Cytokine Release Evaluation Studies
Cytokine release syndrome (CRS) is a life-threatening side-effect of immuno-therapeutics. Current assays for CRS lack the sensitivity and specificity to provide translationally meaningful results, leading to challenges in early clinical trials. JAX has a rapid, sensitive and reproducible human immune cell based preclinical platform enabling safety assessment of immuno-therapeutics.
JAX’s Solution Demonstrates the Donor to Donor Variability of Response Typically Observed in the Clinical Setting

**Treatment**
- Two human PBMC donors
- Anti-CD28 mAb
- TGN1412 biosimilar
  - CD28 superagonist
- Quantitation of human cytokines at 1 & 4 hrs

**Observations**
- Replication of clinically observed cytokine release
- Human donor specific differences in drug induced CRS response
- Highly reproducible responses for any given donor
CRS with Bispecific Antibodies Requires Both Arms Engaged and Can Be Enhanced with Addition of Checkpoint Blockade Drugs

**Treatment**

- Three human PBMC donors
- Anti-CD28 mAb
- Keytruda
- Bispecific: EGFRxCD3
- Combination: EGFRxCD3 + Keytruda (Donors “B” and “C”)

**Observations**

- Replication of clinically observed cytokine release with anti-CD28 within a given donor
- Cytokine releases with the bispecific is tumor dependent and variable across donors
- Cytokine releases with the bispecific is enhanced with addition of checkpoint inhibitor in some donors
  - Increase in cytokines is variable between the two donors

![TNF-α (3 Donors, 6h Post Treatment)](image1)

![IFN-γ (3 Donors, 6h Post Treatment)](image2)
Cytokine Release Assay (In Vitro Assay) versus JAX CRS Evaluation Study (In Vivo Assay)

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<thead>
<tr>
<th>CRA</th>
<th>CRS Evaluation Study</th>
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<tbody>
<tr>
<td>Inconsistent Response across assays (whole blood or isolated PBMC),</td>
<td>Holistic physiological conditions (human immune cells and mouse circulation/vasculature) offering in vivo relevance</td>
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<td>dependent on test article MoA, evidence of false positives</td>
<td>Permits tumor engraftment to assess on-target toxicity and efficacy</td>
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<td>CRAs don’t offer dynamic range to discriminate low, medium or high</td>
<td>Reproducible results, independent of test article MoA (OKT-3, TGN1412, α-CD28, lenalodome)</td>
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<tr>
<td>responders</td>
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<td>Quantification of cytokine levels vary across platforms and are</td>
<td>Curated PBMC bank with pre-characterized response to positive and negative controls</td>
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<td>disconnected from potential clinical implications</td>
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<td>Lack physiological conditions to offer in vivo relevance</td>
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