Clinical Oncology xxx (2016) 1–8

Contents lists available at ScienceDirect

Clinical Oncology

journal homepage: www.clinicaloncologyonline.net

Overview

Biological Features of Human Papillomavirus-related Head and Neck Cancers Contributing to Improved Response

C. Cleary *, J.E. Leeman *, D.S. Higginson *, N. Katabi †, E. Sherman ‡, L. Morris §, S. McBride *, N. Lee *, N. Riaz *

* Department of Radiation Oncology, Memorial Sloan Kettering Cancer Center, New York, NY, USA
† Department of Pathology, Memorial Sloan Kettering Cancer Center, New York, NY, USA
‡ Department of Medical Oncology, Memorial Sloan Kettering Cancer Center, New York, NY, USA
§ Department of Surgery, Memorial Sloan Kettering Cancer Center, New York, NY, USA

Received 8 February 2016; received in revised form 29 February 2016; accepted 1 March 2016

Abstract

Head and neck squamous cell carcinomas (HNSCC) are the sixth most common malignancy globally, and an increasing proportion of oropharyngeal HNSCCs are associated with the human papillomavirus (HPV). Patients with HPV-associated tumours have markedly improved overall and disease-specific survival compared with their HPV-negative counterparts when treated with chemoradiation. Although the difference in outcomes between these two groups is clearly established, the mechanism underlying these differences remains an area of investigation. Data from preclinical, clinical and genomics studies have started to suggest that an increase in radio-sensitivity of HPV-positive HNSCC may be responsible for improved outcomes, the putative mechanisms of which we will review here. The Cancer Genome Atlas and others have recently documented a multitude of molecular differences between HPV-positive and HPV-negative tumours. Preclinical investigations by multiple groups have explored possible mechanisms of increased sensitivity to therapy, including examining differences in DNA repair, hypoxia and the immune response. In addition to differences in the response to therapy, some groups have started to investigate phenotypic differences between the two diseases, such as tumour invasiveness. Finally, we will conclude with a brief review of ongoing clinical trials that are attempting to de-escalate treatment to minimise long-term toxicity while maintaining cure rates. New insights from preclinical and genomic studies may eventually lead to personalised treatment paradigms for HPV-positive patients.

Key words: Biology; DNA repair; head and neck cancer; HPV; radiosensitivity

Statement of Search Strategies Used and Sources of Information

To find relevant literature articles, articles discussing the response of human papillomavirus (HPV)-positive head and neck squamous cell carcinoma (HNSCC) to treatment were found using PubMed and Google Scholar with key words such as head and neck cancer, HNSCC, HPV, radiation, P16, survival and sensitivity. Abstracts were skimmed for relevance and reviews on related topics were read for additional references that may have had data relevant to our discussion. Articles on subjects discussed in this review were then searched for using the same databases and combinations of the aforementioned key words together with DNA repair, DNA damage, genomics, invasiveness, E6, E7, immune response, hypoxia, apoptosis, Rb, p53 and CCND1.

Introduction

Head and neck squamous cell carcinomas (HNSCC) have long been recognised to arise as a result of environmental exposures including tobacco and alcohol. However, in the past 15 years, human papillomavirus (HPV) positivity has been recognised as an aetiological factor for a substantial and increasing subset of these tumours [1]. Since the discovery of a separate viral aetiology of HNSCC, multiple

Authors for correspondence: N. Lee and N. Riaz, Department of Radiation Oncology, Memorial Sloan Kettering Cancer Centre, New York, NY, USA.
E-mail addresses: leen2@mskcc.org (N. Lee), riazn@mskcc.org (N. Riaz).

http://dx.doi.org/10.1016/j.clon.2016.03.001
0936-6555/

Please cite this article in press as: Cleary C, et al., Biological Features of Human Papillomavirus-related Head and Neck Cancers Contributing to Improved Response, Clinical Oncology (2016), http://dx.doi.org/10.1016/j.clon.2016.03.001

groups have shown significantly improved outcomes after treatment for HPV-positive tumours when compared with HPV-negative tumours [2–4]. Most of these series have involved patients treated either with radiotherapy or chemoradiotherapy, suggesting that HPV-positive tumours may be particularly sensitive to ionising radiation. HPV positivity is also a favourable prognostic factor in anal cancers, which are also typically treated with definitive regimens of chemoradiation with generally favourable outcomes [5–11]. Cervix cancers are nearly 95–100% associated with HPV infection, but nevertheless some series show that HPV-negative or certain HPV subtypes are associated with worse outcome [10,12–14]. Taken together, HPV positivity across multiple disease sites results in a favourable outcome to chemoradiation, and strongly suggests a particular cellular sensitivity to DNA-damaging agents. Further *in vitro* characterisations of HPV-associated cell lines has suggested increased radiosensitivity [15,16].

Potential mechanistic explanations for improved outcomes will be the focus of this review. HPV-positive HNSCCs have some molecular similarities, but many differences compared with HPV-negative HNSCCs (Table 1). In some senses, HPV-positive HNSCCs can seem molecularly more similar to HPV-positive tumours in other parts of the body (anal and cervical cancer) than HPV-negative HNSCC [17–19]. We will first review the key molecular and genetic differences between HPV-positive and HPV-negative tumours that may underlie some of the differences in sensitivity. Second we will summarise research investigating the underlying mechanisms of improved radiation response with a special focus on three main areas of investigation: hypoxia, the immune response and the DNA damage response (DDR) (Figure 1). Historically, the efficacy of ionising radiation has been attributed to the four Rs of radiobiology: repair of DNA damage, re-oxygenation, redistribution in the cell cycle and repopulation. HPV oncogenesis affects each of these components: it hijacks the cellular machinery for DNA repair, HPV-positive tumours display unique kinetics of hypoxia during radiation treatment, HPV alters the cell cycle distribution of infected cells by regulating checkpoint mediators and it induces rapid cellular proliferation [20]. Third, we will review other possibilities for improved outcomes rather than increased sensitivity, such as phenotypic differences between HPV-positive and HPV-negative tumours. Finally, we will discuss current efforts to de-escalate treatment and minimise toxicity for patients. A more precise understanding of the mechanism of improved response will ultimately help guide rational de-escalation trial design.

Table 1
Prevalence of genetic alterations in notable genes between human papillomavirus (HPV)-negative and HPV-positive samples

	HPV-	HPV+
ATM	Common	Very Common
CCND1	Very Common	Rare
CDKN2A	Most	Rare
EGFR	Common	Rare
FGFR1	Common	Rare
NOTCH1	Very Common	Common
p53	Most	Rare
PIK3CA	Common	Very Common
Rb	Rare	Rare
TRAF3	Rare	Very Common

Rare defined as < 5%, Common 10–20%, Very Common 20–50%, Most > 50%.

Genomic and Molecular Differences between Human Papillomavirus-positive and -negative Tumours

The HPV genome contains two oncogenes, E6 and E7, which inhibit p53 and Rb, respectively, and are thought to be primarily responsible for viral oncogenesis. Although most HPV-negative HNSCCs have a *TP53* mutation, HPV-positive HNSCCs rarely do because E6 expression disables *TP53* instead [21,22]. Nearly half to three-quarters of HPV-negative HNSCCs have mutated *TP53* [21]. CDKN2A, the gene coding for the protein p16, is inactivated in an overwhelming portion of HPV-negative tumours. About 30% of HNSCC have homozygous deletions of CDKN2A; 10–20% have loss of function mutations and a similar fraction have inactivating epigenetic alterations [22]. Degradation of Rb by E7 drives overexpression of p16. Thus, high expression of p16 is a surrogate marker for HPV-driven oncogenesis and has also shown to be a prognostic factor for radiotherapy outcome [23,24]. Mutations in Rb itself are more likely to appear in HPV-positive tumours, although such occurrences are rare, probably due to a lack of selective pressure in the presence of E6 [22].

Presumably as a response to viral infection, HPV-associated tumours display evidence of induction of a component of innate immunity known as the apoliprotein B mRNA editing enzyme catalytic polypeptide-like (APOBEC) family of proteins. This is a class of editing enzymes that change cytosine to uracil by deamination, thereby hypermutating and inactivating viral DNA. The replacement of the uracil commonly results in a mutation to thymine or guanine. The mutation signature of these enzymes is frequently found in virally transformed cancers and it has been found to be especially active in HPV-positive HNSCC [25]. Potentially as a consequence of high APOBEC activity, two specific codons (E542K and E545K) within the helicase domain of PIK3CA are frequently mutated in HPV-positive HNSCC due to C>T transitions. In HPV-negative tumours, PIK3CA mutations are spread throughout the gene and are significantly less frequent (12% versus 33%) [25]. This APOBEC pathway may be a driving mechanism behind HPV-associated transformation, but whether activation of this pathway affects therapeutic sensitivity is unknown.

On a gene-expression basis, HNSCC has been divided into four subtypes – basal, mesenchymal, atypical and classical – as first described by Chung *et al.* in 2004 [26]. These subtypes were confirmed by Walter *et al.* in 2013 [27] and by TCGA in 2015 [17]. HPV-positive tumours tend to be

Please cite this article in press as: Cleary C, et al., Biological Features of Human Papillomavirus-related Head and Neck Cancers Contributing to Improved Response, Clinical Oncology (2016), http://dx.doi.org/10.1016/j.clon.2016.03.001