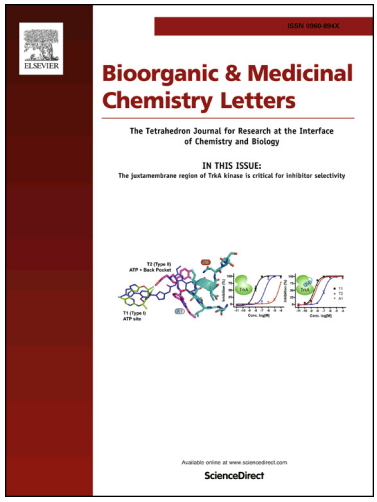
Accepted Manuscript



Easy and rapid preparation of benzoylhydrazides and their diazene derivatives as inhibitors of 15-lipoxygenase

Cristian Tirapegui, Williams Acevedo-Fuentes, Pablo Dahech, Claudia Torrent, Pablo Barrias, Macarena Rojas-Poblete, Carolina Mascayano

PII: S0960-894X(17)30253-6
DOI: http://dx.doi.org/10.1016/j.bmcl.2017.03.017
Reference: BMCL 24769

To appear in: *Bioorganic & Medicinal Chemistry Letters*

Received Date: 16 January 2017
Revised Date: 5 March 2017
Accepted Date: 6 March 2017

Please cite this article as: Tirapegui, C., Acevedo-Fuentes, W., Dahech, P., Torrent, C., Barrias, P., Rojas-Poblete, M., Mascayano, C., Easy and rapid preparation of benzoylhydrazides and their diazene derivatives as inhibitors of 15-lipoxygenase, *Bioorganic & Medicinal Chemistry Letters* (2017), doi: http://dx.doi.org/10.1016/j.bmcl.2017.03.017

Bioorganic & Medicinal Chemistry Letters
journal homepage: www.elsevier.com

Easy and rapid preparation of benzoylhydrazides and their diazene derivatives as inhibitors of 15-lipoxygenase

Cristian Tirapegui[a,*], Williams Acevedo-Fuentes[a], Pablo Dahech[b], Claudia Torrent[b], Pablo Barrias[b], Macarena Rojas-Poblete[c], Carolina Mascayano[b]

[a]*Department of Chemistry, Faculty of Sciences, University of Chile, Santiago, Chile, Casilla 653, Santiago, Chile. cristian.tirapeguic@usach.cl*
[b]*Faculty of Chemistry and Biology, University of Santiago, Santiago, Chile.*
[c]*Relativistic Molecular Physics(ReMoPh) Group, Universidad Andrés Bello, Santiago, Chile.*

ARTICLE INFO

Article history:
Received
Revised
Accepted
Available online

Keywords:
15-lipoxygenase inhibition
Phenylhydrazides
Benzoyldiazene
Antioxidant capability
Lipophilicity.

ABSTRACT

Two series of diaza derivatives were prepared by solvent-free condensation of benzoic acid and 4-substituted phenylhydrazines in order to obtain phenylhydrazides (HYD series) and, by oxidation of these compounds, the corresponding benzoyldiazenes (DIA series). Both sets were evaluated as inhibitors of soybean 15-lipoxygenase activity and antioxidant capability in the FRAP and CUPRAC assays. The most potent inhibitors of both series exhibited IC_{50} values in the low micromolar range. Kinetic studies showed that at least the more active compounds were competitive inhibitors. Docking results indicated that the most potent inhibitor interacts strongly with Ile-839 and iron in the active site.

Lipoxygenases (LOXs) catalyze the production of eicosanoid leukotrienes and lipoxins, both biosynthesized from arachidonic acid (AA)[1] derived from the cell membrane, and important biological mediators of inflammatory processes.[2] This family of enzymes is well conserved among mammalian species[3] and catalyzes stereo- and regiospecific introduction of dioxygen into the polyunsaturated chain of AA as hydroperoxide, which finally is converted to the hydroxyl group.[4] Due to this activity, the name of this enzyme is often preceded by the number of the carbon atom in the AA chain which undergoes oxidation. Thus, using the usual acronym, 5-LOX, 12-LOX and 15-LOX isoforms introduce an oxygen atom at positions C-5, C-12 and C-15 respectively.[5]

Some benzothiophene derivatives and esters of caffeic acid have been reported to be potent inhibitors of 5-LOX. Examples of these are the commercial Zileuton (IC_{50} = 0.4 μM)[6] and CAPE (IC_{50} = 0.13 μM).[7] Likewise, some phenylhydrazones inhibit the dual cyclooxygenase/peroxidase activity of prostaglandin synthase,[8] phenylhydrazide derivatives selectively inhibit cycloxygenase-2,[9] and pyrazole carboxamides inhibit 15-lipoxygenase-1[10,11] but until now we are not aware of any systematic studies of these compounds as 15-LOX inhibitors. The same is true for the related diazene derivatives.

During the last decades a growing interest has arisen to develop new, greener synthetic strategies whereby the amount of residual waste becomes progressively smaller, and a many such reactions can be found in the literature. An interesting example of a green oxidation of arylhydrazides was proposed by Hashimoto,[12] where a phtalocyanine was used as the oxidant. Another example of a green strategy applicable to our work was reported by Metro,[13] who prepared different amide derivatives by mechanochemical means. Specifically, the latter procedure was used to obtain the first set of compounds in this research.

The aim of the present work was to study the effects of two different structural features on the inhibitory behavior of two families of inhibitors: the influence of the oxidation of the central nitrogen-nitrogen single bond of phenylhydrazides, and the systematic substitution of the *para* position of the *N*-phenyl moiety. Both series proposed here were prepared using short and clean procedures, and some of the resulting compounds proved to be fairly potent inhibitors of 15-LOX.

Scheme I shows the synthesis of both series. The first step is the preparation of phenylhydrazide derivatives (HYD series) where both moieties, hydrazine and benzoic acid, where connected by a solvent-free reaction carried out by grinding the reactants in a simple mortar. The oily material prepared by this method was rinsed with pure water affording a white or yellowish white solid that was crystallized in ethanol to obtain fine needles in almost every case. This procedure is an environmentally friendly coupling reaction, with easy recovery of the desired compounds and negligible formation of side products. The second step in Scheme I is the oxidation of the former substances with potassium ferricyanide[14]. This reaction was carried out by shaking two immiscible solutions, one containing the substituted hydrazide dissolved in dichloromethane, and the other, the ferricyanide in a strongly basic aqueous solution. Using