# SEA0400

**Cat. No.:** HY-15515  
**CAS No.:** 223104-29-8  
**Molecular Formula:** C₂₁H₁₉F₂NO₃  
**Molecular Weight:** 371.38  
**Target:** Na+/Ca²⁺ Exchanger  
**Pathway:** Membrane Transporter/Ion Channel  
**Storage:**  
- Powder:  
  - -20°C: 3 years  
  - 4°C: 2 years  
- In solvent:  
  - -80°C: 6 months  
  - -20°C: 1 month

## SOLVENT & SOLUBILITY

**In Vitro**  
DMSO: ≥ 32 mg/mL (86.17 mM)  
* "≥" means soluble, but saturation unknown.  

<table>
<thead>
<tr>
<th>Preparing Stock Solutions</th>
<th>Solvent Mass</th>
<th>1 mg</th>
<th>5 mg</th>
<th>10 mg</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1 mM</td>
<td></td>
<td>2.6927 mL</td>
<td>13.4633 mL</td>
<td>26.9266 mL</td>
</tr>
<tr>
<td>5 mM</td>
<td></td>
<td>0.5385 mL</td>
<td>2.6927 mL</td>
<td>5.3853 mL</td>
</tr>
<tr>
<td>10 mM</td>
<td></td>
<td>0.2693 mL</td>
<td>1.3463 mL</td>
<td>2.6927 mL</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 (6.73 mM); Clear solution

## BIOLOGICAL ACTIVITY

**Description**  
SEA0400 is a novel and selective inhibitor of the Na⁺-Ca²⁺ exchanger (NCX), inhibiting Na⁺-dependent Ca²⁺ uptake in cultured neurons, astrocytes, and microglia with IC₅₀S of from 5 to 33 nM.

**IC₅₀ & Target**  
IC₅₀: 5-33 nM (NCX)

**In Vitro**  
SEA0400 inhibits Na⁺-dependent ⁴⁵Ca²⁺ uptake in cultured neurons, astrocytes, and microglia. IC₅₀ values of SEA0400 are 33 nM (neurons), 5.0 nM (astrocytes), and 8.3 nM (microglia)[¹]. SEA0400 prevents sodium nitroprusside (SNP) to increase ERK and p38 MAPK phosphorylation, and production of reactive oxygen species (ROS) in an extracellular Ca²⁺-dependent manner[²].

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In Vivo

SEA0400 (3 mg/kg + 3 mg/kg/h for 2 h, i.v.) attenuates the infarct volume in the cerebral cortex and striatum, does not affect the mean the regional cortical blood flow in anesthetized rats[1]. SEA0400 protects against the dopaminergic neurotoxicity (determined by dopamine levels in the midbrain and striatum, tyrosine hydroxylase immunoreactivity in the substantia nigra and striatum, striatal dopamine release, and motor deficits) in MPTP-treated C57BL/6J mice[3].

PROTOCOL

Kinase Assay [1]

Na+–Ca2+ exchange activity is determined by assaying Na+-dependent 45Ca2+ uptake as reported previously. Briefly, the cells are preincubated in Hanks’ balanced saline solution (HBSS) for 20 min, and the medium is switched to HBSS containing 45Ca2+ and incubated for 5 min. To increase intracellular Na+ concentration, 1 mM ouabain plus 20 μM monensin (for astrocytes and microglia) and 10 μM monensin (for neurons) are used. Monensin is added simultaneously with the isotope. Ouabain is added 5 min before monensin in astrocytes and microglia. SEA0400 and KB-R7943 are added 5 min before monensin and present during 45Ca2+ uptake reaction.

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

Cell Assay [1]

Cells, plated in 96-well plastic tissue culture plates, are incubated at 37°C for 30 min in normal or Ca2+-free HBSS containing 10 μM H2DCF-DA and 0.25 μg/mL Cremophor EL, and then rinsed twice with normal HBSS to remove excess dye. The cells are reperfused in normal HBSS for 1 h, and the conversion of H2DCF-DA to its fluorescent product dichlorofluorescein by ROS, presumably H2O2 and hydroxyl radical, is determined with excitation at 485 nm and emission at 535 nm using a Wallac Multilabel counter. ROS production is expressed as a percentage of control cells. The linearity and sensitivity of ROS assay are confirmed using H2O2 prior to the experiment. SEA0400 at the indicated concentrations is added 10 min before Ca2+ reperfusion and present until assay.

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

Animal Administration [1]

Male Sprague-Dawley rats, weighing 289 to 325 g, are anesthetized with 1 to 2% halothane. A catheter is inserted into the femoral artery and connected to a pressure transducer to record blood pressure. Regional cortical blood flow is measured by a laser Doppler flowmeter, with probe placement at 2 mm posterior and 6 mm lateral to the bregma. SEA0400 or its vehicle with an equivalent volume is i.v. injected at 3 mg/kg and then infused at 3 mg/kg/h for 2 h under normal conditions without MCA occlusion.

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

CUSTOMER VALIDATION

- JCI Insight. 2019 Apr 25;5. pii: 128765.

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REFERENCES
