Gene Expression Profiling and Regression Rate of Irradiated Uveal Melanomas

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BACKGROUND AND OBJECTIVE: Uveal melanoma is the most common primary intraocular cancer; however, the molecular features that predict response to therapy are poorly understood. Our objective was to determine whether gene expression profiling (GEP) is associated with rate of tumor regression after I-125 plaque brachytherapy for uveal melanoma.

PATIENTS AND METHODS: Retrospective review of 138 patients with posterior uveal melanoma treated with I-125 plaque brachytherapy in which GEP class and 3-month post-radiation ultrasonographic tumor thickness data were available. Statistical analysis was performed using t test and Fisher’s exact test.

RESULTS: GEP class assignment was class 1 in 83 (60.1%) and class 2 in 55 (39.9%) patients. Mean patient age was 60.9 years for class 1 and 68.1 years for class 2 tumors (P = .002). Mean initial tumor diameter was 13.0 mm for class 1 and 14.1 mm for class 2 tumors (P = .02). Mean initial tumor thickness was 5.2 mm for class 1 and 6.1 mm for class 2 tumors (P = .047). Three months after I-125 plaque radiotherapy, mean reduction in tumor thickness was 26.5% for class 1 and 16.7% for class 2 tumors (P = .03).

CONCLUSION: Class 1 uveal melanoma tumors exhibit more rapid early tumor regression than class 2 tumors after I-125 plaque radiotherapy. [Ophthalmic Surg Lasers Imaging Retina. 2015;46:333-337.]

INTRODUCTION

Uveal melanoma is the most common primary intraocular cancer and is most frequently treated in the U.S. with iodine-125 (I-125) episcleral plaque brachytherapy (EPB).1 Although EPB is associated with excellent local tumor control, 50% of patients will go on to develop metastasis. Gene expression profiling (GEP) accurately classifies patients according to metastatic risk: class 1 tumors have a low risk and class 2 tumors have a high risk of metastasis independent of primary tumor treatment.2 A GEP clinical test that is now commercially available is the most accurate molecular prognostic test available for uveal melanoma and the only one to be validated in a prospective, multicenter study.3 The objective of this study was to determine whether GEP class status predicts tumor regression at 3 months after EPB.

PATIENTS AND METHODS

To determine whether GEP correlates with response to iodine-125 plaque brachytherapy, we studied 138 patients who met the following three criteria: (1) underwent I-125 plaque brachytherapy for posterior uveal melanoma, (2) obtained the GEP test from a fine needle biopsy, and (3) had ultrasonographic tumor thickness measurement at baseline and at the 3 month postoperative visit by the same examiner (JWH). These 138 patients were treated between June 1, 2001, and October 28, 2011, and were identified from a total of 563 patients with choroidal melanoma treated at Washington University over a 15-year period, from November 1, 1996, through October 28, 2011. The study was approved by the Washington University institutional review board.

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RESULTS

Study patients included 67 (48.5%) women and 71 (51.5%) men (Table). GEP testing found tumor class 1 in 83 (60.1%) and class 2 in 55 (39.9%) patients. Mean patient age was 60.9 years (median: 62.0 years) for class 1 and 68.1 years (median: 67.7 years) for class 2 tumors ($P = .002$). Mean initial tumor diameter was 13.0 mm (median: 13.0 mm) for class 1 and 14.1 mm (median: 14.9 mm) for class 2 tumors ($P = .02$). Mean initial tumor thickness was 5.2 mm (median: 4.8 mm) for class 1 and 6.1 mm (median: 6.0 mm) for class 2 tumors ($P = .047$). Mean post-treatment tumor thickness was 3.9 mm (median: 3.6 mm) for class 1 and 5.0 mm (median: 4.2 mm) for class 2 tumors ($P = .002$). Mean decrease in tumor thickness was 1.35 mm (median: 1.0 mm) for class 1 and 1.19 mm (median: 0.8 mm) for class 2 ($P = .2$). Mean percent decrease in tumor thickness 3 months after radiotherapy was 26.5% for class 1 and 16.7% for class 2 tumors ($P = .03$). Four class 1 tumors, but no class 2 tumors, exhibited complete regression to a flat scar (ultrasonographic thickness = 0 mm; Figure); however, this was not a statistically significant difference ($P = .16$). Additionally, these regressed tumors were associated with a transient panuveitis.

DISCUSSION

GEP class is derived from the expression of 15 genes by the melanoma. This profile has been shown to correlate with metastatic risk, with class 1 tumors considered low-risk and class 2 tumors considered high-risk. It has been previously shown that uveal melanomas with class 1 GEP have a 2% to 21% risk of metastasis at 5 years, whereas those with the class 2 profile have a 72% risk of metastasis at 5 years. In our population, tumors with a class 1 GEP regressed more rapidly after brachytherapy than tumors with class 2 GEP. Although absolute decreases in tumor thickness were similar between the two groups, proportional decrease was greater in tumors with class 1 GEP (26.5% vs 13.0%; $P = .01$).

Interestingly, four cases of complete regression, in which the tumor regressed completely to a flat scar (3-month ultrasonographic measurement = 0 mm), were present in class 1 tumors. All such cases were associated with transient panuveitis, perhaps suggesting that immune-mediated regression may account for the more rapid regression of some class 1 tumors (Figure). Immune-mediated regression of melanoma has been observed, yet the mechanisms remain unclear. For instance, regression of cutaneous melanoma, which develops spontaneously in a species of miniature pigs, is positively associated with acute uveitis and is associated with migration of lymphocytes and monocytes into the stroma of the ciliary body, iris, choroid, band keratopathy, cataracts, and death of uveal melanocytes. Other studies have described enhanced regression of intraocular melanoma when melanoma cells are mutagenized in vitro prior to intraocular injection in a murine model of intraocular melanoma. Mutagenesis enhanced expression of class I major histocompatibility complex and increased susceptibility of the tumor graft to CD8+ cytotoxic T lymphocyte-mediated killing and to tumor necrosis factor-mediated cytolysis. Whether certain class 1 tumors are distinctly susceptible to these particular uveitic or immunogenic-killing mechanisms remains uncertain and deserves further study.

<table>
<thead>
<tr>
<th>Variable (Mean ± SD)</th>
<th>Class 1 (n = 83)</th>
<th>Class 2 (n = 55)</th>
<th>$P$ Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age at Diagnosis (yrs)</td>
<td>60.9 ± 13.4</td>
<td>68.1 ± 12.5</td>
<td>.002</td>
</tr>
<tr>
<td>Sex</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>36</td>
<td>47</td>
<td>.1</td>
</tr>
<tr>
<td>Male</td>
<td>32</td>
<td>23</td>
<td></td>
</tr>
<tr>
<td>Initial tumor largest diameter (mm)</td>
<td>13.0 ± 2.9</td>
<td>14.1 ± 2.7</td>
<td>.02</td>
</tr>
<tr>
<td>Initial tumor thickness (mm)</td>
<td>5.2 ± 2.2</td>
<td>6.1 ± 2.7</td>
<td>.047</td>
</tr>
<tr>
<td>Tumor thickness 3 mos after treatment (mm)</td>
<td>3.9 ± 1.9</td>
<td>5.0 ± 2.3</td>
<td>.002</td>
</tr>
<tr>
<td>Absolute decrease in tumor thickness 3 mos after treatment (mm)</td>
<td>1.4 ± 1.4</td>
<td>1.2 ± 1.7</td>
<td>.6</td>
</tr>
<tr>
<td>Percent reduction in tumor thickness 3 mos after treatment</td>
<td>26.5 ± 27.2</td>
<td>16.7 ± 24.6</td>
<td>.03</td>
</tr>
</tbody>
</table>
Figure 1. Complete regression of a class 1 posterior uveal melanoma 16 months after plaque therapy. (A) Funduscopic photograph of a macular uveal melanoma (arrowhead), with orange pigment and associated exudative retinal detachment inferiorly (double arrowheads). The ultrasonographic pre-treatment thickness was 2.66 mm. (B) Following episcleral plaque therapy, the tumor regressed to a flat scar (arrowhead) at 3 months as measured by B-scan ultrasonography, with resolution of the exudative retinal detachment (double arrowheads). The 3-month ultrasonographic post-treatment thickness was 0 mm.
The 5-year rate of metastasis for class 1 tumor ranges between 2% and 21%. Of the four patients who developed flat scars during the study period within 3 months of radiotherapy, one developed metastasis (25%) approximately 3 months after plaque radiotherapy. However, this patient had concurrent cutaneous melanoma and developed metastases to the lung and mediastinum, which are relatively unusual sites of metastases for primary uveal melanoma. He underwent ipilimumab therapy for late-stage cutaneous melanoma. Because the metastatic foci were not biopsied, we cannot determine whether the observed metastases originated from uveal or cutaneous sites. Even if the metastases were from the class 1 uveal melanoma, our small sample size of rapidly and completely regressed tumors and lack of statistical significance that distinguishes the likelihood of class 1 versus 2 tumor undergoing complete regression preclude a determination of a possible link between GEP class status, metastasis, and rapid and complete regression. A larger study would be helpful to further assess whether a relationship exists among these variables.

Recently, Correa et al performed a similar analysis of GEP class and post-brachytherapy regression, but found no correlation based on either absolute or proportional decrease in thickness. At 3 months, we observed a mean magnitude of regression of 1.35 mm in class 1 tumors and 1.19 mm in class 2 tumors; however, Correa et al found a mean regression of 1.3 mm in both class 1 and class 2 tumors. Their reported 3-month percentage decrease in the size of class 1 tumors was similar to ours (22.4% vs 20.9%) but higher for class 2 tumors (21.0% vs 16.7%). Their study included 50 patients, 25 with class 1 and 25 with class 2 tumors, whose initial tumor thicknesses were matched within 0.5 mm. In our study, consistent with a previous study, pre-treatment thicknesses and largest basal diameters of class 2 tumors were found to be greater than class 1 tumors (Table; \( P = .047 \)). In contrast to Correa et al, our report and six other studies that have calculated tumor regression rates following I-125, ruthenium-106, cobalt-60 brachytherapy, and proton beam irradiation as a function of genetic attributes (eg, GEP class or chromosome 3 status) or likelihood of subsequent metastasis have not matched individual initial tumor thicknesses. The reason for this may be because such pair-wise matching may disproportionately exclude tumors with distinct genetic features that tend to present with thicker initial tumor thicknesses, such as tumors with class 2 status, monosomy 3, or with otherwise high propensities to metastasize. Taken together, these differences may account for some of the distinct findings observed between Correa et al and our report.

Similar to GEP status and tumor regression after I-125 brachytherapy, the relationship among initial tumor thickness, genetic features, regression rate, metastasis, and mortality remains controversial with regard to other treatment modalities such as ruthenium-106 brachytherapy and proton beam irradiation. A previous study by one of us (JWH), which included 126 patients treated with proton beam irradiation, did not find an association among the absolute magnitude of tumor thickness regression in the velocity or rate of thickness change at 24 months after treatment between class 1 and class 2 GEP. In this report, however, class 1 and class 2 tumors had a statistically significant difference in initial thickness, and tumor thickness at 3 months after proton beam irradiation was not measured. With regard to proton beam irradiation, regression, and mortality before the era of genetic testing, Glynn et al described a more complex response relationship with proton beam irradiation: rapid initial regression during the first 2 years after irradiation portended a higher risk of metastasis during this post-treatment period. However, tumors that regressed slowly after the first 2 years had a higher risk of metastasis 2 or more years after treatment.

In the case of ruthenium-106, Kaiserman et al reported that tumors with a greater decrease in tumor thickness in the first 3 months had a higher rate of metastasis and a lower rate of survival. Shields et al and Marathe et al seemed to confirm these findings by assessing monosomy 3 status, a genetic feature of choroidal melanoma highly associated with metastasis and death, in patients with I-125 brachytherapy. Marathe et al found that initial thickness and absolute and proportional reduction in thickness were greater among tumors with monosomy 3 versus disomy 3 (a feature associated with increased metastasis-free survival) when measured 0.5 to 3.33 years after I-125 brachytherapy. Shields et al’s findings were more complex: monosomy 3 was associated with a more rapid regression rate at 12 and 15 months, but in contrast to what one would predict based on Kaiserman et al’s study, monosomy 3 was not associated with more rapid regression 4, 8, or 18 months after I-125 brachytherapy. Also in contrast to Kaiserman et al, Chiam et al found that tumors with monosomy 3 did not have a significant difference in regression at 6 months when compared to tumors with disomy 3. The relationship between response to brachytherapy and metastasis and survival has been studied even before the advent of genetic profiling. Augsburger et al showed that rapid regression of choroidal melanoma after cobalt-60 brachytherapy is associated with lower metastatic-free survival.
In summary, the conclusions of previous studies relating to tumor regression, genetic features, metastasis, and survival remain conflicting, with no consensus.

While our report arrives at conclusions distinct from conflicting previous studies, it has important limitations including its relatively small sample size and retrospective study design. The follow-up time was short, although by design, because our goal was to analyze early tumor response after brachytherapy. As a result of this short follow-up time, we are unable to identify any relationship between response of brachytherapy and metastasis, precluding a determination of whether response to brachytherapy can predict tumor metastasis within the class 1 or class 2 tumors. Interestingly, four of the class 1 tumors but no class 2 tumors exhibited rapid regression to a flat scar at 3 months by ultrasonography. While these cases of complete regression in class 1 may appear to be outliers, statistical analysis of all cases in the GEP class 1 data set indicates that all Z-scores (representing individual percentage changes in tumor thicknesses subtracted from the mean percentage change, with the corresponding difference divided by the standard deviation) fall within –3 to +3. Thus, the cases of complete regression in class 1 are unlikely to represent outliers and were not excluded from analysis.

In conclusion, we demonstrate that class 1 tumors regress proportionally more than do class 2 tumors. Complete regression at 3 months was observed only among class 1 cases.

REFERENCES


