NARRATIVE REVIEW

Cancer stem cell markers in oral squamous cell carcinoma – Revisited

M. G. Madhura¹, Lipika Kansal², B. Veerendra Kumar¹

¹Department of Oral Pathology and Microbiology, DA Pandu Memorial RV Dental College and Hospital, Bengaluru, Karnataka, India, ²Department of Oral Pathology and Microbiology, DA Pandu Memorial RV Dental College and Hospital, Bengaluru, Karnataka, India

Introduction

Oral squamous cell carcinoma (OSCC) ranks among the top three types of cancer in Indian population. OSCC is the result of genetic and epigenetic instability with the resultant mutations affecting various cell populations. Carcinogenesis is extremely complex multifactorial phenomenon with the interplay of innumerable molecules.¹,² Treatment for cancer is painful, traumatic, and disfiguring. A continuous desire to improve the diagnostic acumen and designing of tailored treatment has improved our understanding of the pathogenesis of carcinogenesis. Cancerous tissue is heterogeneous and comprises various cell types; cancer stem cells (CSCs) constitute a small subpopulation in it.

CSCs have been characterized as the most critical of the cancer cell population as these CSCs are known for progression of the cancer, the development of metastasis, and failure of the treatment. Although developments in the field of research on CSC are nascent, much is explored on various proteins and enzymatic processes that would facilitate isolation of the CSCs. This in turn sheds light on our understanding of CSCs in cancer progression, and appropriate targeted therapy may be instituted.

CSCs are a small group of cells having the potential of self-sustainability and ability to result in heterogeneous lineages.

The term CSCs denote functional properties of the cells rather than their origin.³,⁴ The study of CSCs in different neoplasms has shown that CSCs are tissue specific and it is difficult to have a universal CSC marker.

The CSCs could be derived either from the normal stem cells or from the mutated progenitor cells. Either enzymatic reactions or differential expression of specific molecular markers helps in identification of CSCs among other cell populations in cancerous tissue.

Various CSC markers studied in OSCC are CD44, CD133, Bmi-1, c-Met, ALDH, and so on. Isolation of the CSCs sheds light on our understanding of CSCs in cancer progression and institution of appropriate targeted therapy.⁵-25

The present paper gives a quick overview of various CSC markers and their role in head-and-neck cancer and OSCC.

CSC Biomarkers in Head-and-Neck Squamous Cell Carcinoma

CD44

CD44 is a cell-surface glycoprotein; it is involved in cell–cell interactions such as cell adhesion and migration. CD44
is a well-known marker for CSCs. CD44 is an indicator of tumor recurrence, metastasis, and mortality. CD44<sup>high</sup> CSCs are expressed in head-and-neck squamous cell carcinoma (HNSCC).

CD44 modulates cancer metastasis through facilitating easy adherence to blood vessels and transendothelial migration into blood vessels and secondly by promoting epithelial mesenchymal transition (EMT) of CSCs.

The interactions of CD44 with hyaluronic acid, heparan sulfate, and chondroitin sulfate bring about evasion of apoptosis, degradation of collagen, tumor invasion, and neovascularization.

On one side, studies have shown that CD44 is a proven biomarker in tumors of breast, colon, central nervous system, prostate, pancreas, and head and neck including OSCC. It is perceived that CD44 is a useful marker for assessing the progression of HNSCC and a possible target for therapy. On the other hand, scientific reports have questioned the validity of CD44 as a marker for tumor because of overexpression of CD44 in normal epithelium of head-and-neck region, as evidenced by tissue culture models (where CD44<sup>high</sup> and CD44<sup>low</sup> cells derived from squamospheres could regenerate the spheres from single-cell suspensions).<sup>[7-11]</sup>

Thus, under the light of current scientific evidence, CD44 as a biomarker in HNSCC/OSCC needs further validation.

**Aldehyde dehydrogenases (ALDH)**

Comprise of intracellular cytosolic isoenzymes found in the liver. ALDHs convert retinol to retinoic acid during early stem cell differentiation and catalyze the oxidation of toxic aldehyde metabolites. Expression of ALDH has been reported in malignancies of breast, lung, colon, and liver. In vitro tissue culture models have shown ALDH<sup>+</sup> CSCs in OSCC sphere formation, tumor formation and invasion, resistance to chemotherapeutic agents. Higher expression of ALDH in OSCC has been reported to correlate with disease staging, radio-resistance, and poorer patient outcome.<sup>[12,13]</sup>

**CD133**

CD133 also called as Prominin-1 is a CSC marker characterized in epithelial cells and in somatic stem cells from kidney, prostate, neural tissues, colorectal, liver, lung, and skin. CD133<sup>+</sup> cells in HNSCC and OSCC have shown higher clonogenicity, self-renewal, proliferation, tumorigenicity, tumorsphere formation, and EMT phenotype. Higher expression of CD133 has shown positive correlation with Oct-4, Nanog (transcription factors/stemness markers) in oral cancer patients.

CD133 may be regarded as a suitable CSC marker in OSCC, to identify patient’s resistance to chemotherapy.<sup>[14,15]</sup>

**c-Met**

c-Met (mesenchymal-epithelial transition factor) being a proto-oncogene encodes for hepatocyte growth factor tyrosine kinase receptor. c-Met expression is shown to be associated with tumor angiogenesis, tumor invasion, metastasis, and decreased patient survival. c-Met positive cells in HNSCC have demonstrated self-renewal and more tumorigenic potential than CD44.

Higher expression of c-Met has been reported in OSCC when compared to that of normal oral mucosa. Cell membrane and cytoplasm were the sites of c-Met localization. Evidence from OSCC tissues and cell lines have unveiled that c-Met could be functionally important in human OSCC progression.<sup>[16,17]</sup>

**Stemness Markers**

**Bmi-1**

Bmi-1 (B-cell-specific Moloney murine leukemia virus integration site 1) plays a significant role in cell cycle, development, senescence, stem cell genesis, DNA damage response, and cancer. Altered expression of Bmi-1 has been reported in various types of human cancer. In HNSCC, Bmi-1 regulates epithelial-mesenchymal transition through Twist1. In tongue carcinoma, Bmi-1 is overexpressed and it is found to be associated with cervical node metastasis.<sup>[18-20]</sup>

**Oct-4, Nanog, and Sox2**

The transcription factors – Oct-4, Sox2, and Nanog – interact with other transcription factors (STAT3, Zic3, and HesX1) and cell-signaling molecules (LEFTY2, FGF2, and TCF3), thereby maintaining pluripotency and self-renewal of embryonic stem cells. Tissue culture models in HNSCC have shown positive correlation of overexpression of Oct-4 and Nanog genes with disease stage and treatment failure and negative correlation with differentiation status. Thus, cells with stem-like features in cancer do express Oct-4, Sox-2, and Nanog, the transcription factors.<sup>[21,22]</sup>

**Kruppel-like factor 4 (Klf4)**

Klf-4 is a zinc finger transcription factor which is observed in the upstream of Akt in oral potentially malignant disorders. Klf-4 has been implicated in reprogramming of somatic cells to acquire stem cell-like state, thus maintaining the self-renewal capacity of cells, also in regulating cell growth and differentiation. Increased expression of klf-4 has been reported in human HNSCC. Reports on klf-4 expression in various types of cancer have revealed context-dependent function; Klf4 may function as an oncogene or a tumor suppressor gene. In head-and-neck cancer, klf4 as a stem cell factor promotes epithelial-mesenchymal transition through TWIST1-JAGGED1-KLF4 in angiogenesis.<sup>[23]</sup>

**Lgr5**

Lgr5 (also called as GPR49, G-protein coupled receptor 49) being a transmembrane receptor protein is a marker for adult stem cells in intestinal epithelium and hair follicle; Lgr5 is responsible for erroneous activation of Wnt signaling pathway resulting in cytoplasmic accumulation of β-catenin that in turn is associated with carcinogenesis.<sup>[24]</sup> Lgr5 is responsible for tumor initiation, progression, metastasis, and recurrence of the tumor. In OSCC, expression of Lgr5 and vasculogenic mimicry have
shown to be potential biomarkers for metastasis and prognosis and also may act as therapeutic targets.[26]

**Conclusion**

CSCs have the ability for self-renewal and proliferation. CSCs confer heterogeneity to tumor mass and are overexpressed in tissues, thereby contributing to tumorigenesis, metastasis, recurrence, and chemoresistance. Various biomarkers have been explored which are considered to be CSCs. Increased density of CSCs, as identified by overexpression of various primitive cells and CSC markers have been associated with poor prognosis of head-and-neck cancer. CSCs may act as therapeutic targets for the tailored treatment of HNSCC/OSCC.

**References**