

BEN versus CY

Purpose: Compare the effect of bendamustine (BEN) compared to cyclophosphamide (CY) treatment on dendritic cells (DCs) by RNAsequencing.

Mice:

6 BALB/c females, age-matched

Groups:

(3 mice/group)

BEN 40 mg/kg

CY 200 mg/kg

Timeline:

Day -2:

Injection of 200 mg/kg CY intraperitoneally or 40 mg/kg BEN intravenously (based on average weight on day -2).

Day 0:

Dendritic Cell Isolation

Harvest spleens from BEN- or CY-treated mice. Create single cell suspensions in Complete Media (**CM**) (RPMI-1640 with 10% FBS, 1% Sodium Pyruvate, 1% MEM NEAA, and 100 U/mL penicillin-streptomycin). Lyse red blood cells with Pharm Lyse (BD), wash cells in PBS, and then count total splenocytes. Isolate splenic Pan-DCs using the Pan DC Isolation Kit from Miltenyi (130-100-875). Count total DCs and then centrifuge.

Cells were resuspended in 100 μ L PBS before adding 500 μ L RNeasy Lysis Buffer (Thermo Fisher). Samples were kept on ice, transferred to labeled 1.7 mL Eppendorf tubes, and delivered to the University of Arizona Genomics Core (UAGC) for processing.