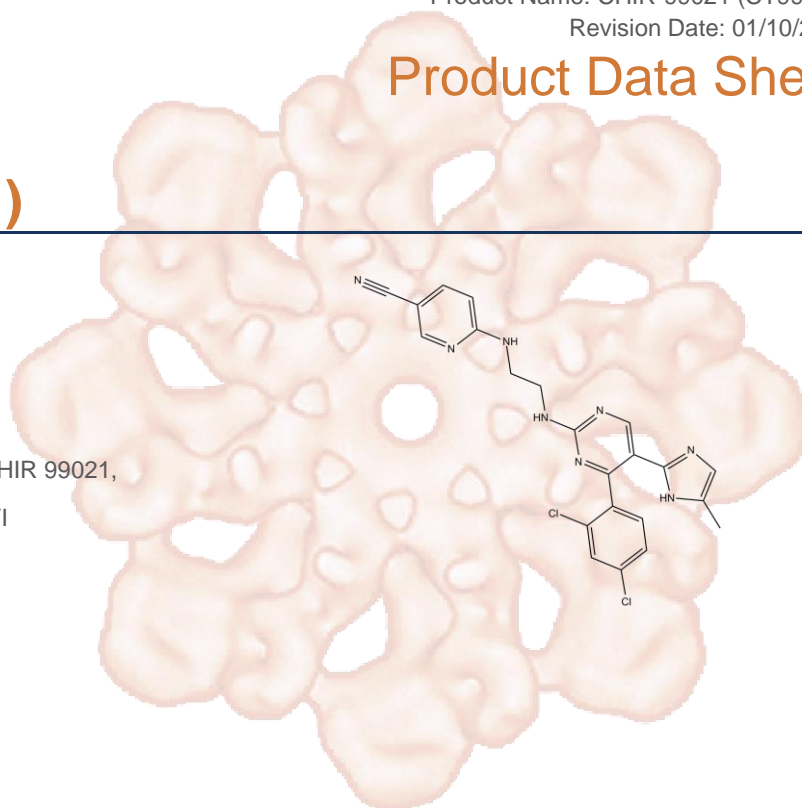


## CHIR-99021 (CT99021)

<b>Cat. No.:</b>	A3011
<b>CAS No.:</b>	252917-06-9
<b>Formula:</b>	C <sub>22</sub> H <sub>18</sub> Cl <sub>2</sub> N <sub>8</sub>
<b>M.Wt:</b>	465.34
<b>Synonyms:</b>	CHIR99021, CHIR-99021, CHIR 99021, CT99021, GSK-3 Inhibitor XVI
<b>Target:</b>	PI3K/Akt/mTOR Signaling
<b>Pathway:</b>	GSK-3
<b>Storage:</b>	Store at -20°C



## Solvent & Solubility

≥23.3mg/mL in DMSO

In Vitro

Preparing Stock Solutions	Solvent		Mass		
	Concentration		1mg	5mg	10mg
	1 mM		2.1490 mL	10.7448 mL	21.4897 mL
	5 mM		0.4298 mL	2.1490 mL	4.2979 mL
	10 mM		0.2149 mL	1.0745 mL	2.1490 mL

Please refer to the solubility information to select the appropriate solvent.

## Biological Activity

Shortsummary

GSK-3 inhibitor, Cell-permeable, ATP-competitive

IC<sub>50</sub> & Target

7 nM (GSK-3)

In Vitro

### Cell Viability Assay

Cell Line: Human embryonic stem cells (ESCs)

Preparation method: The solubility of this compound in DMSO is >10 mM. General tips for obtaining a higher concentration: Please warm the tube at 37 °C for 10 minutes and/or shake it in the ultrasonic bath for a while. Stock solution can be stored below -20°C for several months.

Reacting conditions: 8 μM, 24 hours

	Applications:	On day 0, differentiation was initiated with 8 $\mu$ M CHIR-99021 for 24 h conferring canonical Wnt/ $\beta$ -catenin activation, followed by Wnt inhibition on day 3 by addition of 4 $\mu$ M IWR-1 for 48 h. For all groups tested, first signs of eGFP-fluorescence and first beating EBs were observed on day 6 with a constant increase until d10, reaching almost 100% of beating EBs for the groups seeded with 666-2000 cells per aggregate. Flowcytometry analysis of dissociated EBs on day 10 showed the highest yield of 5.9 Nkx2-5-eGFP+ cells for the group seeded with 666 cells/aggregate; higher cell numbers per aggregate resulted in lower yields. Immunofluorescence stainings of EB cryosections and dissociated/reseeded cells confirmed a high content of Nkx2-5+ and cTnT+ cardiomyocytes, thereby demonstrating efficient cardiomyogenic differentiation of human ESC-derived EBs after aggregation on agarose microwells and induction with small molecule-based media.
In Vivo	<b>Animal experiment</b>	
	Animal models:	Akita type 1 diabetic mice and wild-type mice
	Dosage form:	Intraperitoneal injection, 50 mg/kg, daily
	Applications:	At 15 min after the propranolol injection, the 2-min average HF fraction increased from $46.8 \pm 2.9\%$ before CHIR-99021 treatment to $67.8 \pm 5.1\%$ after CHIR-99021 treatment. Treatment of Akita mice with CHIR-99021 increased SREBP-1 from $0.53 \pm 0.07$ - to $1.17 \pm 0.11$ - fold. CHIR-99021 treatment increased GIRK4 levels from $0.28 \pm 0.06$ - to $1.08 \pm 0.14$ - fold of those in WT mice, which was significantly higher than in placebo.
Other notes:	Please test the solubility of all compounds indoor, and the actual solubility may slightly differ with the theoretical value. This is caused by an experimental system error and it is normal.	

## Product Citations

1. Kaisari S, Shomer P, et al. "Role of Polo-like kinase 1 in the regulation of the action of p31(comet) in the disassembly of mitotic checkpoint complexes." Proc Natl Acad Sci U S A. 2019 Jun 11;116(24):11725-11730.PMID:31118282
2. Karuna EP, Choi SS, et al. "Identification of a WNT5A-Responsive Degradation Domain in the Kinesin Superfamily Protein KIF26B." Genes (Basel). 2018 Apr 5;9(4). pii: E196.PMID:29621187
3. Siller R, Sullivan GJ. "Rapid Screening of the Endodermal Differentiation Potential of Human Pluripotent Stem Cells." Curr Protoc Stem Cell Biol. 2017 Nov 15;43:1G.7.1-1G.7.23.PMID:29140570

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## References

- [1] Dahlmann J, Kensah G, Kempf H, et al. The use of agarose microwells for scalable embryoid body formation and cardiac differentiation of human and murine pluripotent stem cells. Biomaterials, 2013, 34(10): 2463-2471.
- [2] Zhang Y, Welzig C M, Picard K L, et al. Glycogen synthase kinase-3 $\beta$  inhibition ameliorates cardiac parasympathetic dysfunction in

## Caution

### FOR RESEARCH PURPOSES ONLY.

### NOT FOR HUMAN, VETERINARY DIAGNOSTIC OR THERAPEUTIC USE.

*Specific storage and handling information for each product is indicated on the product datasheet. Most APExBIO products are stable under the recommended conditions. Products are sometimes shipped at a temperature that differs from the recommended storage temperature. Shortterm storage of many products are stable in the short-term at temperatures that differ from that required for long-term storage. We ensure that the product is shipped under conditions that will maintain the quality of the reagents. Upon receipt of the product, follow the storage recommendations on the product data sheet.*

**APExBIO Technology**

**[www.apexbt.com](http://www.apexbt.com)**

7505 Fannin street, Suite 410, Houston, TX 77054.

Tel: +1-832-696-8203 | Fax: +1-832-641-3177 | Email: [info@apexbt.com](mailto:info@apexbt.com)

