Preclinical Evaluation of HDP-101, a Novel anti-BCMA Antibody-Drug Conjugate, in Multiple Myeloma

1Max-Eder Group “Experimental Therapies for Hematologic Malignancies”, Internal Medicine V, Heidelberg University Hospital and German Cancer Research Center, Heidelberg, Germany; 2Heidelberg Pharma Research GmbH, Ladenburg, Germany; 3Department of Internal Medicine V, Heidelberg University, Germany; 4National Center of Tumor Diseases (NCT), Heidelberg, Germany

Results

Cytotoxic potency

MM cell lines U266, MM.1S, INA-6, SKMM.1, NCI-H929 and LP-1 were used to test the cytotoxicity of HDP-101 (Figure 1). Doses of 10 μM were used for most cell lines, reduced viability of U266 and SKMM.1 at 10 μM.

Specificity

MM cell lines U266, MM.1S, INA-6, SKMM.1, NCI-H929 and LP-1 were digested with Cog Spartan and Cog SPC T and CTL control (Figure 5). D5D is sterically only found in certain plant species and absent in the human genome. Current Cog ATAC showed no cytotoxic activity up to a concentration of 0.1 ml for most cell lines, reduced viability of U266 and SKMM.1 at 10 μM.

In vivo efficacy

Antitumor activities of ADCs were determined in a MM.1S Luc intravenous xenograft model and NCI-H929 subcutaneous xenograft model (data not shown) in vivo.

Methods

Cell lines and antibodies: The human multiple myeloma cell lines U266, MM.1S, NCI-H929 as well as the bone marrow stroma cell line HS-5 were obtained from the ATCC, SKMM.1 was obtained from the DSMZ (Germany), LP-1 was obtained from the University center of myeloma Heidelberg (Germany). NCI-115 was obtained from Renate Burger at the department of stemcell and bone marrow therapy in Kiel (Germany). NCI-H-929 was obtained from the Life Science Institute (Japan). The Luciferase-transfected multiple myeloma cell line MM.1S-Luc was provided by the Max Delbrück Center for Molecular Medicine (MDC, Berlin). Antibodies were obtained from anti-BCMA antibodies developed at the NDC. Thiomab derivatives thereof were produced by HDP using Espal biotechnology and Humanization transfection methods.

Patient: Patient samples were provided from patients according to the criteria of the International Myeloma Working Group were included.

Primary MM cell: Primary MM cell lines were isolated from MM patient whole bone marrow samples with magnetic CD123 microbeads (Miltenyi Biotec). Cell lines were examined by flow cytometry (ACS-1, Bioscience) and microarrays (Oligo, CO803 1188 1184 02) (bright markers).

Flow cytometry: Bone marrow stroma cells (BMSC) were isolated from MM patient whole bone marrow samples by taking microcarrier cells (MSC) from sample in culture medium. MPhi were obtained from Blood/Fetal Medium (DMEM) 10% Fetal Calf Serum (FCS) (YAC, CO803 1188 1184 02). The conjugate was purified by SEC and dialysis. DAR (drug-antibody ratio) according to LC-MS analysis was 3.30±1.05 amantin per IgG.

Synthesis of anti-IG-ATAC: The Anti-IG-ATAC was synthesized similar to HDP-101 by conjugating HDP 30 2115 to engineered cysteine residues resulting in an ADC with a DAR of 2.0.

Cell viability assay: Quantitative determination of cell viability was performed by CellTiter-Glo 2.0 assay (Promega).

Flow cytometry: Viability test of BCMA on cells was determined with PE-labeled anti-BCMA antibody (Heidelberg Pharma) and Quantibrite PE beads (BD) as a reference using ACCURI C6 (BD Biosciences).

Animal models: MM.1S-Luc: 6 to 8 week-old female SCID big mice were obtained from Charles River. 10 5 MM.1S-Luc cells were intravenously into their tail vein. Once a mean total flux of around 1.5 x 10⁶ - 1.5×10⁷/10⁸/10⁹/d after implantation was reached, animals were intravenously. Luciferase activity was monitored by non-invasive bio imaging (Edgetech). NTP studies were performed in treatment-naive female cynomolgus monkeys at LPT (Germany, Hamburg).

Conclusion

- HDP-101 showed in vitro cytotoxic potency on several BCMA-positive myeloma cell lines and non-proliferating primary CD138+ cells at pico- to nanomolar concentrations.
- Amanitin-based ADCs (ATACs) in the therapy of multiple myeloma are a novel promising approach with a distinct mode of action to overcome drug resistance and improve patient outcome.

References


Disclosures

- BMF35.01.16.2.02.002: Biohit research funding from Heidelberg Pharma Research GmbH.

Contact details

Andreas-Paahl: andreas-paahl@heidelberg-pharma.de
Marc-Stein-Fraulich: m.stein-fraulich@heidelberg-pharma.de