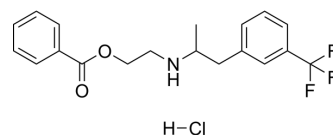


Benfluorex hydrochloride

Cat. No.:	HY-B1058
CAS No.:	23642-66-2
Molecular Formula:	C ₁₉ H ₂₁ ClF ₃ NO ₂
Molecular Weight:	387.82
Target:	Others
Pathway:	Others
Storage:	4°C, sealed storage, away from moisture * In solvent : -80°C, 6 months; -20°C, 1 month (sealed storage, away from moisture)



SOLVENT & SOLUBILITY

In Vitro

DMSO : ≥ 100 mg/mL (257.85 mM)
 H₂O : 2.27 mg/mL (5.85 mM); ultrasonic and warming and heat to 60°C
 * "≥" means soluble, but saturation unknown.

Preparing Stock Solutions	Solvent Concentration	Mass		
		1 mg	5 mg	10 mg
	1 mM	2.5785 mL	12.8926 mL	25.7852 mL
	5 mM	0.5157 mL	2.5785 mL	5.1570 mL
	10 mM	0.2579 mL	1.2893 mL	2.5785 mL

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

In Vivo

- Add each solvent one by one: PBS
Solubility: 5.88 mg/mL (15.16 mM); Clear solution; Need ultrasonic and warming and heat to 60°C
- Add each solvent one by one: 10% DMSO >> 40% PEG300 >> 5% Tween-80 >> 45% saline
Solubility: ≥ 2.5 mg/mL (6.45 mM); Clear solution
- Add each solvent one by one: 10% DMSO >> 90% (20% SBE-β-CD in saline)
Solubility: ≥ 2.5 mg/mL (6.45 mM); Clear solution
- Add each solvent one by one: 10% DMSO >> 90% corn oil
Solubility: ≥ 2.5 mg/mL (6.45 mM); Clear solution

BIOLOGICAL ACTIVITY

Description	Benfluorex hydrochloride (JP-992 hydrochloride) is a hepatic nuclear factor 4 alpha (HNF4α) activator.
IC₅₀ & Target	HNF4α ^[1]
In Vitro	Benfluorex hydrochloride consistently activates insulin promoter activity as measured by an increased number of GFP-

positive cells. Benfluorex hydrochloride increases the number of GFP-positive cells in a dose-responsive manner and increases the level of endogenous insulin mRNA. Consistent with being HNF4 α activator, Benfluorex hydrochloride stimulates HNF4 α expression. Benfluorex hydrochloride alters HNF4 α protease sensitivity, while the inactive control compound does not^[1]. Benfluorex hydrochloride decreases, in a concentration-dependent manner, the synthesis of acid-soluble products and ketone bodies from oleate, whereas the production of ¹⁴CO₂ into citric acid cycle is markedly increased by Benfluorex hydrochloride. Benfluorex hydrochloride inhibits in a dose-dependent manner the rates of gluconeogenesis from lactate/pyruvate (10/1 nM)^[2].

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

PROTOCOL

Kinase Assay ^[1]

HepG2 cells are treated with DMSO or Benfluorex hydrochloride at a concentration of 20 μ M or 40 μ M for 16 hr. Total cell protein is extracted, measured by BCA protein assay. Each sample is split into two aliquots for proteolysis without (-) or with (+) Subtilisin. Twenty μ g of cell lysate is incubated with or without protease (20 ng/mL subtilisin) for 35 minutes at room temperature. Western blot is then performed with primary anti-HNF4 α polyclonal antibody (1:1000 dilution) and secondary HRP conjugated anti-goat IgG (1:2000 dilution), detected with chemiluminescence ECL kit^[1].

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

Cell Assay ^[2]

Hepatocytes are isolated by in situ perfusion of the liver with 0.025% collagenase. Hepatocytes (1 to 2 \times 10⁶ cells/mL) are incubated at 37°C in 2 mL of oxygenated (O₂:CO₂; 95:5) Krebs-Henseleit bicarbonate buffer (pH 7.4) for 1 h in a gyratory shaking water bath. Benfluorex hydrochloride is dissolved in DMSO and added (10 μ L) to the incubation medium at a final concentration of 0.1 or 1 nM^[2].

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

CUSTOMER VALIDATION

- EBioMedicine. 2022 Jul 28;82:104181.
- Biochem Pharmacol. 6 August 2022, 115198.
- Virol J. 2021 Sep 28;18(1):196.
- Research Square Print. December 19th, 2022.

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REFERENCES

[1]. Lee SH, et al. Identification of alverine and benfluorex as HNF4 α activators. ACS Chem Biol. 2013 Aug 16;8(8):1730-6.

[2]. Kohl C, et al. Effects of benfluorex on fatty acid and glucose metabolism in isolated rat hepatocytes: from metabolic fluxes to gene expression. Diabetes. 2002 Aug;51(8):2363-8.

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

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