**Proteins** 

# **Product** Data Sheet

## ZM 336372

Cat. No.: HY-13343 CAS No.: 208260-29-1 Molecular Formula:  $C_{23}H_{23}N_3O_3$ Molecular Weight: 389.45

Target: Raf; Apoptosis

Pathway: MAPK/ERK Pathway; Apoptosis

Storage: Powder -20°C 3 years

 $4^{\circ}C$ 2 years

-80°C In solvent 2 years

> -20°C 1 year

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## **SOLVENT & SOLUBILITY**

DMSO: 50 mg/mL (128.39 mM; Need ultrasonic) In Vitro

H<sub>2</sub>O: < 0.1 mg/mL (insoluble)

Preparing Stock Solutions	Solvent Mass Concentration	1 mg	5 mg	10 mg
	1 mM	2.5677 mL	12.8386 mL	25.6772 mL
	5 mM	0.5135 mL	2.5677 mL	5.1354 mL
	10 mM	0.2568 mL	1.2839 mL	2.5677 mL

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 (6.42 mM); Clear solution
- 2. Add each solvent one by one: 10% DMSO >> 90% (20% SBE-β-CD in saline) Solubility: 2.5 mg/mL (6.42 mM); Suspended solution; Need ultrasonic
- 3. Add each solvent one by one: 10% DMSO >> 90% corn oil Solubility: ≥ 2.5 mg/mL (6.42 mM); Clear solution

## **BIOLOGICAL ACTIVITY**

Description ZM 336372 is a potent inhibitor of the protein kinase c-Raf. The IC $_{50}$  value is 0.07  $\mu$ M in the standard assay, which contains 0.1 mM ATP.

c-Raf

 $0.07 \, \mu M \, (IC_{50}, \, 0.1 \, mM \, ATP)$ 

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IC<sub>50</sub> & Target

#### In Vitro

ZM 336372 is a potent inhibitor of human c-Raf. The IC $_{50}$  value is 0.07  $\mu$ M in the standard assay, which contains 0.1 mM ATP. The IC $_{50}$ , decreases to 0.01 $\mu$ M at 0.025 mM ATP and increases to 0.9  $\mu$ M at 2.5 mM ATP indicating that ZM 336372 is a competitive inhibitor with respect to ATP. ZM 336372 inhibits c-Raf tenfold more potently than B-Raf $^{[1]}$ . Cell proliferation analysis of ZM336372. 3,4-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide growth assay of H727 and BON treated as control, DMSO, and 100  $\mu$ M ZM336372 to days 16 and 10, respectively. Both H727 and BON cell proliferation is inhibited in the presence of drug compared with controls. H727 cells treated with ZM336372 are growth suppressed, whereas control treatments have significantly more growth by day 6, continuing up to 16 days. A similar response is also seen in BON cells as growth suppression occurred as early as day 4 and was maintained out to day  $10^{[2]}$ . MCE has not independently confirmed the accuracy of these methods. They are for reference only.

### **PROTOCOL**

#### Cell Assay [2]

Proliferation of H727 and BON cells after treatment with ZM336372 is measured using a 3,4-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide. Cells are trypsinized and plated in triplicate to 24-well plates and allowed to adhere overnight. Then, cells are treated with either 100  $\mu$ M ZM336372 or DMSO (2%) and incubated. Media are changed every 2 days with new treatment. At each time point, cell growth rates are analyzed after the addition of 3,4-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide reagent to the cultured cells. Absorbance is determined using spectrophotometer at a wavelength of 540 nm<sup>[2]</sup>.

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

## **CUSTOMER VALIDATION**

• Cell Death Discov. 2023 Apr 28;9(1):139.

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#### **REFERENCES**

[1]. Hall-Jackson CA, et al. Paradoxical activation of Raf by a novel Raf inhibitor. Chem Biol. 1999 Aug;6(8):559-68.

[2]. Van Gompel JJ, et al. ZM336372, a Raf-1 activator, suppresses growth and neuroendocrine hormone levels in carcinoid tumor cells. Mol Cancer Ther. 2005 Jun;4(6):910-7.

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

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