

# 1. Technical Data Sheet

<b>Summary</b>	<p>StimCytes™ CD19 Evaluation Kit synthetic cell mimics are lyophilized particles that feature selected biomarkers for use in CAR-T potency assays. This product contains no bio-hazardous material, so it is safe to use in any environment and requires no special disposal.</p> <p><b>For Research Use Only. Not for use in diagnostic or therapeutic procedures.</b></p>
<b>Application</b>	<p>Specific activation of CD19-targeted CAR-T for potency evaluation in co-culture assay with negative control.</p>
<b>Materials</b>	<p>StimCytes™ are hydrogels that are in lyophilized form. This product is lyophilized for stability and ease of use.</p> <p>StimCytes™ CD19 Evaluation Kit (SL-00035) is composed of three vials:</p> <ol style="list-style-type: none"> <li>1. StimCytes™ CD19 (SL-00037)</li> <li>2. StimCytes™ CD19 with Enhancers (SL-00038)</li> <li>3. StimCytes™ Negative (SL-00039)</li> </ol> <p>Each sealed glass vial contains 2M lyophilized particles.</p>
<b>Handling and Safety</b>	<p>No special handling or safety precautions are necessary.</p>
<b>Storage</b>	<p>Store lyophilized products at -20°C upon receipt.</p>
<b>Expiration</b>	<p>24 months from the date of manufacturing when stored at -20°C. Use the entire vial immediately upon reconstitution of lyophilized product.</p>
<b>Instructions for Use</b>	<ol style="list-style-type: none"> <li>1. Tap down the vial to ensure that all cell mimics are collected at the bottom of the vial.</li> <li>2. Add 1000 µL of media of choice to the vial and transfer contents to 15mL conical tube. Add additional 1000 µL of media to vial, incubate 30 seconds to one minute, and transfer to same 15mL conical tube.             <ol style="list-style-type: none"> <li>a. Note: Multiple vials can be combined into one 15mL conical.</li> <li>b. Note: PBS with 1% BSA or RPMI with 10% FBS are recommended buffers for reconstitution. Please reach out to Slingshot Biosciences (support@slingshotbio.com) for alternate media suggestions.</li> </ol> </li> <li>3. Pellet cell mimics at 3000xg 3 mins at room temperature. Carefully remove supernatant from 15 mL tube with 1000 µL pipette, or aspirate if mimic pellet is easily identifiable.</li> <li>4. Resuspend pellet in 1000 µL media per starting vial and repeat step 3.</li> </ol>

5. Resuspend pellet in 1000 µL media. Remove sample and count particles using a flow cytometer, hemacytometer, or automated counting instrument that incorporates phase contrast imaging to identify cells.
  - a. Note: If counts are performed on automated cell counter instrument with trypan blue exclusion, use total cell count. Automated cell counter instruments that rely exclusively on dye-based detection will require alternate methodology. Please reach out to Slingshot Biosciences (support@slingshotbio.com) for instrument-specific protocols or to discuss alternative options or custom solutions.
6. Pellet remaining sample as in (3), carefully remove supernatant, and resuspend mimics at desired concentration for assay.

QC Data

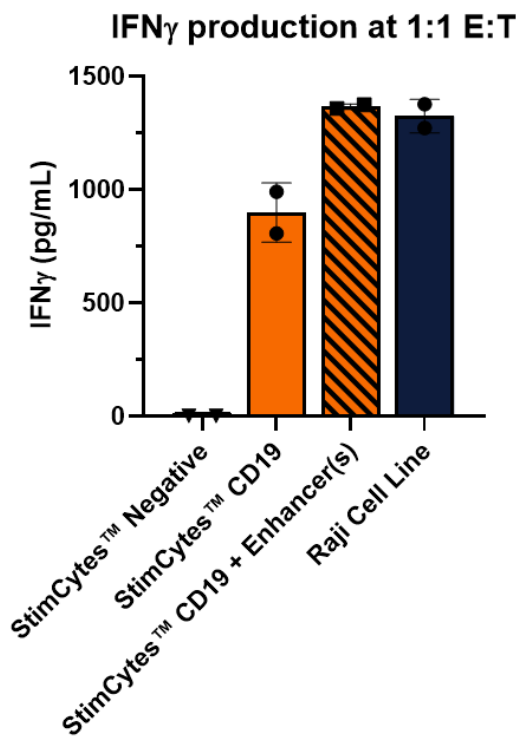


Figure 1. **Specific activation of CD19 CAR-T.** CD19 CAR-T were cultured at 1:1 E:T with StimCytes™ Negative (SL-00039), StimCytes™ CD19 (SL-00037), and StimCytes™ CD19 with Enhancers (SL-00038), or Raji cell line. Supernatant collected after 18 hours of culture was quantified for IFN $\gamma$  with BD CBA on Cytex Aurora.

Technical Support

For technical support from our Cell Therapy scientists, please contact [support@slingshotbio.com](mailto:support@slingshotbio.com)

Individual results may vary. Slingshot Biosciences Cell Therapy scientists are available for technical support and suggestions for customization to achieve optimal results.