DESIGN AND SYNTHESIS OF THE INHIBITORS OF CYSTEINE BIOSYNTHESIS AS ENHANCERS OF ANTIBACTERIAL THERAPY

Giannamaria Annunziato[1], Marco Pieroni[1], Claudia Beato[1], Barbara Campanini[1], Roberto Benoni[2], Stefano Bettati[2], Thelma Pertinhez[3], Andrea Mozzarelli[1] and Gabriele Costantino[1].

[1]Dipartimento di Farmacia, [2]Dipartimento di Neuroscienze and [3]Centro Interdipartimentale Misure, Università degli Studi di Parma, Parma, Italy.

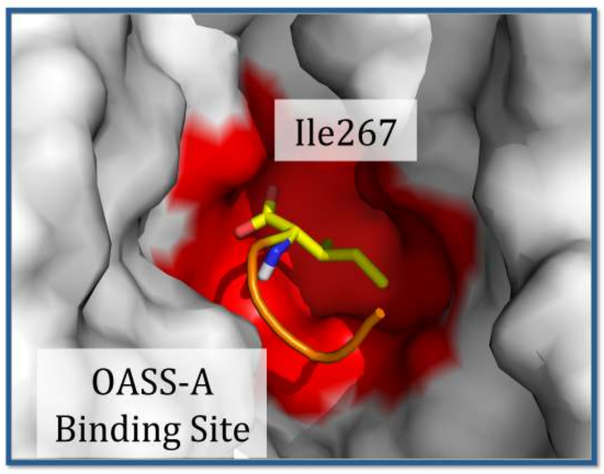
Introduction

Antimicrobial resistance is the ability of bacteria to withstand the effect of drugs. As a global health problem, the development of new molecules targeting the mechanism of resistance is strongly needed.

Within the sulfur assimilation pathway, the **biosynthesis of cysteine** is involved in the virulence of many bacteria and in their capability to bypass the oxidative stress induced by antibacterials.

The last step of cysteine biosynthesis is catalyzed by the enzyme **O-acetylserine sulphydrilase** (**OASS**), absent in humans, and expressed in two isoforms: OASS-A and OASS-B.

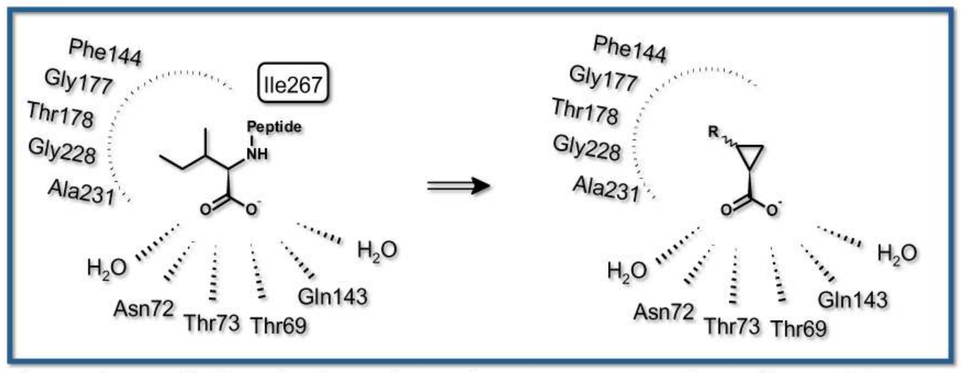
OASS is physiologically inhibited by **Serine acetyl transferase (SAT)** through its C-terminal portion, constituted, in general, by a pentapeptide unit. The last residue of the pentapeptide is a isoleucine (Ile), highly conserved throughout many bacterial spaces.



EcCysE	N H T F E Y G D G I
StCysE	H H T F E Y G D G I
HiCysE	G I D D G M N L N I
NmCysE	I Q F T E I D F M I
YpCysE	I Q G F E Y G D G I
BaCysE	V M D Q M L G D G I
MtCysE	G I W H G E D F S I
SaCysE	N G E I Q D D Y I I
BsCysE	E D R K E R I N Q K
AtCysE	Y L T E W S D Y V I
OsCysE	F I Q Q W S D Y S I

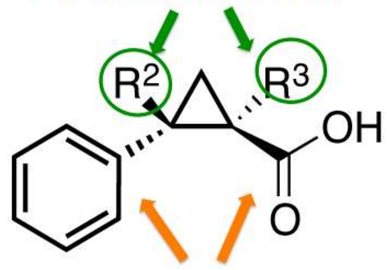
Aim of the work

Based on computational studies and biological evaluations, we propose a series of compounds that mimicking the **Ile267** of **SAT**.



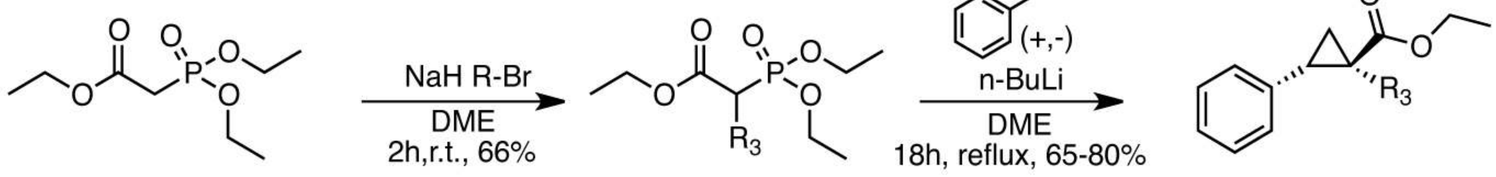
A series of 2-substitued cyclopropane carboxylic acids are synthesized as potential inhibitors of both isoforms of OASS enzymes.

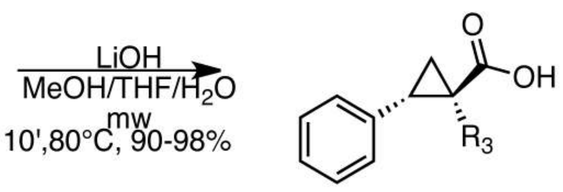
Site of modifications

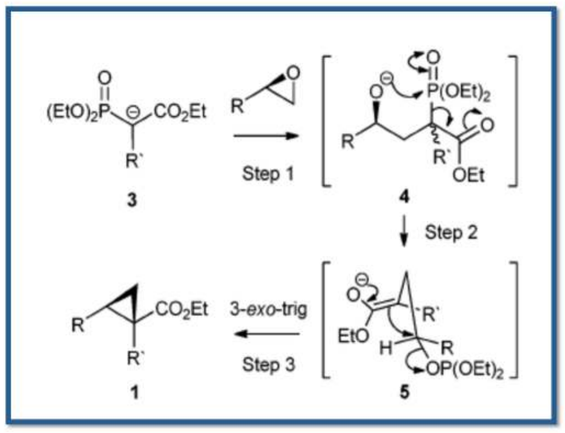


Trans configuration is required

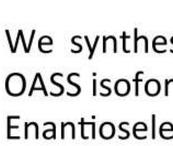
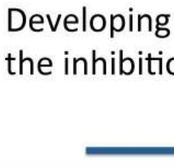
Synthesis



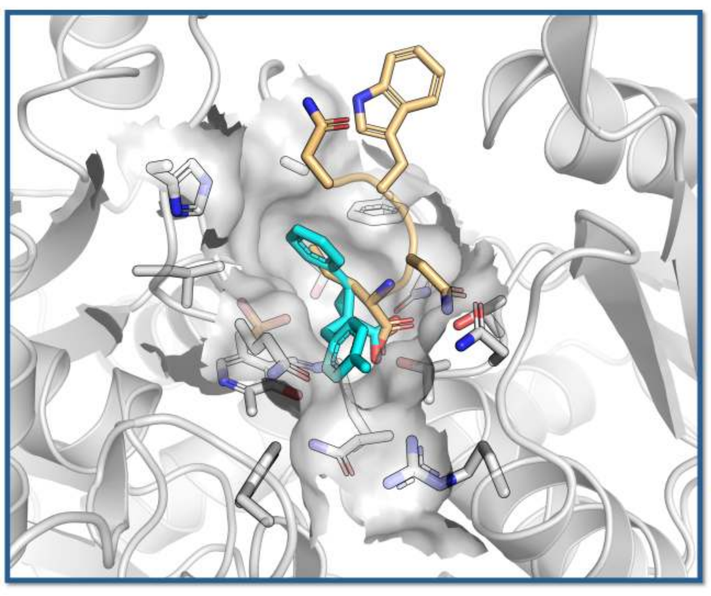




Results

Structure	Ligand		$K_{d,}$ CysK (μM)	$K_{d,}$ CysM (μM)
	UPAR-315	±	15 ± 1	560 ± 100
	UPAR-393	1S, 2R (+)	12.1 ± 0.5	860 ± 90
	UPAR-421	1R, 2S (-)	1200 ± 300	>2000
	UPAR-316	±	0.20 ± 0.02	4.1 ± 0.2
	UPAR-394	1S, 2S (+)	0.067 ± 0.007	1.66 ± 0.07
	UPAR-419	1R, 2R (-)	50 ± 8	197 ± 27
	UPAR-414	±	0.075 ± 0.007	1.2 ± 0.2
	UPAR-415	1S, 2S (+)	0.067 ± 0.003	0.49 ± 0.05
	UPAR-422	1R, 2R (-)	81 ± 17	121 ± 12
	UPAR-416	±	0.156 ± 0.007	0.76 ± 0.10
	UPAR-417	1S, 2S (+)	0.074 ± 0.008	0.42 ± 0.06
	UPAR-420	1R, 2R (-)	84 ± 10	152 ± 19

Docking studies



- UPAR-415 (1S, 2S) were docked into OASS-A binding site.
- UPAR-415 binds into Ile267 binding pocket;
- The carboxylic moieties assume a similar orientation;
- The phenyl moiety protrudes in the Ile297 side chain pocket;
- The benzyl moiety fills a new bulky and hydrofobic pocket.

Conclusions and future perspectives

- We synthesized compounds with high activity against both OASS isoforms, in low nanomolar range;
- Enantioselective, reproducible and scalable synthesis;
- Developing a new library of compounds which could lead to the inhibition of SAT activity.

References

Tai, *et al. Biochem*. **1993**, 6433;
Mozzarelli *et al. Biochim. Biophys. Acta*, **2011**, 1497;
Bruno, *et al. Mol. Inform.* **2013**, *447;*
Amori, *et al. Med. Chem. Comm.* **2012**, *1111.*