Arylalkylamine Derivatives as Myeloperoxidase Inhibitors, Synthesis and Pharmacological Activity

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Arylalkylamine Derivatives as Myeloperoxidase Inhibitors, Synthesis and Pharmacological Activity
Abstract: Myeloperoxidase (MPO) is an important target for drug design because of its contributing role in many inflammatory syndromes such as atherosclerosis, rheumatoid arthritis, end-stage renal disease or neurodegeneration. Rational drug design assisted by virtual screening is an interesting tool to design new chemical entities that could inhibit MPO. After a high throughput virtual screening of a database, bis-2,2’-[(dihydro-1,3(2H,4H)-pyrimidinediyI)bis(methylene)]phenol was chosen as a starting hit and we used different strategies of chemical synthesis to perform pharmacomodulation described by the three approaches. This led to 36 compounds that have been assessed in an in vitro inhibition MPO test. We found that the arylalkylamine compounds were active but to a lesser extent than the starting hit. Exception for propylamine derivatives with a phenyl cycle should be noticed. As indolic compounds have demonstrated interesting inhibiting properties, we combined indole ring with the phenolhydropyrimidine structure which led to compounds more active than the hit. Among them, propylamine derivatives were new MPO inhibitors with a nanomolar IC50. Kinetics studies for the most potent inhibitors were conducted and reflected a fast reaction with compound I resulting in the accumulation of compound II Structure-activity.

Keywords: Myeloperoxidase; Inhibitors; Arylalkylamine
Introduction

MPO
Neutrophils, monocytes, immune defense system
Phagocytosis
Kills microorganisms
Produces HOCl
MPO is a contributing factor in many inflammatory syndromes such as:

- Atherogenic lesions
- Rheumatoid arthritis
- End-stage renal disease
- Neurodegeneration

Introduction
Finding New MPO inhibitors

MPO

Pharmaceutical Database

HTVS

8 HITS of MPO inhibitors

Selected Hit for pharmacomodulation

\[
\text{IC}_{50} = 0.5 \, \mu\text{M}
\]
Results and discussion

Pharmacomodulation and docking
Validation of docking using poses in HX1, SHA X-ray data

HX1 IN PDB 4C1M

SHA in PDB 1DNW
All compounds were designed and docked in MPO receptors 1DNW-4C1M.

Best poses of the docked compounds were compared with X-Ray data of the known inhibitors HX1 and SHA.

And redocked in same receptors.
Pharmacomodulation

The role of hydroxyl groups on both aromatic cycles A and B

The role of aromatic cycle A and one atom of nitrogen.

The role of the position of the two nitrogen atoms

The role of bridge length between one nitrogen atom and cycle B after removing the hexadrodroperimidine cycle and different substitution on both cycles A and B.
Designed compounds

1. Designed compounds:

   - Compound 2
   - Compound 3
   - Compound 4
   - Compound 5
   - Compound 6
   - Compound 7
   - Compound 8
   - Compound 9
   - Compound 10

2. 1st International Electronic Conference on Medicinal Chemistry
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3. Sponsors: MDPI, Pharmaceuticals
Designed compounds

R₁=OH, R₂= R₃= R₄= H  n= 2  11
R₁=OH, R₂= R₃= R₄= H  n= 3  12
R₁=R₂= R₃=R₄=H  n= 2  13
R₁=R₂= R₃=R₄=H  n= 3  14
R₁=OH, R₂= R₃=H, R₄= F  n= 2  15
R₁=OH, R₂= R₃=H, R₄= F  n= 3  16

R₁=OH, R₂= R₃= H,  n= 2  17
R₁=OH, R₂= R₃= H,  n= 3  19
R₁= R₂= R₃=H,  n= 2  21
R₁= R₂= R₃=H,  n= 3  23
R₁=F, R₂= R₃=H  n= 2  25
R₁=F, R₂= R₃=H  n= 3  27
R₁=R₂= H, R₃=F  n= 2  29
R₁=R₂= H, R₃=F  n= 3  31
R₁=H, R₂=F, R₃=H  n= 2  33
R₁=H, R₂=F, R₃=H  n= 3  35

R₁=OH, R₂= R₃= H,  n= 2  18
R₁=OH, R₂= R₃= H,  n= 3  20
R₁=R₂= R₃=H,  n= 2  22
R₁=R₂= R₃=H,  n= 3  24
R₁=F, R₂= R₃=H  n= 2  26
R₁=F, R₂= R₃=H  n= 3  28
R₁=R₂= H, R₃=F  n= 2  30
R₁=R₂= H, R₃=F  n= 3  32
R₁=H, R₂=F, R₃=H  n= 2  34
R₁=H, R₂=F, R₃=H  n= 3  36

n=2  37
n=3  38
Some docked poses of the designed compounds in MPO Receptor

Compound 4 Shows hydrogen bonds with Glu102 and Phe147 Arg239 and salt bridge with Glu102

Compound 20 Shows hydrogen bonds with Glu102 and Phe147 and salt bridge with Glu102

Docking results gave some similar interactions as with HX1 and SHA A1 and different free Energy levels -ΔG or affinities with MPO receptors
Chemistry
Chemistry

Reagents and conditions:
(i) NaBH₄, EtOH
(ii) formaldehyde (37 wt % aqueous solution), EtOH
(iii) BnCl, NaH, DMF, Reflux

Reagents and conditions:
(i) NaBH₄, acetic acid, EtOH for 4
(ii) NaBH₃CN, acetic acid, EtOH for 5-8
Reagents and conditions: (i) NaBH₃CN, acetic acid, EtOH (ii) BBr₃, DCM

Reagents and conditions: (i) NaBH₄, MeOH
Chemistry

Reagents and conditions: (i) NaBH<sub>4</sub>, MeOH
(ii) BBr<sub>3</sub>, DCM

Reagents and conditions: (i) NaBH<sub>4</sub>, MeOH
MPO inhibition assay
Best synthesized compounds with its IC50

16
IC50 = 0.37 μM

20
IC50 = 0.37 μM

28
IC50 = 0.27 μM

24
IC50 = 0.36 μM

36
IC50 = 0.37 μM

32
IC50 = 0.49 μM

38
IC50 = 0.054 μM

37
IC50 = 0.058 μM
Transient-State Kinetics

**Chlorination cycle**
- \( \text{HOCl} \rightarrow \text{Cl}^- \)
- \( \text{Cl}^- + \text{H}_2\text{O}_2 \rightarrow \text{MPO}^{2+} - \text{Fe}^{4+} - \text{O} \)

**Peroxidase cycle**
- \( \text{A}^+ + \text{H}_2\text{O} \rightarrow \text{MPO} - \text{Fe}^{4+} - \text{OH} \)
- \( \text{A}^+ \rightarrow \text{AH} \)
- \( \text{AH} \rightarrow \text{MPO} - \text{Fe}^{3+} \)

**Reactions**
- \( k_2 \)
- \( k_3 \)
Mechanism of action
Transient-State Kinetics

<table>
<thead>
<tr>
<th>Compound</th>
<th>Compound I reduction rate constant ((M^{-1}s^{-1}))</th>
<th>Compound II reduction rate constant ((M^{-1}s^{-1}))</th>
<th>Ratio of compound I rate to compound II rate</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image1" alt="Compound I" /></td>
<td>1.5 \times 10^6</td>
<td>4.8 \times 10^3</td>
<td>313</td>
</tr>
<tr>
<td><img src="image2" alt="Compound II" /></td>
<td>5.7 \times 10^6</td>
<td>1.4 \times 10^3</td>
<td>4071</td>
</tr>
<tr>
<td><img src="image3" alt="Compound III" /></td>
<td>1.4 \times 10^7</td>
<td>3.5 \times 10^3</td>
<td>4000</td>
</tr>
</tbody>
</table>
The kinetic rate constant of reaction of the three best compounds with MPO/compound I /compound II have been measured.

\[ y = 14.378x + 29.649 \]
\[ R^2 = 0.9983 \]

The reaction from compound I to compound II is too fast but the reaction with compound II is slow leading to the accumulation of compound II.
Conclusion
Pharmacomodulation

**Conclusion**

A1-IC$_{50}$ 0.5 µM

MPO IC$_{50}$

with IC$_{50}$ 0.3-0.05 µM. More active to 2-10 times than the A1

n=2 37
n=3 38

R$_1$=OH, R$_2$=R$_3$=H, R$_4$=F 16
R$_1$=OH, R$_2$=R$_3$=H, R$_4$=OH 20
R$_1$=R$_2$=R$_3$=H, R$_4$=OH 24
R$_1$=F, R$_2$=R$_3$=H, R$_4$=OH 28
R$_1$=R$_2$=H, R$_3$=F R$_4$=OH 32
R$_1$=H, R$_2$=F, R$_3$=H R$_4$=OH 36
Conclusion

*best compounds have shown high reduction rate constants of compound I and II and their ratio can explain the accumulation of compound II, illustrating a reversible mechanism of inhibition..

*Arylpropylamine derivatives and adding the indole structure to the original scaffold A1 have given us new effective MPO inhibitors

IC₅₀ = 0.3-0.05 µM
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