# DPAP1 Inhibitor | BI-2051

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## Summary

BI-2051 is very potent and highly selective dipeptidyl aminopeptidase 1 (DPAP1) inhibitor. BI-2051 inhibits recombinant *P. falciparum* DPAP1 with an $IC_{50}$ of 0.3 nM. BI-2051 is highly soluble at pH 2.2, 4.5, 7 and displays good *in vitro* PK properties in rat.

## Chemical Structure

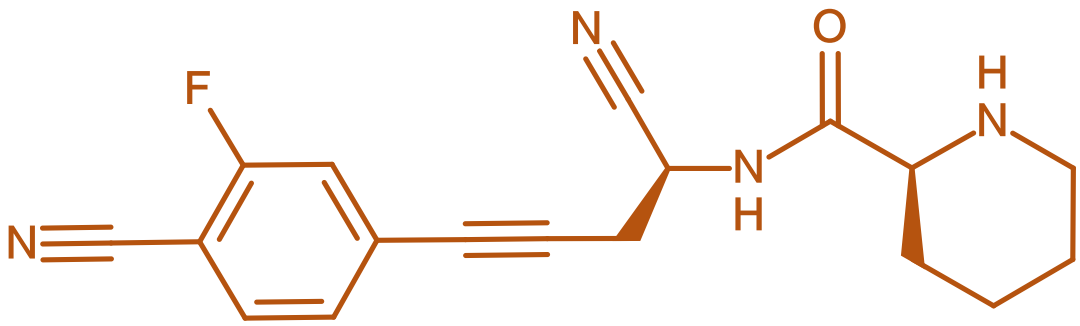


**Figure 1: 2-D structure of BI-2051**

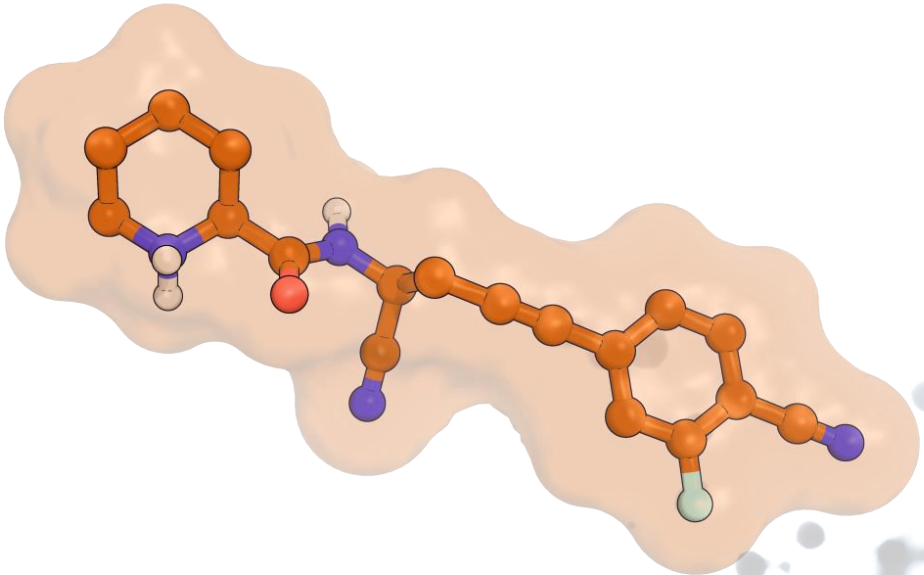

**Figure 2: BI-2051, 3D conformation**

# Highlights

BI-2051 is a highly selective, soluble and cellular permeable inhibitor of the *P. falciparum* protease dipeptidyl aminopeptidase 1 (DPAP 1).

# Target information

Dipeptidyl aminopeptidase 1 (DPAP1) is a cysteine exopeptidase expressed in the food vacuoles of the parasite *Plasmodium falciparum*[1].
*P. falciparum* uses the hemoglobin in the host erythrocytes as a source of amino acids and this catabolism is essential for the intraerythrocytic growth of the parasite. Hemoglobin is taken up by the cytostome and delivered into the food vacuole. In the food vacuole, hemoglobin is degraded by aspartic, cysteine and metalloproteases. DPAP1 catalyzes the final step, the release of small peptides or amino acids from globin-derived oligopeptides[1]. DPAP1 is therefore a potential target to interfere with the growth of *P. falciparum* during the erythrocytic phase of its life cycle[2].
DPAP1 shows significant sequence homology to the human protease Cathepsin C (CatC).

# *In vitro* activity

BI-2051 displays an $IC_{50}$ = 0.3 nM in a DPAP1 assay using recombinant protein.

| PROBE NAME | BI-2051 |
|---|---|
| MW [Da] | 324 |
| DPAP1 ($IC_{50}$) [nM][a] | 0.3 |
| Human CatC ($IC_{50}$) [nM] | 2.691 |
| Human CatK ($IC_{50}$) [nM] | 4.301 |
| Human CatL ($IC_{50}$) [nM] | > 100.000 |

[a]Assay conditions for the CatC assay are available in the patent WO2014140075. For DPAP1, CatK, and CatL the assay conditions are identical except for the enzyme nature, concentration, buffer and

substrates. More detailed experimental conditions can always be obtained via the "Contact us" formular.

For DPAP1, the substrate is H-Pro-Arg-AMC
For CatC, the substrate is Gly-Arg-AMC
For CatL, the substrate is Z-Phe-Arg-AMC
For CatK, the substrate is Z-Gly-Pro-Arg-AMC

## *In vitro* DMPK and CMC parameters

BI-2051 is a highly soluble and permeable compound. It has good *in vitro* PK properties in rats, but displays a weaker microsomal stability in mice.

| PROBE NAME | BI-2051 |
|---|---|
| logP | 2.6 |
| Solubility @ pH 6.8 [µg/ml] | >70 |
| CACO permeability @ pH 7.4 [*$10^{-6}$ cm/s] | 53.7 |
| CACO efflux ratio | 0.45 |
| MDCK permeability $P_{app}$a-b/b-a @ 1µM [$10^{-6}$ cm/s] | n.d. |
| MDCK efflux ratio | n.d. |
| Microsomal stability (human/mouse/rat) [% $Q_H$] | <23 \| 40 \| 29 |
| Hepatocyte stability (human/mouse/rat) [% $Q_H$] | 25 \| n.d. \| n.d. |
| Plasma protein binding (human/mouse/rat) [%] | n.d. |
| hERG [inh. % @ 10 µM] | 59 |
| CYP 3A4 ($IC_{50}$) [µM] | n.d. |

| | |
|---|---|
| CYP 2C8 ($IC_{50}$) [µM] | n.d. |
| CYP 2C9 ($IC_{50}$) [µM] | n.d. |
| CYP 2C19 ($IC_{50}$) [µM] | n.d. |
| CYP 2D6 ($IC_{50}$) [µM] | n.d. |

## *In vivo* pharmacology

No *in vivo* data available.

## Selectivity

BI-2051 is > 8000x selective for *P. falciparum* DPAP1 versus the homologous human enzymes CatC, CatK and CatL.

| BI-2051 | SELECTIVITY DATA AVAILABLE |
|---|---|
| Cerep® | No |
| Panlabs® | No |
| Invitrogen® | No |
| DiscoverX® | No |
| Dundee | No |

## Co-crystal structure of the Boehringer Ingelheim probe compound and the target protein.

No co-crystal structure available.

## Reference molecule(s)

No reference molecules available

## Summary

BI-2051 is a very potent inhibitor of the *P. falciparum* protease DPAP1. It inhibits recombinant DPAP1 with an $IC_{50}$ of 0.3 nM and is highly selective versus the homologous human proteases CatC, CatK and CatL (all $IC_{50}$ > 2500 nM). BI-2051 is highly soluble at pH 2.2, 4.5, 7 has a good cellular permeability and displays good *in vitro* PK properties in rats. Microsomal stability in mice is weaker than in human and rats.

## References

1. Michael Klemba, Ilya Gluzman and Daniel E. Goldberg A Plasmodium falciparum dipeptidyl aminopeptidase I participates in vacuolar hemoglobin degradation *Journal of Biological Chemistry*, **2004** *279,*43000-43007. Epub 2004 Aug 10. DOI: 10.1074/jbc.M408123200, PubMed

2. Edgar Deu, Melissa J. Leyva, Victoria E. Albrow, Mark J. Rice, Jonathan A. Ellman, and Matthew Bogyo Functional studies of Plasmodium falciparum dipeptidyl aminopeptidase I using small molecule inhibitors and active site probes *Chemical & Biology* **2010,** *27*, 808-819. DOI:10.1016/j.chembiol.2010.06.007, PubMed