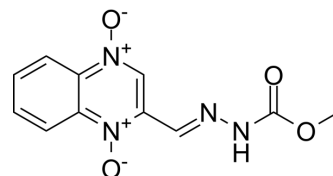


## Carbadox

<b>Cat. No.:</b>	HY-B1340		
<b>CAS No.:</b>	6804-07-5		
<b>Molecular Formula:</b>	C <sub>11</sub> H <sub>10</sub> N <sub>4</sub> O <sub>4</sub>		
<b>Molecular Weight:</b>	262.22		
<b>Target:</b>	Bacterial; Antibiotic; Endogenous Metabolite		
<b>Pathway:</b>	Anti-infection; Metabolic Enzyme/Protease		
<b>Storage:</b>	Powder	-20°C	3 years
		4°C	2 years
	In solvent	-80°C	2 years
		-20°C	1 year



### SOLVENT & SOLUBILITY

#### In Vitro

DMSO : 3.57 mg/mL (13.61 mM; Need ultrasonic)  
 H<sub>2</sub>O : < 0.1 mg/mL (insoluble)

Preparing Stock Solutions	Solvent		Mass		
	Concentration		1 mg	5 mg	10 mg
	1 mM		3.8136 mL	19.0680 mL	38.1359 mL
	5 mM		0.7627 mL	3.8136 mL	7.6272 mL
	10 mM		0.3814 mL	1.9068 mL	3.8136 mL

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

### BIOLOGICAL ACTIVITY

#### Description

Carbadox is a quinoxaline-di-N-oxide antibiotic compound which is widely fed to nursery-age pigs to control enteric diseases and improve feed efficiency.

#### IC<sub>50</sub> & Target

Bacterial<sup>[1]</sup>

#### In Vitro

The results of MTT assay demonstrate a dose-dependent decrease in mitochondrial activity in Vero cells at all concentrations of Carbadox. Treatment with Carbadox at the highest concentration of 160 µg/mL results in cell viability down to only 12%. Cells following Carbadox treatment show a dose-dependent increase of the DNA migration (p<0.01). The nuclear division index (NDI) reduces markedly with the increase doses of Carbadox<sup>[1]</sup>.

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

#### In Vivo

Alpha diversities (Shannon diversity, Heips evenness, and inverse Simpson indices) of samples from medicated piglets compare to non-medicated piglets are significantly different at 2, 3, and 4 days after continuous Carbadox, but not different in either late Carbadox or at any time during the withdrawal period. Analysis of the community structure of bacteria in

animals shows significant differences at days 3 and 4 of early Carbadox treatment ( $[R=0.32, p=0.015]$  and  $[R=0.54, p=0.003]$ , respectively), but not before starting antibiotic treatment ( $p=0.82$ ). No significant differences in *E. coli* colony forming units (CFUs) are observed during the Carbadox-treatment period of the study or late in the withdrawal period. *E. coli* CFUs are significantly different between the medicated and non-medicated groups on day 2 after the withdrawal of Carbadox<sup>[2]</sup>. MCE has not independently confirmed the accuracy of these methods. They are for reference only.

## PROTOCOL

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

Exponentially growing Vero cells are seeded at  $10^4$  cells/well density in 96 microplates and exposed to various concentrations of Carbadox (5, 10, 20, 40, 80, 160, 210, 260, 310 and 360  $\mu\text{g}/\text{mL}$ ). Cells incubated with the same concentration DMSO are used as a control. After 4 h or 24 h, each well is added 100  $\mu\text{L}$  MTT solution (200  $\mu\text{g}/\text{mL}$ ) followed incubation for 4 h at 37 °C, and the medium containing MTT is removed. The formazan crystals in the viable cells are solubilized with 100  $\mu\text{L}$  DMSO and the absorbance at 570 nm of each well is read using a microplate reader. All experiments are performed at least 3 times, with 6 wells for each concentration of Carbadox ( $n=6$  per experiment). Final results are the average of three independent experiments. The cell viability is calculated as follows:  $\text{OD of experimental group}/(\text{OD of control group}-\text{OD of blank group})\times 100\%$ . The data are presented as  $\text{means}\pm\text{SE}$ <sup>[1]</sup>.

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### Animal Administration <sup>[2]</sup>

At 3 weeks of age, 12 piglets from 2 litters are divided into two rooms of six pigs each, with equal representation of littermates and gender. All pigs are fed a standard starter diet ad libitum for 3 weeks, after which six control pigs continue to receive non-medicated feed while the other group receives feed containing Carbadox (50 g/ton). After 21 days of continuous feed with or without Carbadox, all pigs (60 days old) are switched to a non-medicated maintenance diet. Feces are collected from each pig at multiple times before, during, and after antibiotic withdrawal<sup>[2]</sup>.

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

## REFERENCES

[1]. Chen Q, et al. Investigation of the genotoxicity of quinocetone, carbadox and olaquinox in vitro using Vero cells. *Food Chem Toxicol.* 2009 Feb;47(2):328-34.

[2]. Looft T, et al. Carbadox has both temporary and lasting effects on the swine gut microbiota. *Front Microbiol.* 2014 Jun 10;5:276.

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

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