Bioorganic & Medicinal Chemistry Letters 27 (2017) 5326–5331

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry Letters

journal homepage: www.elsevier.com/locate/bmcl

Discovery and evaluation of $Ca_v3.2$-selective T-type calcium channel blockers

Olivier Bezençon *, Luboš Remeň, Sylvia Richard, Catherine Roch, Melanie Kessler, Eric A. Ertel, Richard Moon, Jacques Mawet, Thomas Pfeifer, Bruno Capeleto

Drug Discovery Chemistry, Biology and Pharmacology, Idorsia Pharmaceuticals Ltd., Hegenheimermattweg 91, CH-4123 Allschwil, Switzerland

ARTICLE INFO

Article history:
Received 14 August 2017
Revised 27 September 2017
Accepted 29 September 2017
Available online 30 September 2017

Keywords:
T-type calcium channel
Selective Cav3.2-blockers
Pyrroles
Absence-type epilepsy

ABSTRACT

We identified and characterized a series of pyrrole amides as potent, selective $Ca_v3.2$-blockers. This series culminated with the identification of pyrrole amides **13b** and **26d**, with excellent potencies and/or selectivities toward the $Ca_v3.1$- and $Ca_v3.3$-channels. These compounds display poor physicochemical and DMPK properties, making their use difficult for *in vivo* applications. Nevertheless, they are well-suited for *in vitro* studies.

T-type calcium channels belong to the very large family of voltage-regulated channels. Voltage-regulated calcium channels are classically divided into two main groups.[1] The major group represents the high voltage-gated channels, comprising the L-type channels ($Ca_v1.1$–$Ca_v1.4$), the P/Q-type channel ($Ca_v2.1$), the *N*-type channel ($Ca_v2.2$), and the R-type channel ($Ca_v2.3$). These channels typically open upon membrane depolarization to potentials around −20 mV. The second group contains the low voltage-gated T-type channels, represented by three distinct channels named $Ca_v3.1$, $Ca_v3.2$, and $Ca_v3.3$, respectively. These channels typically open at potentials around −60 mV. T-type channels are highly expressed in the brain ($Ca_v3.1$ and $Ca_v3.3$) as well as in the female reproductive system ($Ca_v3.1$ and $Ca_v3.2$), the endocrine system ($Ca_v3.2$ and $Ca_v3.3$), and the gastro-intestinal and cardiovascular systems ($Ca_v3.2$) (www.gtexportal.org). If many functions of T-type calcium channels in these organs have been described, the specific role of each of the three channel subtypes remain largely unknown, not least due to the absence of selective blockers (i.e. $Ca_v3.1$-, $Ca_v3.2$-, or $Ca_v3.3$-selective blockers).

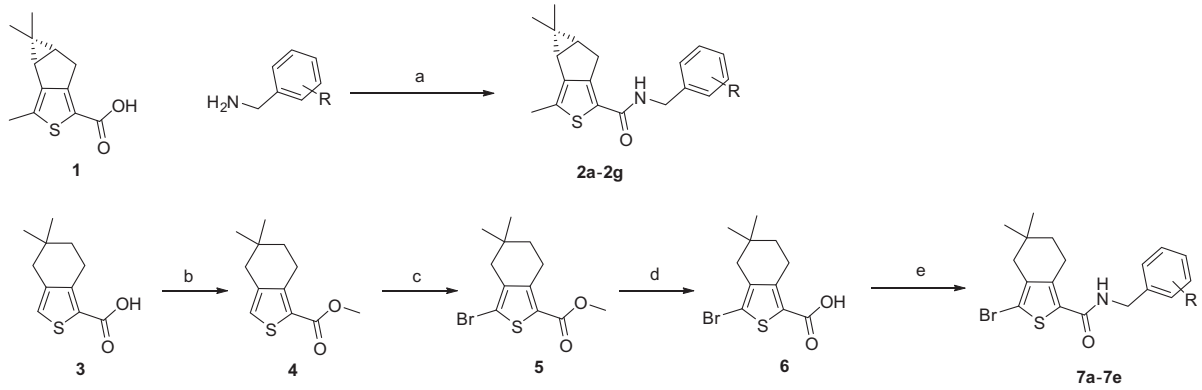
The development of T-type calcium channel blockers has been the subject of recent efforts, as illustrated by the clinical development of MK-8998[2] and Z-944.[3] These compounds have been described as being selective T-type calcium channel blockers, meaning that they block the $Ca_v3.1$, $Ca_v3.2$, and $Ca_v3.3$ channels with similar potencies, while not blocking other channels, in contrast to the prototypical blocker mibefradil.[4] We recently disclosed dihydropyrazole[5] and benzodiazepine[6] derivatives as T-type calcium channel antagonists, and demonstrated their *in vivo* efficacy in the WAG/Rij rat model of absence-like epilepsy. Simultaneously, we observed in spontaneously hypertensive rats that these compounds induced a prolongation of the PR interval of the electrocardiogram (ECG). Due to the excellent PK/PD correlation and a strong parallelism with the desired antiepileptic effect, we hypothesized that PR prolongation was directly linked to the blockade of the T-type calcium channels in the rodent heart. We hypothesized that we might be able to dissociate both effects (i.e. desired antiepileptic effect without PR prolongation) using subtype selective T-type calcium channel blockers, i.e. compounds that would block only one of the three T-type channel. In this paper, we describe the discovery and the profiling of $Ca_v3.2$-subtype blockers, while the discovery of $Ca_v3.1$-subtype blockers is presented in the following paper. Some of the benzodiazepines presented in a previous paper present a moderate selectivity for the $Ca_v3.3$ channel.[5]

An HTS campaign run on $Ca_v3.2$ channels yielded a series of *N*-benzylthiophene-2-carboxamides as moderately potent blockers for this channel. Compound **2a** (Scheme 1, Table 1), which is

Abbreviations: AcOH, acetic acid; Boc, *tert*-butylocycarbonyl; DIPEA, *N*,*N*-diisopropylethylamine; DMAP, 4-(dimethylamino)pyridine; DMF, *N*,*N*-dimethylformamide; EDC·HCl, 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride; HOBt, hydroxybenzotriazole; KHMDS, potassium hexamethyldisilylazide; Ms, mesyl (methylsulfonyl); NBS, *N*-bromosuccinimide; NCS, *N*-chlorosuccinimide; ^{t}Bu, *tert*-butyl; Tf, trifluorosulfonyl; THF, tetrahydrofuran.

* Corresponding author.
E-mail address: olivier.bezencon@idorsia.com (O. Bezençon).

https://doi.org/10.1016/j.bmcl.2017.09.062
0960-894X/



Scheme 1. Preparation $Ca_v3.2$-subtype selective blockers and key nomenclature. (a) EDC·HCl, HOBt, DMAP, DIPEA, CH_2Cl_2, rt, overnight. (b) H_2SO_4, MeOH, reflux, 44 h, 90%. (c) $BnMe_3N^+Br_3^-$, $ZnCl_2$, AcOH, rt, 1 h, 87%. (d) LiOH, THF, H_2O, 40 °C, 24 h, 35%. (e) EDC·HCl, HOBt, DMAP, DIPEA, CH_2Cl_2, rt, overnight.

Table 1
Potencies of selected T-channel blockers (FLIPR®).

Compound	R-substituent[a]	$Ca_v3.1$ IC_{50} (nM)[a]	$Ca_v3.2$ IC_{50} (nM) [a]	$Ca_v3.3$ IC_{50} (nM)[a]	Ratio $Ca_v3.1/Ca_v3.2$
2a	*o*Cl–	600	670	1660	0.9
2b	*o*-Me–	870	990	1500	0.9
2c	*o*-CF_3–	320	61	1060	5.3
2d	*m*-CF_3–	540	530	1150	1.0
2e	*m*-OCF_3–	330	370	1640	0.9
2f	*p*-Me–	1000	1300	1900	0.7
2g	*p*-$EtOCH_2$–	230	160	620	1.5
7a	*o*-$MeOCH_2$–	260	25	810	10
7b	*o*-EtO–	210	64	2760	3.3
7c	*o*-$EtOCH_2$–	140	39	540	3.6
7d	*o*-^{i}PrO–	530	340	2930	1.6
7e	*p*-$MeOCH_2$–	460	470	350	1.0

o: ortho; *m*: meta; *p*: para.
[a] Geometric mean of at least two measurements.

prepared in one step from known carboxylic acid **1**,[7] can be considered a good representative of this series. Using a FLIPR® assay as described in previous publications,[4,5] this compound blocks all three T-type channels with similar potencies. Early SAR studies focused on the amide moiety, applying chemistry described in Scheme 1. Compounds **2a**–**2 g**, based on carboxylic acid **1**, led to a rather shallow SAR. Varying the electronic properties or the position of the substituent led to moderately potent blockers, with the exception of compound **2c**; this compound, bearing an *ortho*-trifluoromethyl substituent, blocked $Ca_v3.2$ with a higher potency. This was a first example of a subtype-selective $Ca_v3.2$ blocker. To simplify chemistry, we prepared compounds **7a** to **7e**, bearing a cyclohexyl ring instead of the 3-carene derived bicyclo[3.1.0]-hexane moiety. They were prepared from known carboxylic acid **3**,[8] via esterification (**4**), bromination (**5**), saponification (**6**) and amide couplings. Both series of blockers **2a**–**2g** and **7a**–**7e** proved to be rather equivalent in terms of potency. Compounds with an *ortho*-substituent at the benzyl substituent emerged as subtype selective $Ca_v3.2$ blockers (**7a**–**7c**), the best one bearing an *ortho*-methoxymethyl substituent (**7a**). Furthermore, the substituent at position 5 of the thiophenyl moiety seemed to be of secondary importance (bromine in series **7** vs. methyl in series **2**).

All compounds remained lipophilic. In a first effort to decrease the lipophilicity, we successfully switched from the thiophene moiety to a pyrrole moiety. Following the chemistry described in Scheme 2, we prepared compounds **13a** to **13l**. Position 5 of the pyrrole moiety had to be substituted by an electro-withdrawing group in order to obtain chemically stable derivatives. These compounds were prepared from 5,5-dimethyl-2-oxocyclohexane-1-carbaldehyde and sarcosine via Schiff-base formation (**9**) and cyclization (**10**). Subsequent chlorination (**11**), saponification (**12**), and amide couplings led to the desired products. It should be noted that the chlorination step proved to be capricious and did not allow a scale-up to multigram quantities. Also, this chlorination step was sometimes more successful when implemented after amide coupling. A chlorine atom was selected for this position as being an electron withdrawing group leading to chemically stable derivatives, and as being rather close to the bromine atom that was tolerated on the thiophenyl system. Pyrrole analogue **13a** confirmed that the pyrrolyl template was adequate for our task (Table 2). Replacing the ethoxy substituent by an isopropoxymethyl substituent on the benzyl group led to compound **13b**, which was clearly identified as a subtype selective $Ca_v3.2$ blocker (selectivity ratio >20 toward $Ca_v3.1$ and $Ca_v3.3$). The selectivity of compound **13b** was confirmed using patch-clamp (IC_{50} ~810 nM and 130 nM for block of $Ca_v3.1$ and $Ca_v3.2$, respectively; $Ca_v3.3$ was not measured).

Pyrrole **13b**, with a molecular weight of 402.9, a clogP of 4.5, and polar surface area of 43.3 A^2, represented a suitable starting point for further investigation. Its calculated ligand efficiency[9] for $Ca_v3.2$ is around 0.40, a rather high value for a T-type channel blocker, and its lipophilic ligand efficiency is of 3.7. With the aim to develop a compound that should penetrate the brain, we calculated a CNS MPO value[10] of 3.5 only for this compound. Here again, the nature of the *ortho*-substituent seemed to be rather unimportant for potency, as can be noticed by comparing compound **13c** with compounds **13d** and **13e**, for which different conformations of the side-chain are expected. Heteroaryl systems like a pyrazolyl