Multiple Hereditary Infundibulocystic Basal Cell Carcinoma Syndrome Associated With a Germline SUFU Mutation

Joshua M. Schulman, MD; Dennis H. Oh, MD, PhD; J. Zachary Sanborn, PhD; Laura Pincus, MD; Timothy H. McCalmont, MD; Raymond J. Cho, MD, PhD

**Importance** Multiple hereditary infundibulocystic basal cell carcinoma syndrome (MHIBCC) is a rare genodermatosis in which numerous indolent, well-differentiated basal cell carcinomas develop primarily on the face and genitals, without other features characteristic of basal cell nevus syndrome. The cause is unknown. The purpose of the study was to identify a genetic basis for the syndrome and a mechanism by which the associated tumors develop.

**Observations** Whole-exome sequencing of 5 tumors and a normal buccal mucosal sample from a patient with MHIBCC was performed. A conserved splice-site mutation in 1 copy of the suppressor of fused gene (SUFU) was identified in all tumors and normal tissue samples. Additional distinct deletions of the trans SUFU allele were identified in all tumor samples, none of which were present in the normal sample.

**Conclusions and Relevance** A germline SUFU mutation was present in a patient with MHIBCC, and additional acquired SUFU mutations underlie the development of infundibulocystic basal cell carcinomas. The downstream location of the SUFU gene within the sonic hedgehog pathway may explain why its loss is associated with relatively well-differentiated tumors and suggests that MHIBCC will not respond to therapeutic strategies, such as smoothened inhibitors, that target upstream components of this pathway.

IMS3

A woman in her 60s presented to our clinic for evaluation of multiple dome-shaped, skin-colored asymptomatic papules located on the face and vulva (Figure 1A and B). These papules first appeared around 50 years of age and gradually increased to approximately 60 lesions. Eight of the lesions underwent biopsy, all of which showed buds and cords of basaloid cells enclosing small cystic spaces, consistent with infundibulocystic BCC (Figure 1C). She had no palmar or plantar pits, and radiographic studies did not reveal any jaw cysts. Therefore, we thought that MHIBCC represented the best diagnosis for her condition. The patient’s medical history was significant for a meningioma. Her family history was notable for a daughter with similar facial papules that had not undergone biopsy.
Owing to her increasing tumor burden, the patient elected to initiate therapy with the smoothened protein (SMO) inhibitor vismodegib; after 9 months of treatment, the size and number of her tumors had not decreased, and therapy was discontinued. The patient provided written informed consent, and our study was approved by the committee on human research at the University of California, San Francisco.

Based on the autosomal dominant inheritance pattern of MHIBCC and the presence of tumors at multiple body sites, we hypothesized that the patient could have a germline mutation in a tumor suppressor gene with acquired second-hit mutations in her tumors. Our approach therefore entailed sampling unaffected (normal) buccal tissue to search for a mutation shared with the patient’s tumor samples, and we selected a total of 5 tumors from the face (n = 3) and vulva (n = 2) for sequencing to identify acquired mutations that, in the case of the vulvar tumors, would not be expected to be exclusively induced by UV radiation.

We prepared libraries for tumor and unaffected samples using a DNA library preparation kit (KAPA Hyper Prep; Kapa Biosystems) and used an exome sequencing system (HiSeq; Illumina) with a reagent kit (Sure Select CRE; Agilent) to provide at least 200× coverage of the transcriptome. Sequencing data for each sample was aligned by the Burrows-Wheeler transform algorithm using default variables, duplicate marked by Samblaster open-source software, and underwent insertion-deletion realignment and base quality recalibration by GATK-Lite (version 2.3; Broad Institute). Tumor vs matched normal tissue variant analysis was performed using an analysis pipeline (Nanomics Contraster; Nanthealth) to determine somatic and germline small variants and estimate relative coverage per exon, as previously described. Small variants were annotated with base-level phastCons conservation scores (UCSC [University of California, Santa Cruz] Genome Bioinformatics), population allele frequencies from the single-nucleotide polymorphism database (dbSNP) (build 137), and their effect, if any, on genes.
Results

We detected deletions of chromosome 10 in all tumor samples by drops in relative coverage and concordant allelic imbalances, estimated by comparing reference and alternate allele sequencing depths in each tumor at heterozygous dbSNP loci in the unaffected sample (Figure 2). Based on the distinct locations and spans of the observed allelic imbalances, the losses on chromosome 10 likely occurred independently in each tumor. We then examined the other copy of chromosome 10 in the tumors, in which we detected a mutation in all of the tumors at a conserved splice acceptor site of intron 6 of the suppressor of fused gene (SUFU [OMIM 607035]) c.757-2A>G. This variant allele was also present on the same copy of chromosome 10 in the normal tissue sample, consistent with a uniparental germline mutation. Unlike in the tumors, the wild-type trans copy of the SUFU gene in the unaffected sample was preserved.

Discussion

In the original report of MHIBCC, 5 individuals from 2 families were identified with numerous infundibulocystic BCCs on
photoexposed (face) and nonphotoexposed (trunk and genital) sites. Genetic analysis of these individuals was performed to search for PTCH1 loss of heterozygosity in their tumors, as would be expected in most cases of BCNS, but this aberration was not detected. In the present investigation, we used whole-exome sequencing to conduct a much broader search for a genetic basis for MHIBCC. We identified a conserved SUFU splice-site mutation in normal and tumor tissue samples from our patient, and we identified additional, distinct deletions affecting the other copy of SUFU in each of the tumors but not in the normal buccal sample. No other mutations were present. Based on these results, we propose that MHIBCC is caused by a germline SUFU mutation with individual infundibulocystic BCCs arising as a result of acquired second-hit SUFU mutations.

The SUFU protein is a component of the sonic hedgehog pathway that acts as a tumor suppressor by binding to and modulating the function of the transcription factor Gli. Mutations of the SUFU gene have been studied mostly in the context of medulloblastoma, in which they have been implicated in sporadic and familial cases, and in the context of familial multiple meningioma. In SUFU also have been identified in several patients who fulfill diagnostic criteria for BCNS but lack PTCH1 mutations. In each of these families with SUFU-associated BCNS, at least 1 family member developed a medulloblastoma or meningioma; this is a much higher rate than is seen in BCNS overall. Our patient also reported a remote history of a meningioma, further supporting this potentially significant association.

Recently, Mann and colleagues described a patient with a germline SUFU splice-site mutation who had 2 children with medulloblastoma; on physical examination, this patient had multiple skin-colored facial papules that histopathologically demonstrated basaloid proliferations with follicular differentiation. We suspect that these facial lesions, characterized as basaloid hamartomas, are actually infundibulocystic BCCs (indeed, the 2 entities may be synonymous) and that this patient shares the same diagnosis as our patient. Consequently, the association between a SUFU mutation and MHIBCC in our patient can be considered a reproducible phenomenon.

Tumors that characterize MHIBCC behave more indolently than other subtypes of BCC. This behavior may reflect the relatively downstream position of SUFU protein in the sonic hedgehog pathway, such that SUFU mutations may be less disruptive to the regulation of the pathway than mutations in upstream components, such as PTCH1 or SMO, might be. The downstream position of SUFU relative to SMO in the hedgehog pathway also means that tumors with SUFU mutations may be resistant to SMO inhibitors or other treatments targeting upstream components of the pathway. This concept is supported by the observation that vismodegib resistance can be achieved through acquired copy number changes or mutations in SUFU, and it likely explains why our patient’s tumors were refractory to vismodegib treatment.

Conclusions

A germline mutation in the tumor suppressor gene SUFU is associated with MHIBCC, and the tumors that develop in this syndrome are induced by additional acquired mutations in the trans copy of that gene. Mutations in SUFU are also found in some patients with BCNS, especially those with a predisposition toward medulloblastoma or meningioma, and MHIBCC and SUFU-associated BCNS may represent phenotypic variants of a common syndrome. A heightened awareness of the risk for medulloblastoma or meningioma in patients with these syndromes and their relatives may be warranted. Patients with these syndromes may not be responsive to vismodegib treatment or other SMO inhibitors. The downstream role of SUFU protein within the sonic hedgehog pathway also provides a potential explanation for the relatively indolent behavior of infundibulocystic BCCs that develop in patients with MHIBCC. Further investigation is warranted to determine whether the same mechanism underlies sporadic infundibulocystic BCCs as well.

Funding/Support: This study was supported by the Well Aging Research Center, Samsung Advanced Institute of Technology, under the auspices of Sang Chul Park, MD, PhD, and by grant K08 CA169865 from the National Cancer Institute, National Institutes of Health (Dr Cho).

Role of the Funder/Sponsor: The funding sources had no role in the design and conduct of the study; collection, management, analysis, and interpretation of the data; preparation, review, or approval of the manuscript; and decision to submit the manuscript for publication.

Additional Contributions: We thank the patient for granting permission to publish this information. We are indebted to Jean Tang, MD, PhD, and Kavita Sarin, MD, PhD, Stanford University, for discussions regarding SUFU mutations in BCNS. NanOnics LLC provided DNA preparation and DNA sequencing. No compensation was given for these contributions.

REFERENCES

Enjoying Opera, Dermatology Style

Abigail L. Alexander, MS; Leonard J. Hoenig, MD

Opera has just about everything: great music, beautiful singing, talented performers, interesting plots, as well as a bit of medicine and dermatology thrown in. To enhance the appreciation of opera among physicians, we provide a discussion of several notable operatic works that explores the intriguing interface between opera and dermatology.

One of the most beloved of all operas is Giacomo Puccini’s La Bohème, which premiered in 1896. The plot centers around the lives of impoverished artists residing in Paris' Latin Quarter during the 1830s and on the romance between Rodolfo, a poet, and Mimi, a seamstress.

In the opening lines, the narrator explains that one of the reasons Rodolfo finds Mimi so attractive is because of the “rosy hue to her clear complexion that had the white velvety bloom of the camellia.”1 Sadly, this appearance was caused by tuberculosis, from which Mimi tragically dies during the final act.

Pallor with red cheeks was often clinically seen in patients with tuberculosis. The pallor resulted from anemia, a common hematologic abnormality in tuberculosis. The rosy cheeks were likely caused by facial flushing from fever.

In discussing La Bohème, it is almost impossible not to mention Rent, a rock opera written and composed by Jonathan Larson as a modern-day reinterpretation of Puccini’s work. Rent, like La Bohème, spotlights the plight of poor artists, this time living in Manhattan’s East Village during 1989. Instead of tuberculosis, Mimi now has human immunodeficiency virus (HIV) infection.

In the 2005 Movie version of Rent, the character Angel suffers from Kaposi’s sarcoma and dies of AIDS. Kaposi’s sarcoma not only disfigured many AIDS patients but also stigmatized them as having HIV infection. Rent, through its humane portrayal of people with AIDS, sought to lessen the stigma of the disease. The musical ends on a positive note as Mimi survives AIDS, surrounded by friends and hopeful for the future.

Another disfiguring disease, syphilis, affects Dr Pangloss, a character in Leonard Bernstein’s 1956 operetta Candide, based on the 1759 satirical novel by Voltaire. Pangloss is described by Voltaire as having “the end of his nose eaten away”2 by syphilis and in the operetta he wears a prosthetic nose. Tertiary syphilis can disfigure the nose in several ways. One clinical presentation was described by the noted French dermatologist Jean-Alfred Fournier (1832-1914). When destruction of the inferior nasal cartilage occurs, the lower nose appears to be pushed into the intact upper nose much like the small tubes of opera glasses which retract into the larger ones.3 Fournier termed this finding “opera glass nose.”

Opera continues to inspire us with its beauty, which touches and elevates the human soul. We hope that this brief dermatologic look at opera will enhance your appreciation of this magnificent form of musical art.

Author Affiliations: University of Miami School of Medicine, Miami, Florida (Alexander); private practice (Hoenig).

Corresponding Author: Leonard J. Hoenig, MD, 601 N Flamingo Rd, No. 201, Pembroke Pines, FL 33028 (gooddoc@h@gmail.com).