opnMe by Boehringer Ingelheim

Cathepsin C Substrate

BI-1750

Boehringer Ingelheim

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Summary

BI-1750 is a stable and highly selective intracellular substrate for the human protease Cathepsin C (CatC).

Chemical Structure

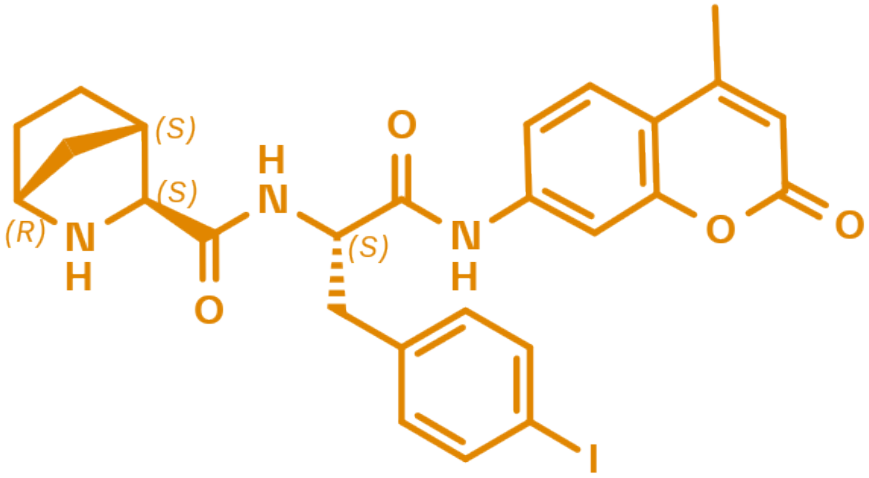

Figure 1: 2D structure of BI-1750

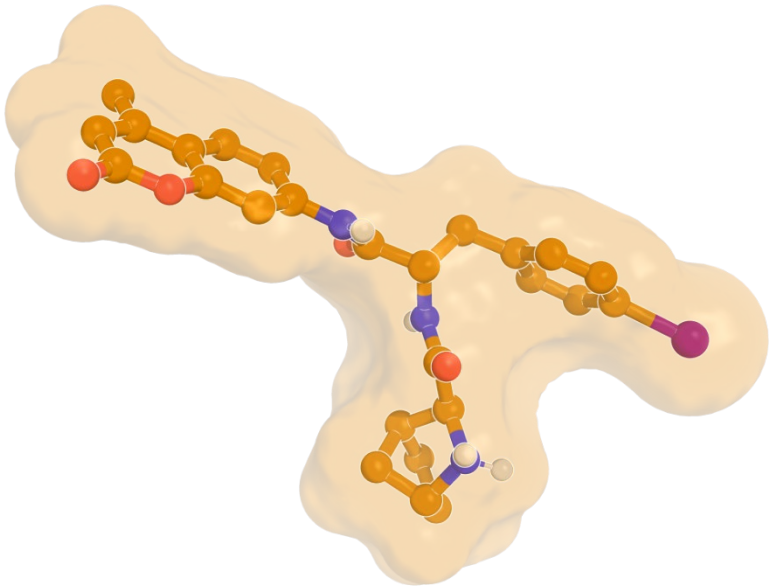

Figure 2: BI-1750, 3D conformation

Highlights

BI-1750 is a novel fluorophore substrate for the human protease Cathepsin C (CatC), which enzymatically cleaves BI-1750 with a Michaelis-Menten kinetic. It is stable and highly selective, with no conversion by the related enzymes CatB, CatF, CatH, CatK, CatL and CatS observed. BI-1750 is cell permeable and may be used to monitor intracellular CatC activity in *ex vivo* human whole blood assays and other cellular systems, including activity in cells from rodents.

Target information

Cathepsin C (CatC) is a lysosomal cysteine protease. It is expressed at high levels in lung, kidney, and placenta and at moderate to low levels in many other organs. Among immune/inflammatory cells, the mRNA is expressed at high levels in polymorphonuclear leukocytes and alveolar macrophages and their precursor cells[1].
In the bone marrow, CatC activates neutrophil serine proteases (NSPs) during myelopoiesis of neutrophils. Inhibition of CatC leads to a decrease in neutrophil elastase (NE), cathepsin G (CG), proteinase 3 (PR3) and NSP4 activities in circulating neutrophils[2].

In vitro activity

BI-1750 behaves like a substrate for Cathepsin C and all assays proving this behavior are given below with different assay conditions.

PROBE NAME	BI-1750
MW [Da, free base][a]	571.4
CatC (IC_{50}) [nM][b]	120,000

[a] For the salt form you will get, please refer to the label on the vial and for the molecular weight of the salt, please refer to the FAQs
[b] Assay conditions for CatC assay are available in the patent WO2014140075. More detailed experimental conditions can always be obtained via the "Contact us" formular.

At 50 µM, BI-1750 is enzymatically cleaved by CatC similar to the standard substrate Gly-Arg-AMC:

SUBSTRATE	VMAX [RFU/SEC]
50 µM Gly-Arg-AMC	29
50 µM BI-1750	22

BI-1750 is converted by isolated primary human neutrophils (PMN) depending on cell-number:

PMN (*1E5)	SUBSTRATE TURNOVER [RFU/30 MIN]
19.4	5,045
9.7	2,763
4.85	1,526
2.43	769
1.21	408
0.61	179
0.3	69

Turnover of BI-1750 in human whole blood: (40 µM BI-1750, 30 min incubation at 37°C)

PLASMA CONTROL [RFU, N=10]	WHOLE BLOOD [RFU, N=10]
750 +/- 32	6,449 +/- 171

In vitro DMPK and CMC parameters

No data available, this tool can be used to monitor intracellular CatC activity in human whole blood assays and other cellular systems but in *in vivo* assays.

In vivo DMPK parameters

No data available, this tool can be used to monitor intracellular CatC activity in human whole blood assays and other cellular systems but in *in vivo* assays.

Selectivity

BI-1750 is not converted by the related enzymes CatB, CatF, CatH, CatK, CatL and CatS.

ENZYME	ENZYME SPECIFIC SUBSTRATE		BI-1750
	SUBSTRATE	TURNOVER [RFU/MIN]	TURNOVER [RFU/MIN]
CatB	Z-Arg-Arg-AMC	80	0
CatF	Z-Leu-Arg-AMC	26	0
CatH	H-Arg-AMC	86	0
CatL	Z-Phe-Arg-AMC	114	0

CatK	Z-GPR-AMC	36	0
CatS	Z-Val-Val-Arg-AMC	69	0.1

SELECTIVITY DATA AVAILABLE	BI-1750
SafetyScreen44™ with kind support of eurofins	not applicable
Invitrogen®	No
DiscoverX®	No
Dundee	No

Reference molecule(s) – Inhibitors

Daniel Guay, Christian Beaulieu and David M. Percival Therapeutic Utility and Medicinal Chemistry of Cathepsin C Inhibitors *Current Topics in Medicinal Chemistry* **2010**, *10*, 2010, 708-716 DOI: 10.2174/156802610791113469, PubMed.

Supplementary data

Selectivity data can be downloaded free of charge from opnMe.

References

1. Rao N. V., Rao G. V., Hoidal J. R. Human Dipeptidyl-peptidase I Gene characterization, localization and expression *J Bio Chem.* **1997**, *272*, 10260-10265. DOI: 10.1074/jbc.272.15.10260, PubMed.

2. Korkmaz B., Horwitz M. S., Jenne D. E., Gauthier F. Neutrophil Elastase, Proteinase 3, and Cathepsin G as Therapeutic Targets in Human Diseases *Pharmacol Rev.* **2010**, *62*, 726-759. DOI: 10.1124/pr.110.002733, PubMed.