DNA Methylation as a Diagnostic, Prognostic, and Predictive Biomarker in Head and Neck Cancer

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ABSTRACT

Head and neck squamous cell carcinoma (HNSCC) is a term collectively used to describe all cancers that develop in the oral and nasal cavities, the paranasal sinuses, the salivary glands, the pharynx, and the larynx. The majority (75%) of all newly diagnosed cases are observed in patients with locally advanced and aggressive disease, associated with significant relapse rates (30%) and poor prognostic outcomes, despite advances in multimodal treatment. Consequently, there is an unmet need for the identification and application of tools that would enable diagnosis at the earliest possible stage, accurately predict prognostic outcomes, contribute to the timely detection of relapses, and aid in the decision for therapy selection. Recent evidence suggests that DNA methylation can alter the expression of genes in a way that it favors tumorigenesis and tumor progression in HNSCC, and therefore represents a potential source for biomarker identification. This study summarizes the current knowledge on how abnormally methylated DNA profiles in HNSCC patients may contribute to the pathogenesis of HNSCC and designate the methylation patterns that have the potential to constitute clinically valuable biomarkers for achieving significant advances in the management of the disease and for improving survival outcomes in these patients.

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INTRODUCTION

Head and neck squamous cell carcinoma (HNSCC) is a general term that includes all cancers that develop in the oral and nasal cavities, the paranasal sinuses, the salivary glands, the pharynx, and the larynx. Patients with locally advanced and aggressive disease, who account for 75% of the newly diagnosed cases, are likely to experience relapse and have a 5-year overall survival (OS) rate of 50%, despite advances in surgical treatment, radiotherapy, chemotherapy, and modern treatments. Consequently, there is a need for the identification and application of tools with high sensitivity and specificity that would enable diagnosis at the earliest possible stage, inform clinicians of the possible prognosis of patients, contribute to the early detection of relapses, and provide information on the progression of the disease after the application of specific treatments. One of the mechanisms by which smoking, alcohol abuse, and HPV infection increase the risk of developing HNSCC is through the induction of epigenetic changes that lead to abnormal cellular physiology. Specifically, these are alterations at the chromosomal level that lead to changes in gene expression without altering the DNA sequence. The most common of these alterations are DNA methylation, non-coding RNAs, and histone modifications. DNA methylation involves the addition of a methyl group from S-adenosyl methionine (SAM) to the fifth carbon of cytosine (C) to form 5-methylcytosine (5 mC). This study summarizes the current knowledge on how abnormally methylated DNA profiles in HNSCC patients may contribute to the pathogenesis of HNSCC and designate the methylation patterns that have the potential to constitute clinically valuable biomarkers for achieving significant advances in managing the disease and for improving survival outcomes in these patients.

RESULTS

Regarding diagnostic biomarkers, a total of thirteen studies were analyzed. Concerning the type of samples studied, three studies included saliva samples, twelve included tissue samples, and one included plasma samples. In addition, ten studies focused on HNSCC in general, while one study focused mainly on oral and oropharyngeal cancer, one concentrated on oral cancer, and one on HPV+ oropharyngeal carcinoma. Subsequently, in six studies, the pathological samples showed hypermethylation of the respective gene promoters, with subsequent reduction of transcriptional expression, five studies showed promoter hypomethylation with subsequent increase in transcript levels, and two studies showed a differential methylation pattern as compared to normal samples. Two studies investigated the methylation status of HOX9, a transcription factor involved in nuclear maintenance, cell proliferation, cell differentiation, and apoptosis, yet the first of these studies referred to promoter hypermethylation, whereas the second study referred to promoter hypomethylation. The other genes found to be differentially methylated include: miR-9, EDNRB, DCC, MED15/PCQAP, ZNF14, ZNF160 and ZNF420, FAM135B, ZNF610, CTHRC, PLAU, MTHFD1L, OPRM1, OPRL1, XPR1, KCNA3, EMBP1, CCDC181, DPP4, ITGA4, BEND4, CTNND2, ELMO1, SFMBT2, C1QL3, MIR129-2, ATP5EP2, OR6S1, NID2, HOXB4, ZNF439, ZNF93, VSTM2B, ZNF137P, and ZNF773. In addition, a gene signature of five methylated genes, namely GATA4, GRIA4, IRX4, ALDH1A2, and OSR2, was identified.

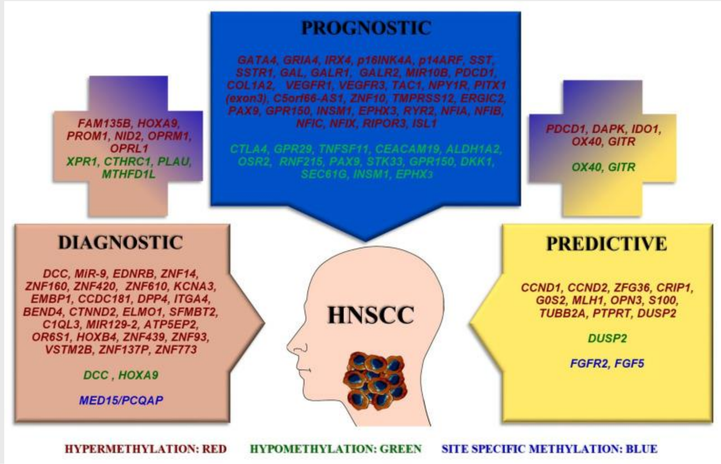

Figure 1:
Differentially methylated genes as biomarkers with diagnostic, prognostic, and predictive value in HNSCC. The genes included in the plus signs have dual roles, i.e., either both diagnostic and prognostic (left) or both prognostic and predictive (right).

RESULTS-CONT.

As for the prognostic biomarkers, twenty-nine studies were reviewed. The samples were derived from tissues, while two studies included additional blood plasma samples and one also involved cell lines. Moreover, twenty-two studies focused on HNSCC, one study focused mainly on oral and oropharyngeal cancer, two on tongue cancer, two on oral cancer, one on HPV+ oropharyngeal carcinoma, one on HPV− cancer of the hypopharynx, larynx and mouth, and one on HPV− HNSCC. Furthermore, hypermethylation was associated with an unfavorable prognosis in seventeen studies, hypermethylation was linked to a favorable prognosis in three studies, hypomethylation was associated with an unfavorable prognosis in nine studies, and hypomethylation was associated with a favorable prognosis in three studies. In total, the following genes were analyzed: methylation signature 1 (GATA4, GRIA4, IRX4, ALDH1A2, OSR2), FAM135B, HOXA9, PROM1/CD133, CTHRC1, PLAU, MTHFD1L, OPRM1, OPRL1, XPR1, p16INK4A, p14ARF, SST, SSTR1, GAL, GALR1/2, MIR10B, methylation signature 2 (FUT3, TRIM5, TSPAN7, MAP3K8, RPS6KA2, SLC9A9, NPAS3, TIMM8A, RNF113A), PDCD1, COL1A2, VEGFR1, COL1A2, DAPK, TAC1, GALR1, NPY1R, SSTR1, VEGFR3, PITX1, C5orf66-AS1 lincRNA gene, methylation signature 3 (ZNF10, TMPRSS12, ERGIC2, RNF215), IDO1, methylation signature 4 (cg17892178 and cg17378966 in NID2 and IDO1), five DMG mode (PAX9, STK33, GPR150, INSM1, EPHX3), RYR2, NFI, DKK1, CALML5, DNAJC5G, LY6D, SEC61G, OX40, GITR, RIPOR3, CTLA4, GPR29, TNFSF11, ISL1, nine-gene multi-omics signature (methylation status of CEACAM19, KRT17, and ST18).

Finally, eleven studies evaluated the predictive biomarkers. The samples analyzed were derived from tissues and cell lines while all the studies were related to HNSCC in general. The therapeutic agents that were tested were immunotherapeutic agents, targeted agents, and chemotherapeutic agents, as well as radiotherapy and chemoradiotherapy.

METHODS AND MATERIALS

A systematic search was performed in PubMed for articles published in English between 1 January 2012 and 20 October 2022. The search terms used were related to DNA methylations and head and neck squamous cell carcinoma.

DISCUSSION

Therefore, during the past decade, a plethora of DNA methylation markers have been identified that show a high level of accuracy and reproducibility in a variety of biospecimens from HNSCC patients that have been obtained with non-invasive or semiinvasive techniques. Epigenetic changes in DNA methylation appear to play an important role in the entire spectrum of HNSCC evolution, from tumor initiation to aggressive disease progression. At the same time, however, there is a need for further identification and stratification of methylation biomarkers, and for the development of robust and efficient detection methods that can be applied to a routine clinical setting of risk assessment, diagnosis (especially at the early stage of the disease), prognosis, treatment management with various therapeutic agents, and post-treatment monitoring.

Currently, in vitro methylation-based biomarker diagnostic tests are commercially available for colorectal cancer, glioblastoma, hepatocellular carcinoma, lung and bladder cancer, as well as cervical and prostate cancer. However, there is no equivalent commercially approved test exclusively for head and neck cancer. The pathfinder II clinical trial is currently in progress and is focused on evaluating a cell-free DNA-based targeted methylation multi-cancer early detection (MCED) blood testing method that includes head and neck cancer.

CONCLUSIONS

In conclusion, there is a large chasm between the identification of promising epigenetic signatures and their transfer to clinical practice, even though the number of described epigenetic signatures is rapidly increasing. The adaptation of new methods and technologies can contribute towards the selection of the most valuable epigenetic biomarkers in clinical practice and achieve the transition to precision and personalized medicine.

INDICATIVE REFERENCES

1. Arantes, L.M.R.B.; De Carvalho, A.C.; Melendez, M.E.; Lopes Carvalho, A. Serum, Plasma and Saliva Biomarkers for Head and Neck Cancer. Expert Rev. Mol. Diagn. 2018, 18, 85–112. [CrossRef] [PubMed]
2. Miranda-Galvis, M.; Loveless, R.; Kowalski, L.P.; Teng, Y. Impacts of Environmental Factors on Head and Neck Cancer Pathogenesis and Progression. Cells 2021, 10, 389. [CrossRef] [PubMed]
3. Leemans, C.R.; Snijders, P.J.F.; Brakenhoff, R.H. The Molecular Landscape of Head and Neck Cancer. Nat. Rev. Cancer 2018, 18,269–282. [CrossRef] [PubMed]
4. Mesia, R.; Iglesias, L.; Lambea, J.; Martínez-Trufero, J.; Soria, A.; Taberna, M.; Trigo, J.; Chaves, M.; García-Castaño, A.; Cruz, J. SEOM Clinical Guidelines for the Treatment of Head and Neck Cancer 2020. Clin. Transl. Oncol. 2021, 23, 913–921. [CrossRef]