



# SZABO SCANDIC

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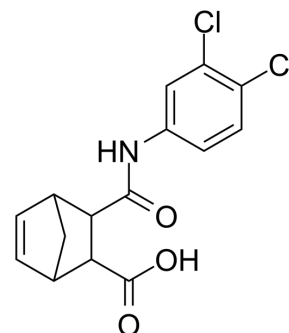
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## CADD522

Cat. No.:	HY-107999
CAS No.:	199735-88-1
Molecular Formula:	C <sub>15</sub> H <sub>13</sub> Cl <sub>2</sub> NO <sub>3</sub>
Molecular Weight:	326.17
Target:	Reactive Oxygen Species
Pathway:	Immunology/Inflammation; Metabolic Enzyme/Protease; NF-κB
Storage:	4°C, stored under nitrogen
	* In solvent : -80°C, 2 years; -20°C, 1 year (stored under nitrogen)



## SOLVENT & SOLUBILITY

In Vitro	DMSO : 250 mg/mL (766.47 mM; Need ultrasonic)					
	Preparing Stock Solutions	<div><div>Solvent</div><div>Concentration</div></div>	Mass	1 mg	5 mg	10 mg
		1 mM		3.0659 mL	15.3294 mL	30.6589 mL
		5 mM		0.6132 mL	3.0659 mL	6.1318 mL
		10 mM		0.3066 mL	1.5329 mL	3.0659 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: ≥ 2.17 mg/mL (6.65 mM); Clear solution					
	2. Add each solvent one by one: 10% DMSO >> 90% corn oil Solubility: ≥ 2.17 mg/mL (6.65 mM); Clear solution					

## BIOLOGICAL ACTIVITY

Description	CADD522 is a RUNX2-DNA binding inhibitor (downregulates RUNX2-mediated transcription of downstream target genes), with an IC <sub>50</sub> of 10 nM. CADD522 inhibits primary tumor growth and experimental metastasis of tumor cells in the lungs of immune-compromised mice. CADD522 can be used in study of cancer <sup>[1][2]</sup> .
IC <sub>50</sub> & Target	RUNX2 10 nM
In Vitro	CADD522 (0-100 μM; 24-72 h) exhibits a strong inhibitory effect on BC cell growth and survival <sup>[1]</sup> . CADD522 (50 μM; 72 h) shows anti-proliferative effect by inducing cell cycle arrest (G1 phase) <sup>[1]</sup> . CADD522 (50 μM; 8 days) inhibits tumorsphere formation and (50 μM; 24 h) in vitro invasion of BC cells (without cellular toxicity) <sup>[1]</sup> .

CADD522 (2, 10, 25, 50, 100  $\mu$ M; 48 h) inhibits RUNX2 transcriptional activity by inhibiting RUNX2-DNA binding in T47D-RUNX2 and T47D-Empty cells<sup>[1]</sup>.

CADD522 (50  $\mu$ M; 72 h) upregulates RUNX2 levels through increased RUNX2 stability in cells<sup>[1]</sup>.

CADD522 (50  $\mu$ M; 6 or 24 h) increases ROS generation of mitochondrial in MCF7 and MDA-468 cells<sup>[2]</sup>.

CADD522 (0-2000 nM, 30 min) inhibits mitochondrial ATP synthase activity in MDA-231 and MDA-468 cells<sup>[2]</sup>.

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

#### Cell Viability Assay<sup>[1]</sup>

Cell Line:	MDA-MB-468, MCF7, MCF10A, IEC-6, GES-1 and C2C12 cells
Concentration:	0-100 $\mu$ M
Incubation Time:	24-72 h
Result:	Displayed a dose- and time-dependent cell growth inhibition over 72 h. Exhibited low cytotoxicity for normal cell growth.

#### Cell Cycle Analysis<sup>[1]</sup>

Cell Line:	MCF7, MDA-468 and MDA-231 cells
Concentration:	50 $\mu$ M
Incubation Time:	72 h
Result:	Induced MDA-231 cells accumulated at the G1 and G2/M phase whereas MCF7 and MDA-468 cells were at the G1 phase.

#### Cell Viability Assay<sup>[1]</sup>

Cell Line:	MCF7, MCF7-tet-off cells
Concentration:	50 $\mu$ M
Incubation Time:	8 days
Result:	Dramatically decreased the size as well as the number of tumorspheres, and severely disrupted tumorspheres at day 4. Showed a relatively selective effect on BC cells (did not have a significant influence on mammosphere formation of the MCF10A non-malignant mammary epithelial cells).

#### Cell Invasion Assay<sup>[1]</sup>

Cell Line:	MCF7-tet-off (+Doxy), MCF7-tet-off (-Doxy) cells
Concentration:	50 $\mu$ M
Incubation Time:	24 h
Result:	Almost abrogated the invasiveness of both MCF7-tet-off (+Doxy) and MCF7-tet-off (-Doxy) cells without cellular toxicity.

#### Cell Viability Assay<sup>[1]</sup>

Cell Line:	T47D-RUNX2 and T47D-Empty cells
Concentration:	2, 10, 25, 50, 100 $\mu$ M
Incubation Time:	48 h

Result:	Resulted in a dramatic decrease of the promoter-luciferase (Luc) activities of RUNX2 downstream target genes such as MMP13 and VEGF (metastasis markers) and OC (osteogenesis marker).
RT-PCR <sup>[1]</sup>	
Cell Line:	T47D and MCF7 cells (ectopic expressing RUNX2)
Concentration:	50 $\mu$ M
Incubation Time:	72 h
Result:	Significantly inhibited the mRNA level (RUNX2-mediated) of Glut-1 and LDHA.
Western Blot Analysis <sup>[1]</sup>	
Cell Line:	T47D-RUNX2 and MCF7-RUNX2 cells
Concentration:	50 $\mu$ M
Incubation Time:	72 h
Result:	Enhanced both mRNA and protein expression of RUNX2.
Western Blot Analysis <sup>[1]</sup>	
Cell Line:	MDA-468 and MDA-231 cells
Concentration:	50 $\mu$ M
Incubation Time:	2, 4, 6 h
Result:	Increased RUNX2 stability by delaying protein degradation.
Cell Viability Assay <sup>[2]</sup>	
Cell Line:	MCF7 and MDA-468 cells
Concentration:	50 $\mu$ M
Incubation Time:	6 or 24 h
Result:	Increased the level of mitochondrial ROS, which was more evident in serum-free than serum-containing condition.
Cell Viability Assay <sup>[2]</sup>	
Cell Line:	MDA-231 and MDA-468 cells
Concentration:	50, 250, 2000 nM (for MDA-231); 500, 2000 nM (for MDA-468)
Incubation Time:	30 min
Result:	Inhibited the activity of A TP synthase.

#### In Vivo

CADD522 (1, 5 and 20 mg/kg; i.p.; twice a week for 45 days) delays the onset of the tumors and suppresses tumor growth in mice<sup>[1]</sup>.  
 CADD522 (10 mg/kg; i.p.; twice a week for 11 days) suppresses tumor metastasis and inhibits expression of Ki-67 in mice<sup>[1]</sup>.

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

Animal Model:	Female mice (6-week-old; MMTV-PyMT transgenic model) <sup>[1]</sup> .
Dosage:	1, 5 and 20 mg/kg
Administration:	Intraperitoneal injection; twice a week for 45 days.
Result:	Delayed the onset of the tumors, delayed tumor development and reduced tumor burden in transgenic MMTV-PyMT mice. Reduced the tumor weight in mice.
Animal Model:	Female NOD scid gamma (NSG) mice and nude mice (TNBC-PDX Br-001 model) <sup>[1]</sup> .
Dosage:	10 mg/kg
Administration:	Intraperitoneal injection; twice a week for 11 days.
Result:	Significant decreased tumor volume and markedly inhibited expression of Ki-67. Inhibited experimental metastasis of BC cells in vivo.(did not significantly decrease body weight or influence the general health of animals).

## CUSTOMER VALIDATION

- Nat Commun. 2022 Nov 4;13(1):6648.

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## REFERENCES

[1]. Kim MS, et al. Characterization of CADD522, a small molecule that inhibits RUNX2-DNA binding and exhibits antitumor activity. Oncotarget. 2017 Aug 10;8(41):70916-70940.

[2]. Kim MS, et al. Targeting breast cancer metabolism with a novel inhibitor of mitochondrial ATP synthesis. Oncotarget. 2020 Oct 27;11(43):3863-3885.

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

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