Disrupting anti-apoptotic BCL-xL sensitizes SHH-driven medulloblastomas to SHH inhibition

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Medulloblastoma Background

- Medulloblastoma is the most common malignant pediatric brain tumor.
- Current treatments result in an ~80% survival rate.
- Issues: therapies still fail 20% of patients and the treatment toxicity can cause long-term neurocognitive deficits.
- Goal: To improve the efficacy of medulloblastoma treatments to increase survival outcomes and decrease treatment toxicity.

SHH signaling drives medulloblastoma growth

BCL-xL prevents apoptosis by inhibiting BAX and promotes tumor progression in a SHH subtype medulloblastoma mouse model

Experimental Design

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Endpoint</th>
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<tbody>
<tr>
<td>M-Smo</td>
<td>Injection terminated at P74, humane endpoint, Loss of 15% of body weight average, sickly, unresponsive, bone loss, Mice harvested for IHC study, Brain fixed with 4% PFA</td>
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<tr>
<td>M-Smo (POx-Vismo)</td>
<td>Injection terminated at P74, humane endpoint, Loss of 15% of body weight average, sickly, unresponsive, bone loss, Mice harvested for IHC study, Brain fixed with 4% PFA</td>
</tr>
<tr>
<td>M-Smo/Bcl-xL cKO (POx-Vismo)</td>
<td>Injection terminated at P74, humane endpoint, Loss of 15% of body weight average, sickly, unresponsive, bone loss, Mice harvested for IHC study, Brain fixed with 4% PFA</td>
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Results

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Conclusions

1. Inhibiting SHH signaling via POx-Vismo prolonged survival of Bcl-xL-intact tumor bearing mice.
2. Bcl-xL deletion increased the long-term survival of POx-Vismo-treated tumor mice, as 35.6% of mice with Bcl-xL-deleted tumors reached P100, compared to 15.7% for the Bcl-xL-intact controls.
3. Bcl-xL deletion prevents recurrence of POx-Vismo-treated tumor mice, as no residual tumors were found in any MSmo/Bcl-xL cKO mice at P100, while all of the P100 MSmo mice had residual tumors.

Future Directions

1. Target both the SHH signaling pathway and BCL-xL pharmacologically.
   - POx-ABT263 = inhibits BCL-xL and induces spontaneous apoptosis in vivo
2. Combine BCL-xL deletion, or pharmacologically inhibit BCL-xL using POx-ABT263, with radiation treatment.
   - Radiation = targets proliferating tumor cells

References


Hypothesis

Targeting proliferation by inhibiting SHH signaling pharmacologically will sensitize medulloblastomas to Bcl-xL deletion.

Table 1: Sample size, median survival age, and percent survival to P100 of MSmo untreated mice, MSmo POx-Vismo treated mice, and MSmoser Bcl-xL cKO POx-Vismo treated mice.

<table>
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<tr>
<th>Sample Size</th>
<th>Median Survival Age</th>
<th>Percent Survival to P100</th>
</tr>
</thead>
<tbody>
<tr>
<td>MSmo (untreated)</td>
<td>29</td>
<td>22</td>
</tr>
<tr>
<td>MSmo (POx-Vismo)</td>
<td>19</td>
<td>28</td>
</tr>
<tr>
<td>MSmo/Bcl-xL cKO (POx-Vismo)</td>
<td>14</td>
<td>60.5</td>
</tr>
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Figure 1: Bcl-xL deletion induces focal necrosis and increases survival of medulloblastoma-bearing mice.

(A) Representative images of Bcl-xL-intact (top, MSmo) and Bcl-xL-deleted (bottom, MSmo/Bcl-xL−/−) SHH subtype medulloblastomas in mice. Bcl-xL-deleted tumors showed regions of focal necrosis (arrow) and regions of acellularity (*), indicating cell death.

(B) Survival of MSmo, MSmo/Bcl-xL−/−, and MSmo/Bcl-xL cKO mice. Deletion of one copy of Bcl-xL did not prolong survival, but deletion of both copies of Bcl-xL did (p = 0.019).

Figure 2: SHH inhibition prolongs survival of SHH medulloblastoma-bearing mice and sensitizes tumors to Bcl-xL deletion.

(A) Survival of MSmo, MSmo (POx-Vismo), and MSmo/Bcl-xL−/− mice. Deletion of both copies of Bcl-xL did not prolong survival (p = 0.14).

(B-C) Representative H&E stains of tumors from MSmo and MSmo/Bcl-xL−/− mice at P100. (B) Bcl-xL-intact MSmo mice had residual tumors at P100, while (C) MSmo/Bcl-xL−/− mice had no residual tumors.