Abstracts

of T-cells into a regulatory phenotype. In a murine B16 xenograft model IL-2 expression significantly enhanced therapeutic effects of an H1 oncolytic influenza virus. Expressed within the background of H5 hemagglutinin, IL-2 did not lead to a significant enhancement of therapeutic efficacy. Interestingly, the empty influenza H5 subtype was significantly more potent in treating B16 xenograft tumors than the H1 subtype, regardless of IL-2 expression. In primary human PBMC models, the virus based on H1 hemagglutinin led to enhanced CD8 T-cell activation compared to H5. This effect was further enhanced by IL-2 expression, although all viruses led to significant activation. Surprisingly, viruses based on H1 hemagglutinin led to increased expression of the immune checkpoint PD-1. The virus based on H5 hemagglutinin did not lead to upregulation of PD-1, indicating a favorable balance between activation and exhaustion. Infection with the H5 based virus led to both enhanced apoptosis and immunogenic calreticulin exposure in human and murine melanoma cell lines compared to H1.

Conclusions IL-2 does not promote Tregs, when expressed in a viral background. H1 viruses induced PD-1 more potently than H5 viruses. The choice of viral entry protein is more relevant for the therapeutic effect of the virus, than the expression of a T-cell stimulating cytokine such as IL-2. Efficacy of oncolytic viral treatment appears to depend more on viral growth than on virally expressed T-cell promoting cargo.


P09.05
IMMUNOGENICITY INDUCED BY THE ACADEMIC CHIMERIC ANTIGEN RECEPTOR CAR19 (ARI-0001) IN PATIENTS WITH CD19-POSITIVE RELAPSED/REFRACTORY B-CELL MALIGNANCIES RECRUITED INTO THE CART19-BE-01 CLINICAL TRIAL

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Background Chimeric Antigen Receptor (CAR)-T cells directed against CD19 have induced high rates of response in patients with relapsed/refractory (R/R) B-cell malignancies. Two CD19-targeting constructs have been approved by the FDA and EMA (Yescarta®, Kymriah®) for B lymphoblastic leukemia (B-ALL) and aggressive lymphoma. Despite deep remissions, there are still major challenges and disparate data are reported about the immunogenicity induced by CART-cell therapy. On May/2017, the Spanish Agency of Medicines approved our first clinical trial (clinicaltrials.gov NCT03144583) with a fully academic CART-19.

Materials and Methods Eligibility criteria included R/R B-ALL (adult and pediatric), non-Hodgkin’s lymphoma (NHL) and chronic lymphocytic leukemia (CLL) who failed standard therapy. The primary objective of the study was safety and secondary objectives were response rate and its duration. The humoral anti-CART response was assessed by a (cell-based) fluorescence assay to detect human anti-murine antibodies (HAMA) in patients sera. Assessment was conducted at different time points: 1) at baseline (pre-dose), 2) on day 14 after the administration of ARI-0001 cells, 3) on day 28, 4) on day 100, and 5) every 3 months thereafter.

Results Forty-seven patients (37 adults/10 pediatrics) received ARI-0001 cells, Thirty-eight patients had a diagnosis of R/R B-ALL (28 adults and 10 children); all but 5 had relapsed after allogeneic hematopoietic stem cell transplant (HCT). Seven patients had a diagnosis of NHL, four of them (57%) had relapsed after HCT, and 2 patients had a diagnosis of CLL (2). Median age was 27 years (3-68). After conditioning with fludarabine (90 mg/m2) and cyclophosphamide (900 mg/m2), a total dose of 0.5–5 x106 ARI-0001 cells/kg was infused. Autologous T-cells from peripheral blood were expanded and transduced with a lentivirus to express a CAR with a single-chain variable fragment (scFv) with anti-CD19 specificity, conjugated with the co-stimulatory regions 4-1BB and CD3z. The scFv was originated from a mouse monoclonal antibody A3B1. Twenty-five per cent of the patients tested positive for the presence of anti-CAR antibodies, all of them post-dose, in contrast to previous data reported on Kymriah® with a significant presence of pre-dose anti-murine CAR19 antibody. Of these 12 patients, 8 patients presented with a weak, and 4 patients with a strong presence of HAMA. The last 4 patients had lost the effectiveness of the CART-therapy at that time point, reflected by simultaneous B-cell recovery in the periphery. Moreover, three of them received a second dose of CART-19, which did not revert the relapse.

Conclusions To conclude, these data suggest the importance of the immunogenicity induced by CART-cell therapies. Immune monitoring should include the assessment of humoral response, especially before considering a second dose after relapse.


P09.06
‘AN ENHANCED CRISPR TOOL FOR TREATING CHRONIC MYELOGENOUS LEUKEMIA’

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Background Chronic myeloid leukemia (CML) is a myeloproliferative neoplastic disease, occurring in 1 to 2 cases per 100,000 adults, which accounts this type of cancer for approximately 15% of newly diagnosed leukemia in adult patients. The diagnosis is based upon the genetic translocation between the t(9;22)(q34;q11.2), resulting in formation