EUROPEAN HEMATOLOGY ASSOCIATION

EHA2021 VIRTUAL

## S199

# BH3-MIMETICS AND AZACITIDINE SHOW SYNERGISTIC EFFECTS ON JUVENILE MYELOMONOCYTIC LEUKEMIA

Topic: 15. Myeloproliferative neoplasms - Biology & Translational Research

Keywords: Apoptosis BCL2 JMML

Naile Koleci[1], Ying Wu[1], Lorena Gallego-Villar[1], Geoffroy Andrieux[2], Jovana Rajak[1], Konrad Aumann[3], Charlotte M. Niemeyer[1], Christian Flotho[1], Miriam Erlacher[1]

[1] Department of Pediatric Hematology and Oncology, University Medical Center Freiburg, Freiburg im Breisgau, Germany

[2] Institute of Medical Bioinformatics und Systems Medicine IBSM, Freiburg, Germany., University Medical Center Freiburg, Freiburg im Breisgau, Germany

[3] Institute of Pathology, Freiburg, Germany., University Medical Center Freiburg, Freiburg im Breisgau, Germany

**Background:** Juvenile myelomonocytic leukemia (JMML) represents an aggressive myeloid neoplasia of early childhood, driven by oncogenic RAS signaling and epigenetic deregulation. Most patients require allogeneic stem cell transplantation (HSCT), but relapse risk remains high. Azacitidine effectively reduces leukemia burden in some JMML patients. Nonetheless, not all patients respond to this treatment, and alternative therapeutic strategies are urgently required. BH3-mimetics inhibit anti-apoptotic BCL-2 proteins (i.e. BCL-2, BCL-XL, MCL-1 and/or BCL-W) thereby inducing apoptosis directly at the mitochondrial level. These compounds hold big promise for anti-cancer therapy, either alone or in combination with other drugs. Recently, venetoclax, a specific BCL-2 inhibitor, was FDA-approved in combination with hypomethylating agents for the treatment of acute myeloid leukemia (AML) in adults.

**Aims:** Based on these promising results obtained in AML, we investigated different BH3-mimetics in JMML, alone or in combination with azacitidine.

**Methods:** For this purpose, we generated patient-derived xenograft (PDX) mice and treated them with ABT737, an analogue of navitoclax known to inhibit BCL-2, BCL-XL and BCL-W. Treatment was initiated 8 weeks after transplantating JMML cells. ABT737 was regularly given for the next 28 days (75 mg/kg/d), and on day 29, mice were sacrificed and analyzed by flow cytometry and histopathology. Mock-treated mice were used as controls. In addition, a group of mice was treated with azacitidine as described earlier (i.e. 2 cycles á 5 days followed by 9-day breaks, 3 mg/kg/d).

**Results:** Both ABT737 and azacitidine led to efficient leukemia depletion. We then analyzed whether both drugs could be used at lower concentration when given in combination, with the aim to minimize any potential side effect. Indeed, a 28 days long treatment with 0.75 mg/kg/d azacitidine (5 days on, 9 days off) and 50 mg/kg/d ABT737 (daily) did not only completely reduce leukemia burden but also depleted all leukemia-initiating cells as confirmed by serial transplantation experiments. The combination of drugs worked significantly better than both drugs alone.

To dissect the roles of BCL-2 and BCL-XL, both inhibited by navitoclax, and to additionally analyze the role of MCL-1 in the survival of JMML cells, we performed *in vitro* experiments using the specific inhibitors ABT199, A1155463 and S63845, respectively. When used alone, only ABT737 and the MCL-1 inhibitor S53845 killed JMML cells efficiently. However, synergistic effects were noted between azacitidine and the BCL-XL inhibitor A1155463. Azacitidine treatment resulted in the upregulation of pro-apoptotic BCL-2 proteins (i.e. BIK, PUMA, BAD and BMF) and a slight downregulation of MCL-1 indicating that azacitidine and the BCL-XL inhibitor synergized at the level of the BCL-2 protein family.

**Summary/Conclusion:** In sum, our study confirms that the effects of azacitidine are significantly increased by adding BH3-mimetics. Such combination strategies will be especially important for those patients that are resistant to azacitidine monotherapy.

**Copyright Information:** (Online) ISSN: 2572-9241


**Abstract Book Citations:** Authors, Title, HemaSphere, 2021;5:(S2):pages. Abstract Book, DOI:
http://dx.doi.org/10.1097/HS9.0000000000000566



EHA2021 Virtual
JUNE 9-17 2021
POWERED BY M-ANAGE.COM