

Product Sheet

GAS Reporter HCT116 Cell Line

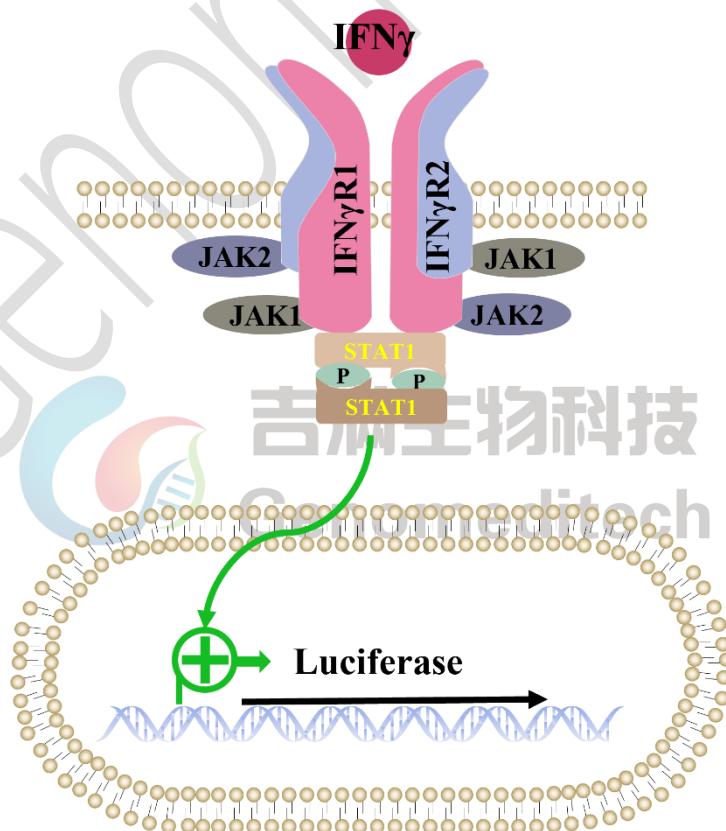
Catalog number: GM-C38096

Version 3.3.1.260316

GAS (Gamma-Activated Sequence) is a key cis-regulatory DNA element in cytokine signaling pathways, regulating gene expression. It is a binding site for transcription factors like STATs, activated by cytokine stimuli. Discovered in interferon (IFN)-induced genes and closely linked with STAT1, GAS is commonly found in promoter regions of genes responsive to IFN stimulation. It is crucial in immunity, proliferation, and differentiation.

The GAS pathway is driven by the JAK-STAT pathway. Cytokines like IFN- γ activate Janus kinases (JAKs), which phosphorylate receptors to recruit STAT proteins. Phosphorylated STATs dimerize, move to the nucleus, and bind GAS sequences to trigger gene expression. For example, IFN- γ activates STAT1 to regulate host defense genes. This pathway plays critical roles in immunity, inflammation, and cellular growth, with abnormalities linked to autoimmune diseases, inflammation, and tumors.

GAS Reporter HCT116 Cell Line is a clonal stable HCT116 cell line constructed using lentiviral technology, along with GAS signal-dependent expression of a luciferase reporter gene. When IFN γ binds to IFN γ R, it activates downstream signaling pathways, leading to the expression of luciferase. The luciferase activity measurement indicates the activation level of the signaling pathway and can thus be used to evaluate the in research of drugs related to GAS.



Specifications

Quantity	5E6 Cells per vial, 1 mL
Product Format	1 vial of frozen cells
Shipping	Shipped on dry ice
Storage Conditions	Liquid nitrogen immediately upon receipt

Recovery Medium	McCoy's 5A+10% FBS
Growth medium	McCoy's 5A+10% FBS+1% P.S+10 µg/mL Blasticidin
Note	None
Freezing Medium	90% FBS+10% DMSO
Growth properties	Adherent
Growth Conditions	37°C, 5% CO ₂

Mycoplasma Testing	The cell line has been screened to confirm the absence of Mycoplasma species.
Safety considerations	Biosafety Level 2
Note	It is recommended to expand the cell culture and store a minimum of 10 vials at an early passage for potential future use.

Materials

Reagent	Manufacturer/Catalogue No.
McCoy's 5A	VivaCell/C3020-0500
Fetal Bovine Serum	ExCell/FSP500
Pen/Strep	Thermo/15140-122
Blasticidin	Genomeditech/ GM-040404
IFN-gamma	MCE/HY-P70610
Recombinant Human IFN- α 2 (carrier-free)	BioLegend/592702
GMOne-Step 2.0 Luciferase Reporter Gene Assay Kit	Genomeditech/ GM-040513

Figures

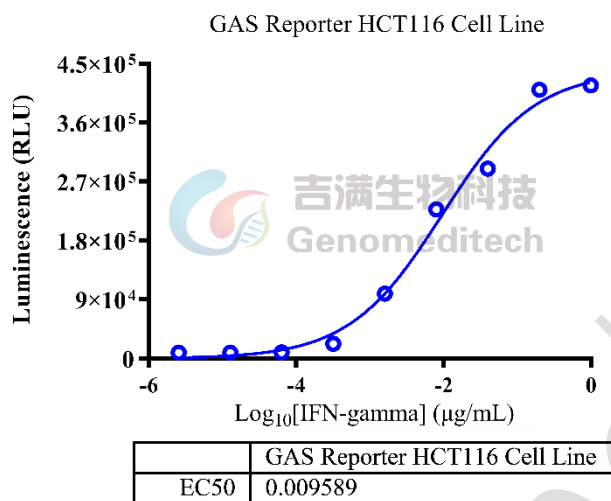


Figure 1 | Response to IFN-gamma. The GAS Reporter HCT116 Cell Line (Cat. GM-C38096) at a concentration of 2E4 cells/well (96-well format) was stimulated with serial dilutions of IFN-gamma (MCE/HY-P70610) in assay buffer (McCoy's 5A + 1% FBS + 1% P.S) for 16 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The maximum induction fold was approximately [50.7]. Data are shown by drug mass concentration.

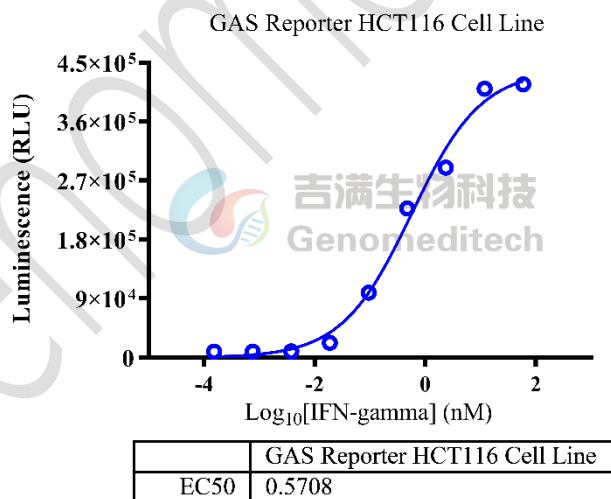


Figure 2 | Response to IFN-gamma. The GAS Reporter HCT116 Cell Line (Cat. GM-C38096) at a concentration of 2E4 cells/well (96-well format) was stimulated with serial dilutions of IFN-gamma (MCE/HY-P70610) in assay buffer (McCoy's 5A + 1% FBS + 1% P.S) for 16 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The maximum induction fold was approximately [50.7]. Data are shown by drug molar concentration.

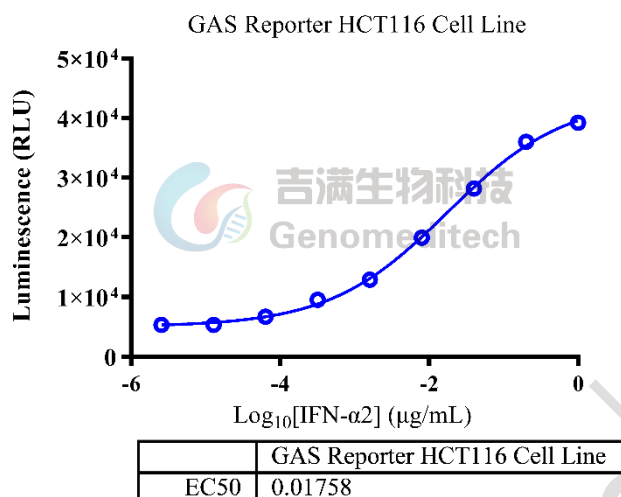


Figure 3 | Response to Recombinant Human IFN- α 2. The GAS Reporter HCT116 Cell Line (Cat. GM-C38096) at a concentration of 2E4 cells/well (96-well format) was stimulated with serial dilutions of Recombinant Human IFN- α 2 (Biolegend/592702) in assay buffer (McCoy's 5A + 1% FBS + 1% P.S) for 16 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The maximum induction fold was approximately [7.8]. Data are shown by drug mass concentration.

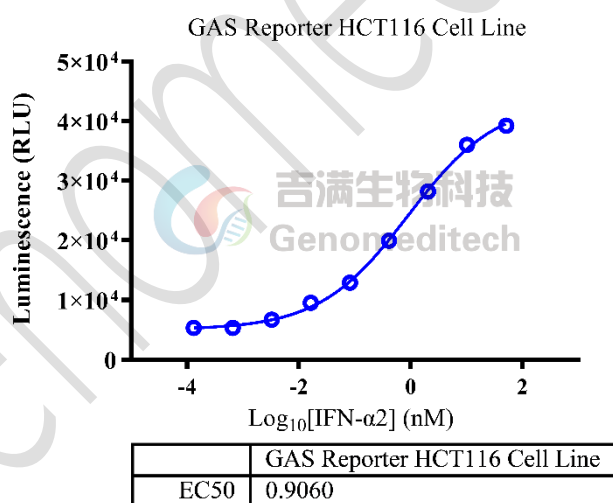


Figure 4 | Response to Recombinant Human IFN- α 2. The GAS Reporter HCT116 Cell Line (Cat. GM-C38096) at a concentration of 2E4 cells/well (96-well format) was stimulated with serial dilutions of Recombinant Human IFN- α 2 (Biolegend/592702) in assay buffer (McCoy's 5A + 1% FBS + 1% P.S) for 16 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The maximum induction fold was approximately [7.8]. Data are shown by drug molar concentration.

Cell Recovery

Recovery Medium: McCoy's 5A+10% FBS

To insure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be stored in liquid nitrogen vapor phase and not at -70°C . Storage at -70°C will result in loss of viability.

- a) Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 - 3 minutes).
- b) Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions.
- c) Transfer the vial contents to a centrifuge tube containing 5.0 mL complete culture medium and spin at approximately $176 \times g$ for 5 minutes. Discard supernatant.
- d) Resuspend cell pellet with the recommended recovery medium. And dispense into appropriate culture dishes.
- e) Incubate the culture at 37°C in a suitable incubator. A 5% CO_2 in air atmosphere is recommended if using the medium described on this product sheet.

Cell Freezing

Freezing Medium: 90% FBS+10% DMSO

- a) Centrifuge at $176 \times g$ for 3 minutes to collect cells.
- b) Resuspend the cells in pre-cooled freezing medium and adjust the cell density to 5×10^6 cells/mL.
- c) Aliquot 1 mL into each vial.
- d) Place the vial in a controlled-rate freezing container and store at -80°C for at least 1 day, then transfer to liquid nitrogen as soon as possible.

Cell passage

Growth medium: McCoy's 5A+10% FBS+1% P.S+10 $\mu\text{g}/\text{mL}$ Blasticidin

For the first 1 to 2 passages post-resuscitation, use the recovery medium. Once the cells have stabilized, switch to a growth medium.

- a) Remove and discard culture medium.
- b) Briefly rinse the cell layer with PBS to remove all traces of serum that contains trypsin inhibitor.
- c) Add 1.0 mL of 0.25% (w/v) Trypsin-EDTA solution to dish and observe cells under an inverted microscope until cell layer is dispersed (usually within 1 to 2 minutes at 37°C).
- d) Note: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate dispersal.
- e) Add 2.0 mL of growth medium to mix well and aspirate cells by gently pipetting.
- f) After centrifugation, resuspend the pellet and add appropriate aliquots of the cell suspension to new culture vessels.
- g) Incubate cultures at 37°C .

Subcultivation Ratio: A subcultivation ratio of 1:3 - 1:4 is recommended

Medium Renewal: Every 1 to 2 days

Notes

- a) After the stabilization of the cell condition, there will be fewer dead cells post-passage, the cell growth rate will tend to stabilize, cell morphology will become uniform, and the cells will appear robust.

Related Products

IFN- α	
IFNα Reporter HEK-293 Cell Line	IFNα Reporter MDCK Cell Line
IFNα Reporter THP1 Cell Line	

License Agreement:

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