# Data Sheet (Cat.No.T0740)



# Metformin hydrochloride

# **Chemical Properties**

CAS No.: 1115-70-4

Formula: C4H12ClN5

Molecular Weight: 165.63

Appearance: no data available

Storage: Powder: -20°C for 3 years | In solvent: -80°C for 1 year

# H<sub>3</sub>C N NH NH

# **Biological Description**

Description	Metformin hydrochloride (1,1-Dimethylbiguanide hydrochloride), a widely used anti- diabetic drug, exhibits potential anti-Y properties by inhibiting the proliferation of various Y cells, including colon and prostate.		
Targets(IC50)	Mitophagy,AMPK,Autophagy		
In vitro	METHODS: Ovarian cancer cells A2780 and SKOV3 were treated with Metformin hydrochloride (0.001-50 mM) for 24-48 h. Cell viability was assayed using the MTS RESULTS: Micromolar concentrations of Metformin did not statistically reduce the viability of the A2780 or SKOV3 cell lines. At 48 h, millimolar concentrations resulted in cell death. [1]  METHODS: Human colorectal cancer cells HCT29 were treated with Metformin hydrochloride (0.6 mM) for 90 h. Cell motility was detected using the wound healing assay and chamber invasion assay.  RESULTS: Metformin inhibited the migration and invasion of HCT29 cells, and Metformin decreased the motility of tumor cells. [2]		
In vivo	METHODS: To model Metformin-induced diarrhea, Metformin hydrochloride (125-500 mg/kg) was administered orally to healthy and diabetic obese db/db C57BL/6J mice twice daily for thirteen days.  RESULTS: Metformin at 1000 mg/kg/day significantly increased fecal water content.  Although no diarrhea symptoms were observed in healthy C57BL/6J mice, the same dose of Metformin induced severe diarrhea in diabetic obese db/db mice. [3]  METHODS: To investigate the protective effect of Metformin in radiation injury, Metformin hydrochloride (200 mg/kg once daily for three days) was administered orally to BALB/c mice, which were then exposed to 6-8 Gy of gamma radiation.  RESULTS: When administered prior to exposure to radiation, Metformin prolonged the survival of mice exposed to 8 Gy-TBI and increased the survival of mice exposed to 6 Gy-TBI. Pretreatment with Metformin attenuated radiation damage. [4]		
Cell Research	Hepatocytes were isolated from male Sprague Dawley (SD) rats by collagenase digestion. For the AMPK assay, cells were seeded in six-well plates at $1.5 \times 10^6$ cells/well in DMEM containing 100 U/ml penicillin, 100 $\mu$ g/ml streptomycin, 10% FBS, 100 nM insulin, 100 nM dexamethasone, and 5 $\mu$ g/ml transferrin for 4 hours. Cells were then cultured in serum-free DMEM for 16 hours followed by treatment for 1 hour or 7 hours with control medium, 5-aminoimidazole carboxamide riboside (AICAR), or metformin at concentrations indicated. For a 39-hour treatment, cells for both control		

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and metformin (10 or 20  $\mu$ M) groups were cultured in DMEM plus 5% FBS and 100 nM insulin, and the fresh control and metformin-containing medium were replaced every 12 hours (last medium change was 3 hours before harvest). After treatment, the cells were directly lysed in digitonin-containing and phosphatase inhibitor-containing buffer A, followed by precipitation with ammonium sulfate at 35% saturation. AMPK activity was determined by measurement of phosphorylation of a synthetic peptide substrate, SAMS (HMRSAMSGLHLVKRR). For ACC assay, the 35% ammonium sulfate precipitate from digitonin-lysed hepatocytes (4  $\mu$ g each) was used for determination of ACC activity via 14CO2 fixation in the presence of 20 mM citrate as done previously. For fatty acid oxidation, the oxidation of 14C-oleate to acid-soluble products was performed as done previously, but in medium M199 in the absence of albumin [1].

#### Animal Research

Oral gavage was used to administer 1 ml of metformin (100 mg/ml) or water alone to male SD rats (300-350 g, n = 7-8). Rats were treated once or twice a day for 5 days. Rats were starved for 20 hours and then re-fed for 2 hours before the final dose; 4 hours after the final dose, the animals were anesthetized and livers rapidly removed by freeze clamping followed by blood withdrawal. RNA was prepared from the freeze-clamped liver by RNA isolation reagent. Nuclear extracts were prepared from a pool of seven rat livers. Glucose levels were determined using the standard glucose oxidase assay kit;  $\beta$ hydroxybutyrate concentrations were assayed by measuring the reduction of NAD to NADH with a standard assay kit. FFA levels were measured with the assay kit [1]. MCF10A-ER-Src cells (5 × 10^6) were injected into the right flank of 18 female nu/nu mice, all of which developed tumors in 10 d with a size of ~ 100 mm^3. The mice were randomly distributed into six groups (three mice/group) that were untreated or treated by intratumoral injections every 5 d (four cycles) with 1 mg/kg or 4 mg/kg doxorubicin, 200 µg/mL metformin (diluted in the drinking water), or the combination. In another experiment, LNCaP and DU145 prostate cancer cells (5 x 10^6) were injected into the right flank of 12 female nu/nu mice, all of which developed tumors in 10 d with a size of ~75 mm^3. The mice were randomly distributed into four groups that were untreated or treated by intratumoral injections every 5 d (four cycles) with 4 mg/kg doxorubicin and/or 200 µg/mL metformin. In another experiment, A375 and MDA-MB-435 melanoma cells  $(7 \times 10^6)$  were injected into the right flank of 12 female nu/nu mice, all of which developed tumors in 10 d with a size of ~50 mm3. The mice were randomly distributed into four groups that were untreated or treated by intratumoral injections every 5 d (four cycles) with 10 mg/kg cisplatin and/or 200 µg/mL metformin. Finally, SNU-449 liver cancer cells (10^7) were injected into the right flank of 12 female nu/nu mice, all of which developed tumors in 10 d with a size of ~50 mm^3. The mice were randomly distributed into four groups that were untreated or treated by intratumoral injections every 5 d (four cycles) with 10 mg/kg cisplatin and/or 200 µg/mL metformin. Tumor volume (mean ± SD) was measured at various times after the initial injection [3].

### **Solubility Information**

Solubility H2O: 193.21 mM, Sonication is recommended.

DMSO: 15 mg/mL (90.56 mM), Sonication is recommended. (< 1 mg/ml refers to the product slightly soluble or insoluble)

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## **Preparing Stock Solutions**

	1mg	5mg	10mg
1 mM	6.0376 mL	30.1878 mL	60.3755 mL
5 mM	1.2075 mL	6.0376 mL	12.0751 mL
10 mM	0.6038 mL	3.0188 mL	6.0376 mL
50 mM	0.1208 mL	0.6038 mL	1.2075 mL

Please select the appropriate solvent to prepare the stock solution, according to the solubility of the product in different solvents. Please use it as soon as possible.

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