# Landscape of Genetic Alterations in Uveal Melanoma


INTRODUCTION

• Uveal melanoma is the most common primary intraocular malignant tumor in adults.
• About 50% of patients develop metastases to the liver within 15 years of their initial diagnosis.
• Uveal melanoma is characterized by frequent mutations in two genes encoding guanine nucleotide binding proteins (G-protein) α-subunits: Gqα (GNAQ) and G-protein α11 (GNA11). These missense mutations arise in G-protein coupled receptors to downstream signaling pathways, including protein kinase C (PKC) ([2], 3).
• Mutations in GNAQ/GNA11 may be the genetic drivers leading to the development of uveal melanoma by causing constitutive activation of pathways involving these genes.
• Here, we conducted genomic profiling of tumor biopsies from patients with metastatic uveal melanoma.

METHODS

Study Design

• Patients with biopsy-proven metastatic uveal melanoma were enrolled in a Phase I, multi-institutional, single-arm study (AEB071).
• Patients received oral AEB071 in one of two dosing schedules at total daily doses ranging from 450 mg to 1450 mg, twice daily or three times daily until disease progression, withdrawal toxicity or withdrawal of consent.
• At baseline, two core needle biopsies were required from each patient for study enrollment.
• If tumor tissue was accessible through a minimally invasive biopsy, then a fresh, pre-dose core needle tumor biopsy was obtained.
• A fresh biopsy could not be collected at study entry, an archival tumor sample from a metastatic lesion was required.
• At least one post-treatment core needle biopsy was also obtained, prior to the morning dose of AEB071 on Cycle 2 Day 25 (±5).

Assessments

• Genetic profiling of FFPE biopsy from patients with metastatic uveal melanoma, primarily from:
  - Genetic profiling was conducted on the baseline pre-treatment biopsy specimens.
  - DNA was assayed by massively parallel sequencing, covering a panel of 388 clinically relevant cancer genes at Foundation Medicine.
  - Sequencing was performed at high depth (median 61X) to characterize mutations, amplifications, translocations, homozygous deletions and loss of heterozygosity (LOH).
• Somatic alterations were found to contain in-frame CNVs or to have insufficient sequence coverage, the CD15 sample was not analyzed. If flow was defined to meet tissue requirements, the sample was excluded from further analysis.

RESULTS

Genetic Alterations

• Aberrations of known or likely functional significance were frequent only in genes previously implicated in uveal melanomas (Figure 2).
• These were distinct from the mutational profile of skin melanoma (Figure 3).
• Two novel pathways were observed.
  - As expected in uveal melanoma biopsies, a large proportion of patients (94%) were found to have activating mutations in either GNAQ-200C [GNAO(+)GNA11], or R143Q or GNA11 (200C). (2006-H).
  - The high prevalence of GNAQ/GNA11 mutations reported here is likely due to the sequencing depth, as evidenced by low allele frequencies in some patients even after correcting for tumor purity.
  - Among the five patients without a GNAQ or GNA11 mutation, all had low tumor cell content (<20%).
  - Truncations or splice-site mutations in the gene encoding BCR1 associated protein 1 (BAP1) were frequently observed (31%), in line with previous reports (Figure 4).
  - Partial or putative complete amplifications of chromosome 8p (16%) and recurrent mutations in splicing factor 3B subunit 1 (SF3B1) (20%) and/or ATP5B were usually mutually exclusive, which is a patient previously unreported (Figure 2).
• In addition, BAP1 mutations were frequently observed, chromosome 3p amplifications (33/48%, 8p), and rare in patients with SF3B1 mutations (1/9).
• Among the 65 patient samples in which LOH could be evaluated, BAP1 spanning LOH occurred in 45 patients (71%), including 35 of 38 patients (95%) with BAP1 mutations.

• Mutations of known or likely functional impact were otherwise rare.
• Somatic alterations in uveal melanoma patients are shown in Figure 4. Genes with mutations in fewer than two patients are not shown. Some patients had short blocks of unknown functional significance. LOH is only included for BAP1.
• The positions of sequenced genes on chromosome 8 are indicated in Figure 4.

CONCLUSIONS

• Genomic profiling has demonstrated that 94% of patients with uveal melanoma had a known activating mutation in either GNAQ or GNA11.
• BAP1 mutations were frequently identified in patients with chromosome 3p amplifications and newly identified in patients with SF3B1 mutations. This suggests a complimentary and redundant consequences of these lesions with BAP1 mutations, respectively.
• High-depth sequencing of clinical patient metastases sheds new light on the interplay among the small group of genetic alterations observed in uveal melanoma.

REFERENCES


Pharmacodynamics

• 51 paired tumor biopsy samples were analyzed to investigate the effect of genetic subtypes on baseline levels of pMARCKS.
• Preliminary analyses of the currently available data suggest no obvious relationship between biomarkers and patient response.

Studied Genes

• pMARCKS
• pERK
• pAEB071

Figure 1. Patient Samples

Figure 2. Somatic Gene Alterations Observed in Tumor Samples

Figure 3. Top Mutated Genes in Skin Melanoma

Figure 4. Somatic Alterations in Uveal Melanoma Patients

Figure 5. Normalized pMARCKS at Baseline vs Genotype (Full Analysis Set)

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