**TWS119**

**Cat. No.:** HY-10590  
**CAS No.:** 601514-19-6  
**Molecular Formula:** C\(_{18}\)H\(_{14}\)N\(_{4}\)O\(_{2}\)  
**Molecular Weight:** 318.33  
**Target:** GSK-3; Autophagy  
**Pathway:** PI3K/Akt/mTOR; Stem Cell/Wnt; Autophagy  
**Storage:**  
- Powder: -20°C 3 years, 4°C 2 years  
- In solvent: -80°C 1 year, -20°C 6 months

**SOLVENT & SOLUBILITY**

**In Vitro**  
DMSO: ≥ 50 mg/mL (157.07 mM)  
H\(_2\)O: < 0.1 mg/mL (ultrasonic; warming; heat to 60°C) (insoluble)  
* "≥" means soluble, but saturation unknown.

<table>
<thead>
<tr>
<th>Preparing Stock Solutions</th>
<th>Solvent Concentration</th>
<th>1 mg</th>
<th>5 mg</th>
<th>10 mg</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 mM</td>
<td>3.1414 mL</td>
<td>15.7070 mL</td>
<td>31.4139 mL</td>
<td></td>
</tr>
<tr>
<td>5 mM</td>
<td>0.6283 mL</td>
<td>3.1414 mL</td>
<td>6.2828 mL</td>
<td></td>
</tr>
<tr>
<td>10 mM</td>
<td>0.3141 mL</td>
<td>1.5707 mL</td>
<td>3.1414 mL</td>
<td></td>
</tr>
</tbody>
</table>

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

**In Vivo**  
1. Add each solvent one by one: 10% DMSO >> 40% PEG300 >> 5% Tween-80 >> 45% saline  
   Solubility: ≥ 2.5 mg/mL (7.85 mM); Clear solution  
2. Add each solvent one by one: 10% DMSO >> 90% (20% SBE-β-CD in saline)  
   Solubility: ≥ 2.5 mg/mL (7.85 mM); Clear solution

**BIOLOGICAL ACTIVITY**

**Description**  
TWS119 is a specific inhibitor of GSK-3β, with an IC\(_{50}\) of 30 nM, and activates the wnt/β-catenin pathway.

**IC\(_{50}\) & Target**  
GSK-3β  
30 nM (IC\(_{50}\))

**In Vitro**  
TWS119 induces neuronal differentiation in P19 EC cells and primary mouse ESCs. TWS119 (< 4 μM) significantly enhances the proliferation and survival of γδ T cells via activation of the mammalian target of rapamycin (mTOR) pathway, upregulation of the expression of anti-apoptotic protein Bcl-2 and inhibition of cleaved caspase-3. TWS119 (0-8 μM) induces
the generation of CD62L⁺γδT or CCR5⁺γδT cell phenotypes. TWS119 (0.5, 1.0 and 2 μM) increases the expression level of granzyme B in a dose-dependent manner. TWS119 also enhances the cytolytic activity of γδT cells against tumour cells in vitro[2].

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

**In Vivo**

TWS119 (30 mg/kg, i.p.) improves the neurologic function and decreases neurologic deficit score in rtPA-treated MCAO rats. TWS119 effectively relieves cerebral edema, and reduces cerebral infarction in rats treated with rtPA. TWS119 also effectively decreases blood-brain barrier permeability in rtPA-Treated MCAO Rats and attenuates rtPA-induced hemorrhage in ischemic brain tissue. Furthermore, TWS119 activates the Wnt/β-Catenin signaling pathway and increases the expression of Claudin-3 and ZO-1[1].

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

### PROTOCOL

**Cell Assay**[3]

PBMCs are cultured with pamidronate disodium for 8 days and then cells are labelled with or without 1.5 μM carboxyfluorescein succinimidyl ester (CFSE) and CFSE-labelled cells are then seeded in 6-well plates (2.5 × 10⁶ cells/well) followed by treatment with various concentrations of TWS119 for 72 h. The total number of cultured cells is evaluated using an automated cell counter and the γδT cell proliferation is examined by flow cytometry[3].

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**Animal Administration**[2]

All rats are randomly divided into four groups as follows: Sham group-rats undergo the same surgical procedure, but the filament is not inserted and they receive 1 mL of dimethyl sulfoxide (1 % DMSO in saline); Vehicle group-rats undergo MCAO and receive 1 mL of DMSO; rtPA group-rats underwent MCAO and receive rtPA (10 mg/kg, Actilyse®) at 4 h after MCAO; and rtPA+TWS119 group-rats undergo MCAO and receive intraperitoneal TWS119 (30 mg/kg, dissolved in 1 mL 1 % DMSO) immediately after rtPA injection at 4 h after MCAO[2].

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

### CUSTOMER VALIDATION

- J Lipid Res. 2019 Dec;60(12):2020-2033.

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### REFERENCES

