PTCH1 Mutation in a Patient With Metastatic Undifferentiated Carcinoma With Clear Cell Change

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ABSTRACT

Clear cell basal cell carcinoma (BCC) is an unusual variant of BCC. Its pathogenesis, prognosis, and optimal management remain poorly described due to its rarity. This report presents a 51-year-old man with a history of excised BCC and cutaneous squamous cell carcinomas of the face, with multiple recurrent poorly differentiated carcinomas with clear cell changes of the shoulder for which further classification using conventional histologic means was not possible. His tumor tissue was sent to Foundation Medicine for testing, which revealed a high number of pathogenic genomic alterations, including a mutation in *PTCH1*. He was diagnosed with dedifferentiated BCC and started on vismodegib. He developed lung metastases while receiving vismodegib, and his disease continued to progress while he was undergoing treatment in a phase I clinical trial. Given the high number of pathogenic alterations suggestive of high tumor mutational burden, immunotherapy was considered and off-label authorization was obtained for treatment with a PD-1 antibody (pembrolizumab). He had a dramatic disease response after 4 infusions of pembrolizumab. Molecular testing was instrumental in determining the correct diagnosis and formulating appropriate treatment options for this patient. Molecular profiling of metastatic BCCs and its subtypes is essential to the development of effective targeted therapies and combination approaches.

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Cutaneous neoplasms have a broad spectrum of morphologic presentations. Neoplasms with clear cell morphology, defined as cells with optically clear cytoplasm caused by lipid, glycogen, or mucin accumulation, have been well described and are comprised of primary or metastatic lesions with epidermal, adnexal, mesenchymal, or melanocytic differentiation.^{1,2} Because clear cell morphology is easily identified but nonspecific, ancillary studies such as immunohistochemical staining or molecular studies are helpful in establishing the histogenesis of these tumors.

Clear cell change has been described in epidermal tumors, including acanthoma, seborrheic keratosis, squamous cell carcinoma (SCC), and basal cell carcinoma (BCC). BCC is the most common cancer in the United States, with >2 million cases occurring annually.³ Although known for its ability to cause significant local destruction of soft tissue, cartilage, and bone, it often has a good prognosis because of its low metastatic rate of <0.1%.^{4–6} The pathogenesis of BCC has also been well characterized, with the sonic hedgehog (Shh) signaling pathway identified as playing a crucial role. Signaling in this pathway is initiated by the cell surface receptor smoothed homolog (SMO). This pathway is normally inhibited by the cell surface receptor PTCH1, and binding of the hedgehog ligand to PTCH1 prevents this inhibition. The *PTCH1* gene on chromosome 9q codes for the Shh receptor, and mutations in this gene result in loss of inhibition of SMO activation of the hedgehog pathway and are drivers of nevoid BCC syndrome and 30% to 90% of sporadically occurring BCCs.^{7–9} Ultraviolet-induced mutations in the tumor suppressor gene *TP53* have also been implicated in BCC development.¹⁰

In contrast, clear cell BCC is rare. It was first described in 1984,¹¹ and to date only approximately 30 cases have been described in the literature. These cases, however, have focused on histopathologic/immunohistochemical characteristics of resected primary clear cell BCCs. One

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See page 886 for related commentary.

case report describes clear cell BCC with oligometastatic disease,¹² but molecular profiling was not performed and disease outcome is unknown; recurrence occurred after surgery, but no description of subsequent systemic therapy was provided. The molecular profile and clinical behavior of clear cell BCC thus remains poorly described.

Accordingly, we report a case of undifferentiated carcinoma with clear cell change. Identification of a *PTCH1* mutation through molecular profiling was instrumental in correctly diagnosing clear cell BCC and in identifying rational treatment options for metastatic disease.

Case Report

A 51-year-old man presented to the UCSF Melanoma and Skin Cancer Program in June 2015 with a rapidly enlarging mass of the right trapezius. He described a 10 pack-year smoking history and previous BCC and SCC excision from the left postauricular region in 2005. His recent history was also notable for growth of a right upper back tumor in February 2015, which was resected in late April 2015, with pathology indicating poorly differentiated clear cell carcinoma (Figure 1). The tumor was 9.8 cm in greatest diameter and was excised with a positive margin. Focal perineural invasion and vaguely squamoid areas were also noted. No adjuvant therapy was administered. On clinical examination at his June 2015 visit, the patient had soft tissue swelling over his right trapezius with ipsilateral supraclavicular lymphadenopathy. PET/CT performed on July 10, 2015, confirmed the presence of a 4.8-cm mass (maximum standardized uptake value [SUV_{max}], 17.7) in the right supraclavicular region extending to the posterior neck, with an ipsilateral trapezius satellite nodule (SUV_{max} , 8.2). A hiatal hernia was noted, but no distant metastatic disease was seen.

Review of the patient's external records revealed an extensive oncologic history. After the BCC and SCC excisions from his left postauricular region in 2005, he developed a nonhealing ulcer of the left mandibular angle in 2009 that was diagnosed as BCC and for which he underwent 3 weeks of radiation followed by surgery with wide excision, left superficial parotidectomy, and left modified neck dissection with a left pectoralis rotation flap.

In 2013, the patient had 2 recurrences at the left neck. In February 2013, biopsy of a left neck lesion showed BCC, for which he underwent wide excision with negative margins and a split-thickness skin graft. In July 2013, a new lesion appeared within the graft. Biopsy again showed BCC, and he underwent surgical excision, with an intraoperative finding of a deeper separate tumor. Final pathology noted a poorly differentiated clear cell carcinoma. After undergoing postoperative radiation to the left neck, he did well until

Figure 1. Dermatopathology images showing (A) low-power overview of clear cell carcinoma extending to the subcutis (hematoxylin and eosin, original magnification $\times 4$); (B) high-power view of clear cell carcinoma (hematoxylin and eosin, original magnification $\times 10$); and (C) high-power view of a BerEP4 immunohistochemical stain showing BerEP4 expression by neoplastic keratinocytes (original magnification $\times 10$).

February 2015, when he noticed the right upper back mass for which he underwent surgery in April 2015.

Surgery was not immediately recommended after results of the July 2015 PET/CT scan noted the masses in the right trapezius and supraclavicular region. This case was presented at a multidisciplinary tumor board. Although the disease was clearly a metastatic poorly differentiated carcinoma with clear cell change, further classification was needed to formulate an appropriate treatment plan. The differential diagnosis included BCC, SCC, or clear cell carcinoma of adnexal origin, all of which would have been treated differently from a clinical standpoint. Further classification through conventional pathologic means was not believed possible, and thus the tumor board recommended tumor molecular profiling to help with diagnosis and identification of possible therapeutic options. A formalin-fixed, paraffin-embedded tumor sample was

submitted to Foundation Medicine for comprehensive genomic profiling. FoundationOne is a next-generation sequencing–based assay developed and validated to identify genomic alterations within hundreds of cancer-related genes.¹³ At the time of this patient's tissue submission, the assay was noted to interrogate 315 genes and introns of 28 genes involved in rearrangements.

On August 11, 2015, the FoundationOne results noted many pathogenic genomic alterations (Table 1). Of particular interest were 2 disabling mutations in *PTCH1*: one frameshift and the other truncating. Also noted was an inactivating mutation in *MLH1*, an alteration that results in defective DNA mismatch repair (dMMR) and can result in microsatellite instability (MSI) in a variety of solid tumors. Given the known role of *PTCH1* gene mutation and the Shh signaling pathway in BCC pathogenesis, the patient was diagnosed with clear cell BCC and was started on vismodegib on August 28, 2015. Although he tolerated treatment well, he experienced increasing discomfort and limited range of motion in his right upper extremity. The right trapezius mass continued to enlarge, and subsequent PET/CT performed on October 28, 2015, confirmed progressive disease characterized by a larger and more avid right trapezius mass (2.2 cm; SUV_{max}, 22.3), new right axillary lymphadenopathy (1.6 cm; SUV_{max}, 13.8), and new pulmonary nodules in the right upper lobe (1 cm; SUV_{max}, 6.2) and lingula (0.8 cm; SUV_{max}, 3.5). Vismodegib was discontinued on December 28, 2015, and he was switched to sonidegib while clinical trial options were considered. Alternatives to trial participation were discussed, including off-label therapies based on other alterations (ie, *MYCN*, *MLH1*, *PIK3R1*, *ARID1A*, and *TSC1*) and continuing sonidegib, but the patient was ultimately enrolled in a phase I study of an investigational agent starting February 23, 2016.

Restaging scans obtained on April 14, 2016, showed progressive disease, and he was withdrawn from the trial. Enlarged masses in the right trapezius (10.7 cm), lingula (2.8 cm), and right axilla (3.3 cm) were noted. Other therapies, including molecularly targeted therapy and immune checkpoint blockade, were considered. The PD-L1 IHC 28-8 pharmDx assay by NeoGenomics Laboratories stained 0% of tumor; however, in light of the high number of pathogenic alterations (22 genomic alterations in 18 genes; Table 1) and the documentation of an inactivating MLH-1 alteration suggestive of dMMR/MSI-high status, immunotherapy was believed to be a viable option. Off-label authorization was obtained for a PD-1 antibody (pembrolizumab) and treatment was started on May 9, 2016. The treatment course was notable for grade 1 fatigue and dramatic reduction of the right trapezius mass (Figure 2), with

Table 1. Genomic Alterations Identified by FoundationOne Testing

FBXW7 V485fs*13
MLH1 Q445*
NF1 R1204W
PIK3CA P471L
PIK3R1 R386*
PTCH1 A925fs*6, W948*
TSC1 splice site 2392-1G>A
CDKN2A p16INK4a R58* and p14ARF P72L
TP53 R248W
ARID1A splice site 2878+2_2878+2delT, truncation exon 1
ATRX P2299S
BCORL1 Q969*, splice site 4472+1G>A
CHD2 duplication exon 15
FAT1 S1103F
KEL M365I
MAGI2 R564Q
MLL2 Q3580*, R5432W
MYCN P44L

systemic response confirmed on PET/CT scans after 4 infusions (Figure 3). The patient received his last infusion on July 24, 2017, after scans confirmed no evidence of disease, and as of February 2018, he remained disease-free.

Discussion

Molecular profiling is important in resolving diagnostic quandaries and also in leading the way to therapeutic opportunities for rare diseases. This has been shown in the evolution of systemic treatment of unresectable BCC from cytotoxic chemotherapy to a molecularly targeted approach. Chemotherapy has shown some activity in case reports and small series, but advances in understanding BCC genetics and the role of the Shh signaling pathway have allowed for more focused treatment strategies. Currently, 2 hedgehog pathway inhibitors are approved for use in patients with locally advanced BCC that is ineligible for or has recurred after surgery/radiation: vismodegib and sonidegib. Vismodegib was FDA-approved in 2012 based on promising data from ERIVANCE, a multisite, single-arm, phase II trial of 104 patients.¹⁴ Subsequent analyses have shown durable responses and consistent efficacy and safety profiles, with an objective response rate (ORR) of 48.5% and median progression-free survival (PFS) of 9.3 months in patients with metastatic BCC.^{15,16} A subsequent, larger phase II trial (STEVIE) of 1,161 patients with locally advanced or metastatic BCC showed an ORR of

Figure 2. Clinical photographs of the right trapezius mass (A) after vismodegib and phase I therapy, before initiation of pembrolizumab; (B) after 4 pembrolizumab infusions; and (C) after discontinuation of pembrolizumab.

36.9% and median PFS of 13.1 months in the metastatic cohort.^{17,18} Sonidegib was FDA-approved in 2015 based on data from BOLT, a phase II trial in 230 patients that showed an ORR of 23.1% (per investigator review) and median PFS of 13.1 months in the metastatic cohort treated with 200 mg of the agent.^{19–21}

Despite these options, resistance remains a problem. Sonidegib and vismodegib remain the sole FDA-approved targeted therapies for advanced BCC, and little evidence is available to support the use of a second hedgehog pathway inhibitor as a second-line treatment option. One trial found that patients whose advanced BCC was resistant to vismodegib had similar resistance to sonidegib; 3 of 9 had stable disease and none experienced an objective response.²² Effective treatment options are thus extremely limited for patients whose disease progresses on chemotherapy or a hedgehog inhibitor; furthermore, efficacy of these agents in a dedifferentiated clear cell variant of BCC is unknown. This highlights the need for more relevant data for these subgroups, and large-scale trial participation remains crucial although inefficient, owing to the rarity of metastatic BCC and of the clear cell variant in particular. Given these barriers, clinical experience can provide insight and direction for further approaches, as we report herein. Although our patient did not receive further targeted treatment after progression on vismodegib, he had other potentially targetable genomic alterations, and a combination approach might have produced a more meaningful response than hedgehog pathway inhibition alone. For example, truncated *PIK3R1* (R386*) may predict sensitivity to inhibitors of the phosphoinositide 3-kinase (PI3K)/Akt/mTOR pathway, and off-label use of mTOR inhibitors currently approved for other indications (eg, everolimus in renal cell carcinoma,

pancreatic neuroendocrine tumors, and breast cancer; temsirolimus in renal cell carcinoma) may have been a viable option. The splice site mutation in *TSC1* (splice site 2392-1G>A) results in loss of function of the hamartin protein and mTOR activation, also making a case for an off-label trial of an mTOR inhibitor. Presence of the *MYCN* P44L mutation also raises the question whether inhibition of N-Myc expression or indirect targeting of N-Myc would be valuable; Aurora kinase B and BET inhibitors have shown some activity in *MYCN*-driven tumors in preclinical models and early-phase trials, although these were in malignancies with *MYCN* amplification, and it is unclear whether a similar approach would have been of value in this particular case. Also of interest, *ARID1A* alterations are associated with clear cell changes in a variety of other cancers, and the mutation in this case (splice site 2878+2_2878+2delT, truncation exon 1) may be responsible for the phenotype seen. Potential therapeutic targets have been proposed in *AIRID1A*-mutated cancer, including inhibition of histone methyltransferase EZH2 and targeting of PI3K/AKT signaling, thus offering further avenues for combination approaches in metastatic BCC.

Although targeted combinations have the advantage of select and limited toxicities, this approach often results in the development of resistance. Immune checkpoint inhibitors have been shown to improve survival outcomes in melanoma and a variety of solid tumors, and they could be considered to provide more durable response. A variety of tumor characteristics, including dMMR/MSI-high status^{23,24} and high tumor mutational burden,^{25,26} have been shown to be biomarkers for response or increased survival in patients treated with immunotherapy. In this case, the inactivating MLH-1

Figure 3. PET/CT images showing metastatic disease (arrows) response in the (A) lingula, (B) right lung, (C) right axillary lymph node, and (D) right trapezius after treatment with vismodegib, phase I therapy, and pembrolizumab.

alteration (as a marker for dMMR/MSI-high) and high number of pathogenic alterations (as a marker of high tumor mutational burden) were compelling grounds on which to offer off-label treatment with an anti-PD-1 agent, despite the PD-L1-negative status of the tumor. An increasing body of nonrandomized data shows immunotherapy to be clinically relevant in the treatment of metastatic BCCs; combination approaches with vismodegib are already being tested (ClinicalTrials.gov identifier: NCT02690948) and monotherapy with anti-PD-1 is under

ongoing clinical investigation (NCT03132636). Detailed molecular profiling may provide valuable insight into the tumor microenvironment and identify tumor characteristics in patients who may benefit most from such an approach. For example, recent data from mouse models of ovarian cancer have shown that *ARID1A* loss and *PIK3CA* mutation cooperate to promote carcinogenesis through sustained IL-6 production.²⁷ Understanding the landscape of driver mutations and progression pathways in BCC and the mechanisms of resistance to hedgehog pathway

inhibition is essential for establishing the optimal treatment approach.²⁸

Conclusions

Given the aggressive clinical course and molecular profiling results described in this case, consideration of upfront combination therapies (targeted/targeted or targeted/immunotherapy) may provide a more effective treatment strategy for metastatic BCC, especially in patients exhibiting clear cell change. More widespread use of molecular profiling to identify common aberrant signaling pathways and increased involvement

of specialized molecular tumor boards to interpret these results will help inform optimal management of this malignancy.

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