### Intoplicine

**Cat. No.:** HY-101647  
**CAS No.:** 125974-72-3  
**Molecular Formula:** C₂₁H₂₄N₄O  
**Molecular Weight:** 348.44  
**Target:** Topoisomerase  
**Pathway:** Cell Cycle/DNA Damage  
**Storage:** Please store the product under the recommended conditions in the COA.

### Solvent & Solubility

**In Vitro**  
10 mM in DMSO

<table>
<thead>
<tr>
<th>Preparing Stock Solutions</th>
<th>Solvent</th>
<th>Mass</th>
<th>1 mg</th>
<th>5 mg</th>
<th>10 mg</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Concentration</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1 mM</td>
<td></td>
<td>2.8699 mL</td>
<td>14.3497 mL</td>
<td>28.6993 mL</td>
<td></td>
</tr>
<tr>
<td>5 mM</td>
<td></td>
<td>0.5740 mL</td>
<td>2.8699 mL</td>
<td>5.7399 mL</td>
<td></td>
</tr>
<tr>
<td>10 mM</td>
<td></td>
<td>0.2870 mL</td>
<td>1.4350 mL</td>
<td>2.8699 mL</td>
<td></td>
</tr>
</tbody>
</table>

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

### BIOLOGICAL ACTIVITY

**Description**  
Intoplicine is a DNA topoisomerase I and II inhibitor.

**IC₅₀ & Target**  
Topoisomerase I; Topoisomerase II

**In Vitro**  
Activity of Intoplicine (RP60475), a new DNA topoisomerase I and II inhibitor, against human tumor colony-forming units in vitro. Intoplicine (RP60475) is the most active analogue evaluated in the 7H-benzo[e]-pyrido-[4,3-b]-indole series of antineoplastic compounds. Intoplicine exerts its activity through inhibition of DNA topoisomerase I and II[1]. Intoplicine is found to unwind DNA and to inhibit purified calf thymus topoisomerase II via a stabilization of the ternary cleavable complex[2].

### PROTOCOL

**Kinase Assay [2]**  
The assay is performed with various concentrations of calf thymus Topo II (20 to 0.1 decatenation units) in a 20 μL final reaction volume containing 0.25 μg of supercoiled pBR322 DNA, 20 mM Tris HCl (pH 7.5), 60 mM KCl, 10 mM
MgCl\(_2\), 30 μg/mL bovine serum albumin, 0.5 mM EDTA, 0.5 mM Dithiothreitol, and 1 μM Intoplicine or water. The final nucleotide concentration is 20.8 μM. The reaction is assembled in ice and the reaction mixture is then incubated at 37°C for 5 min. Then sample is mixed at room temperature with 20 μL of preaggregated silver hydrosol and immediately analyzed by SERS. Control experiments consisting of measurement of the SERS spectra of buffer alone, Topo II alone, Intoplicine alone (1μM), DNA alone, Topo II+Intoplicine, and DNA+Intoplicine are performed under the same conditions, except that distilled water is used to adjust the reaction volume to 20μL\(^2\).

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

**Cell Assay \(^2\)**

The K562 human erythroleukemia cell line is established from a patient with chronic myelogenous leukemia. Cells are in the exponential growth phase at 5-8×10\(^5\) in RPMI 1640 (GIBCO) supplemented with 10% fetal calf serum (Seromed) and 2 mM L-glutamine. Cell growth and viability are determined by phase contrast microscopy and by using the trypan blue test. Cells (2×10\(^6\)) are incubated with 1 μM Intoplicine for 1 h at 37°C, washed twice with PBS by centrifugation (200× g at 4°C) and resuspended in 200 μL PBS\(^2\).

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

**REFERENCES**


---

**Caution: Product has not been fully validated for medical applications. For research use only.**

Tel: 609-228-6898                  Fax: 609-228-5909                  E-mail: tech@MedChemExpress.com
Address: 1 Deer Park Dr, Suite Q, Monmouth Junction, NJ 08852, USA

www.MedChemExpress.com