opnMe by Boehringer Ingelheim

Nav1.2 channel blocker (crobenetine)

BIII 890CL

Boehringer Ingelheim

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Summary

BIII 890CL (crobenetine) is a highly potent and selective Nav1.2 sodium channel blocker that can be used as tool compound to test biological hypotheses *in vitro* and *in vivo*.

Chemical Structure

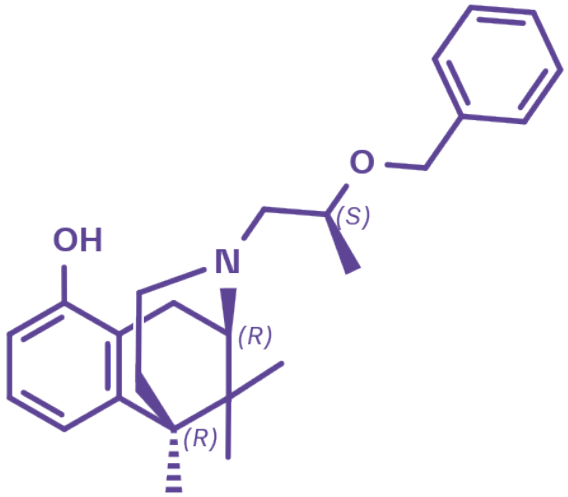

Figure 1: 2D structure of BIII 890CL (crobenetine)

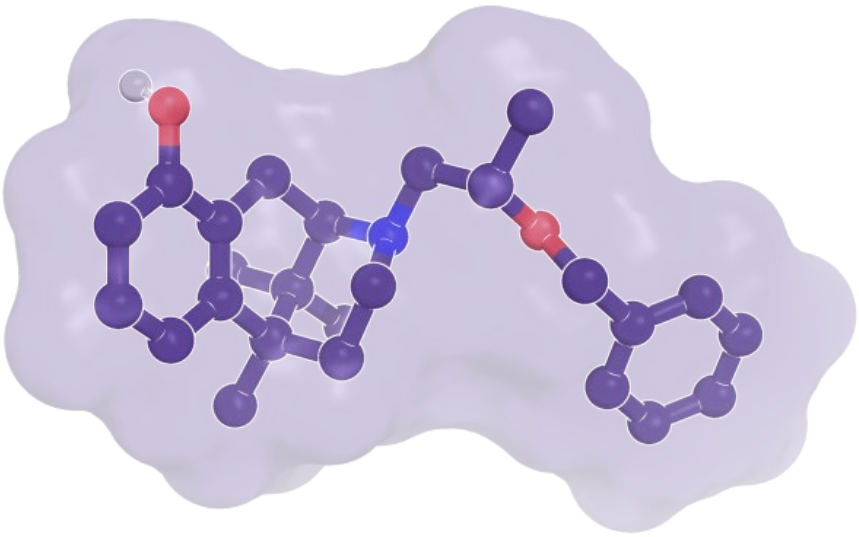

Figure 2: BIII 890CL (crobenetine), 3D conformation

Highlights

BII 890CL is a highly potent, use- and voltage-dependent Nav1.2 sodium channel blocker. It has high selectivity for site 2 of the sodium channel and preferentially binds to its inactive state (IC_{50} = 77 nM), which makes it an excellent tool to discriminate between highly activated neurons and neurons with physiological membrane potentials. This compound is suitable for both *in vitro* and *in vivo* experiments.

Target information

The voltage-gated sodium (Nav) channels are responsible for the rapid rising phase of an action potential, and thus play an essential role in membrane excitability and electrical signalling. Three distinct functional states are known: resting, active, and inactivated and all play a key role in neuronal activation. They are highly selective for the transport of sodium ions across cell membranes and become activated by a change in transmembrane voltage which is initially negatively charged. The activation results in a sodium influx and further depolarization of the neuron as the cause for an action potential. At the peak of the action potential, the sodium channels inactivate themselves by closing their inactivation gate. The neuron has to repolarize to its resting potential to bring the sodium channel back into the resting state[1,2,4].
Sodium channels play a major role in signal propagation within the central nervous system (CNS) but also in cardiac myocytes. Mutations that interfere with Na+ channel inactivation can contribute to cardiovascular diseases or epileptic seizures by over-excitation of muscle or nerve cells[1,2,4].
Based on patch clamp evaluations, BII 890CL preferentially binds to the inactivated state of the sodium channel (IC_{50} = 77 nM). In contrast, the binding at the resting state is only 18 µM. This exceptional selectivity of more than 230-fold for the different states of the sodium channel makes this compound unique compared with other sodium channel blockers[1-4].

Figure 3: Cryo-EM structure of Nav1.2 in complex with cynotoxin KIIIA (X. Pan et al.)[5].

In vitro activity

PROBE NAME / NEGATIVE CONTROL	BIII 890CL	BI-55CL
MW [Da, free base][a]	379.5	303.4
Displacement of [^{3}H] BTX K_i [nM] BTX: batrachotoxin[b]	43	>10,000
Inhibition of veratridine induced glutamate release in rat brain slices IC_{50} [nM][b]	322	n.a.

[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] For detailed assay conditions see reference 3

In vitro DMPK and CMC parameters

PROBE NAME / NEGATIVE CONTROL	BIII 890CL	BI-55CL
Melting point (°C)	258	n.a.
logD @ pH 11	>6	4.9
Solubility @ pH 4 / pH 6 / pH 7 [µg/mL]	2,560 / 1,040 / 8	n.a. / n.a. / >70
Caco-2 permeability AB @ pH 7.4 [*10^{-6} cm/s]	n.a.	44
Caco-2 efflux ratio	n.a.	0.7
Microsomal stability (human, mouse, rat) [% Q_H]	87 / n.a. / n.a.	62 / >88 / >88
Hepatocyte stability (human / mouse / rat) [% Q_H]	n.a.	n.a.
Plasma Protein Binding (rat) [%]	99.2	n.a.
hERG (IC50) [µM]	1.3	n.a.
CYP 3A4 (IC_{50}) [µM]	11.4	>50
CYP 1A2 (IC_{50}) [µM]	25.5	n.a.
CYP 2C9 (IC_{50}) [µM]	6.9	>50
CYP 2C19 (IC_{50}) [µM]	2.3	3.6
CYP 2D6 (IC_{50}) [µM]	0.03	>50

In vivo DMPK parameters

BIII 890CL	RAT[A]
t_{max} [h]	2.83
C_{max} [nM]	11.9
AUC [nM]	24

[a] *p.o.* dose: 3 mg/kg

In vivo pharmacology

In animal studies such as the maximum electrical shock model it could be demonstrated that the compound supresses tonic seizures at doses which do not interfere with physiological functions of the sodium channel.

Maximum electroshock (MES) test in mice ID_{50} = 6.1 mg/kg[3].

Negative control

With BIII 890CL, we offer a structurally close analogue, BI-55CL, with more than 1,000-fold lower potency for site 2 of the sodium channel (K_i ~ 10 µM; [^{3}H]-BTX). Although the selectivity between the two states of the sodium channel has not been fully determined, the compound can be used as a comparator compound in *in vitro* studies[1-4].

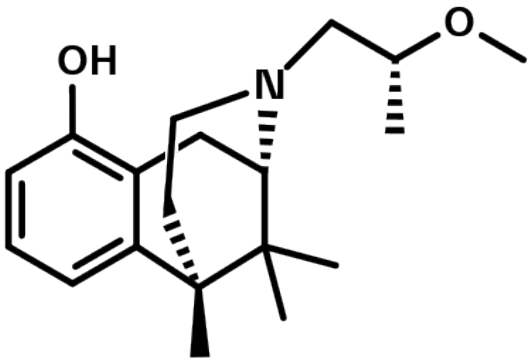

Figure 4: BI-55CL which serves as a negative control

Selectivity

SELECTIVITY DATA AVAILABLE	BIII 890CL	BI-55CL
SafetyScreen44™ with kind support of eurofins	Yes	Yes
Invitrogen®	No	No
DiscoverX®	No	No
Dundee	No	No

Reference molecule(s)

For a review on voltage-gated sodium channel blockers please see reference 6.

Supplementary data

2D structure files can be downloaded free of charge from opnMe.

References

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3. Grauert M., Bechtel W. D., Weiser T., Stransky W., Nar H., Carter A. J. Synthesis and structure-activity relationships of 6,7-benzomorphan derivatives as use-dependent sodium channel blockers for the treatment of stroke *J Med Chem* **2002**, 45(17), 3755–3764. DOI: 10.1021/jm020875j, PubMed: 12166948.

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5. Pan X., Li Z., Huang X., Huang G., Gao S., Shen H., Liu L., Lei J., Yan N. Molecular basis for pore blockade of human Na+ channel Nav1.2 by the μ-conotoxin KIIIA *Science* **2019**, 363(6433), 1309–1313. DOI: 10.1126/science.aaw2999, PubMed: 30765605.

6. Wood J. N., Boorman J. Voltage-gated sodium channel blockers; target validation and therapeutic potential *Curr Top Med Chem* **2005**, 5(6), 529–537. DOI: 10.2174/1568026054367584, PubMed: 16022675.