer and familial adenomatous polyposis. High-dose retinoid have been shown to be active against oral premalignant lesions and in prevention of second primary tumors in the head and neck. [9] The rationale for pharmacologic chemoprevention in patients at risk for the development of invasive cancer is based upon two factors:

Field cancerization: Patients with head and neck cancer have a predilection for cancer development throughout the oropharyngeal mucosa.

Multistep carcinogenesis: Squamous cell cancers of the head and neck result from a multistep process with defined intermediate stages; leading to fully transformed, invasive and metastatic cancer. [10] Thus, chemopreventives are chemicals of natural or synthetic origin, which, unlike other drugs, does not prevent disease but instead, chemopreventives reduce the incidence of diseases such as cancer before clinical symptoms occur.

Classification of Chemopreventive agents (Pharmacological and chemical structural classification):[11]

1. ***Antimutagens/Carcinogen Blocking Agents***
   Phase II metabolic enzyme inducers, N-acetyl L-cysteine Polyphenols Curcumin and dehydroepiendrosterone (DHEA).
2. ***Antiproliferatives***
   Retinoids/caretinoids: β-carotene, 13-cis-retinoic

acids, vitamin-AGlucose-6-phosphate dehydrogenase inhibitors Aspirin.

3. ***Antioxidants***
   Commonly tried chemopreventive agents in oral cancer Vitamin A and other retinoids, β-caroteneVitamin E Dietary agents, Other agents.

Overview

A concept common to chemoprevention is the ability of chemical agents to function as reducing agents, antioxidants or oxidizing agents (pro-oxidants). The chemical character of the chemopreventive will depend on the partial pressure of oxygen and the level of oxidative metabolites produced or derived in the cell. β-carotene, a carotenoid, acts as an antioxidant, but can also act as a pro-oxidant, depending on the oxygen state of the cell. β-carotene as well as ellagic acid (from garlic) is carcinogen-blocking agents that either suppress promotion or act as antioxidants, which are reducing agents. Other chemopreventive agents, such as dλ-alpha-tocopherol (vitamin E), are strong antioxidants that enhance the cellular detoxification system by increasing the levels of glutathione-S-transferases (GSTs; Phase II enzymes). Some chemopreventives may suppress the promotion of cancer and prevent the transformation of premalignant cells by altering differentiation. Another class of chemopreventives, the terpenes, may also inhibit oncogenic expression and cell proliferation, reducing dedifferentiation. Indomethacin, an anti-inflammatory drug, blocks prostaglandin synthesis and reduces tumor development, resulting in a normal differentiation pattern. Cells also protect themselves from reactive oxygen substances (ROSs) by activating antioxidant pathways and molecular systems that use enzymes. Examples are superoxide dismutase, which controls the level of the superoxide anion (e.g., O2−); whereas, catalase modifies the levels of hydroxyl radicals (e.g., OH−) and glutathione- S-transferase (GSTs) alters the level of the intracellular antioxidant glutathione. Less obvious cellular antioxidants are proteins such as Bcl-2, which is comprised of a family of proteins that modifies PCD. There are also several protein families that function as redox, antioxidant/pro-oxidant molecules that also regulate PCD (e.g., p53). Perhaps the most important common feature of chemopreventives is their ability to trigger PCD in transformed cells. The retinoids, carotenoids, tocopherols, isothiocyanates and polyphenols induce PCD in various cell types.[11]

Genetic, molecular and biochemical activities of chemopreventive Retinoid Chemopreventives

The retinoid molecule family consists of vitamin A and its derivatives. The primary source of retinoids in the diet is retinyl esters from animal tissues and retinol from the conversion of carotenoids (e.g., 3-carotene) derived from a vegetable source. Retinol tends to be the biologically active form, but retinoic acid (RA) may substitute for retinol for many functions and both can be found in the serum linked to proteins. The normal plasma level for retinol is approximately 2 mmol/L, while that of RA is 10-20 nmol/L, about 150-fold lower than retinol. In cells, retinol and RA bind to specific cytoplasmic binding proteins, which include cytoplasmic retinol binding proteins I and II (CRABP-I/II). The cellular binding proteins mediate transfer of retinol and RA from the cytoplasm to the nucleus. The nuclear RA receptors (RAR) are members of the steroid-thyroid superfamily of nuclear receptors. Retinoid modulation of gene expression is the result of the activation of one of four possible receptors. The RARs are found in many different isoforms, as shown by mRNA, which indicates that the isoforms arise from different promoters or by alternative RNA splicing in a region at the 5 end of the mRNA. Isoforms appear to be tissue-specific and their expression is stage-specific, suggesting distinctive roles for each isoform. The expression of mRNA for RAR-; -β, -γ and -X has been found in normal oral mucosa. The development of premalignant change and smoking or alcohol use do not appear to change the general distribution of these receptors in oral tissue. However, recent studies have indicated that with retinoid treatment there is an increase in the level of RAR-β in premalignant lesions. Lotan *et al*.,(1995)[12] presented a study where RAR-1 was depressed during premalignant change and retinoid treatment and remission of oral leukoplakia was coincident with an increase in RAR-β. It appears that RAR-β may be an important indicator for a retinoid response in premalignant or malignant oral mucosa. Recent studies have also indicated that the conversion of retinol to RA might be mediated by the RAR. It is well-known that head and neck squamous cell carcinoma (HNSCC) patients, particularly those who smoke tobacco, have low levels of serum retinoids. Their low intake of fruits and vegetables may play a role in the development of oral cancer, but the cellular state of the oral mucosa must also be considered. The concentration of any retinoid metabolite may be initially

dependent on the type, number and affinity of the RARs present on the cell. In addition, RARs appear to be involved in the differentiation of myeloid cells and keratinocytes. RA binding to its receptors has also been shown to induce apoptosis in the differentiating cell population.[13,14,15]

Genetic and molecular activities of carotenoid and tocopherol

The carotenoids and tocopherols produce genetic and molecular responses similar to those observed with the retinoids. The major cellular difference between the retinoid response and the carotenoids or tocopherols is lack of a complex receptor system described above for retinoids. Chemopreventives, as a group, respond to changes in oxygen partial pressure that influences their ability to act as oxygen free radical quenchers or reactive oxygen molecules.[16] In a well-oxygenated environment, the carotenoid, β-carotene, can inhibit the growth of oral cancer cells, because the carotenoid induces an oxidative stress in the tumor cell. At the identical partial pressure of oxygen, tocopherol tends to have an antioxidant activity and reduces the oxidative stress. Some of the more common membrane-related responses to β-carotene and/or tocopherols are the reductions in membrane-associated enzymes such as serine and threonine kinases, connexin 43 and a reduction in ras proteins. Other previously discussed proteins, such as proto-oncogenes, c-fos, c-myb, N-myc and the tumor suppressor p53, have been shown to be affected by treatment with these chemopreventives. Stress proteins, acting as cellular chaperonins, can complex to p53 and following treatment of oral carcinoma cells with β-carotene.In addition, growth factors and immune regulatory factors such as epidermal growth factor (EGF) and TGF-α have a reduced expression, while TGF-β1 and TNF-α expression are elevated during the inhibition of oral carcinogenesis. The EGF receptor's (EGFR's) affinity, number and protein expression were reduced following β-carotene treatment and suppression of oral cancer growth. Studies reveal the broad effects of β-carotene and other similar chemopreventives on many different cellular functions. Importantly, the carotenoids and tocopherols do not appear to produce the toxicity seen with retinoids, but their effectiveness at preventing or reversing premalignant change is unclear.[16,17,18]

Oxidative biochemistry of Chemopreventives

Chemopreventive agents can be defined by their suppression or blocking of mutagenic activity, initiation and/or promotion during oral carcinogenesis. Agents that alter the mutagenic process are generally enzymes that enhance the solubilization or degradation of mutagenic or carcinogenic agents. This process usually activates the intracellular antioxidant, glutathione and its associated system. Glutathione also acts to block the nucleophilic attack of the carcinogen on DNA. Chemopreventive agents, such as glutathione, may also function by enhancing the elimination of the genotoxic agent from the liver and other tissues. In these tissues, the hydroxylation of the cytochrome P450 system and mixed-function oxidases (MFO) eventually terminates with conjugates of reduced protein salts containing glucuronate and sulfates.[14,19-22] Chemopreventives have been shown to inhibit both initiation and promotion during oral carcinogenesis. The inhibition of initiation may occur by preventing the carcinogen from becoming fully active by enhancing DNA repair and/or the activation of tumor suppressor genes. Initiation and promotion may also be affected by the elimination of transformed malignant clones of cells.[23-25] Examples of chemopreventives that have exhibited anti-promotional activity include tamoxifen (an anti-estrogen), retinoids and carotenoids, alpha-tocopherol acid succinate and piroxicam. Leukotriene activity can also be regulated by reducing the activity of lipoxygenase. The inhibition of prostaglandins has been shown to be associated with the suppression of oral carcinogenesis.[18,26-29]

CONCLUSION

It is thought that patients with head and neck premalignant changes consist of a diverse population and should be treated differently depending on their molecular genotype. Patients with minimal genetic changes may be treated with single-agent retinoids or other agents. Those with more accumulated genetic changes will require combination of chemoprevention therapies. Lesions that have advanced genetic changes with mutant p53 may benefit from targeted p53 therapy and those lesions that express EGFR and COX-2 may require inhibitors of EFGR and COX-2. However, the challenge today is achieving long-lasting efficacy with retinoids and/or new agents and determining the optimal dose and duration of therapy while maintaining acceptable toxicities.

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