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Antistaphylococcal activity of new salicylamide analogues

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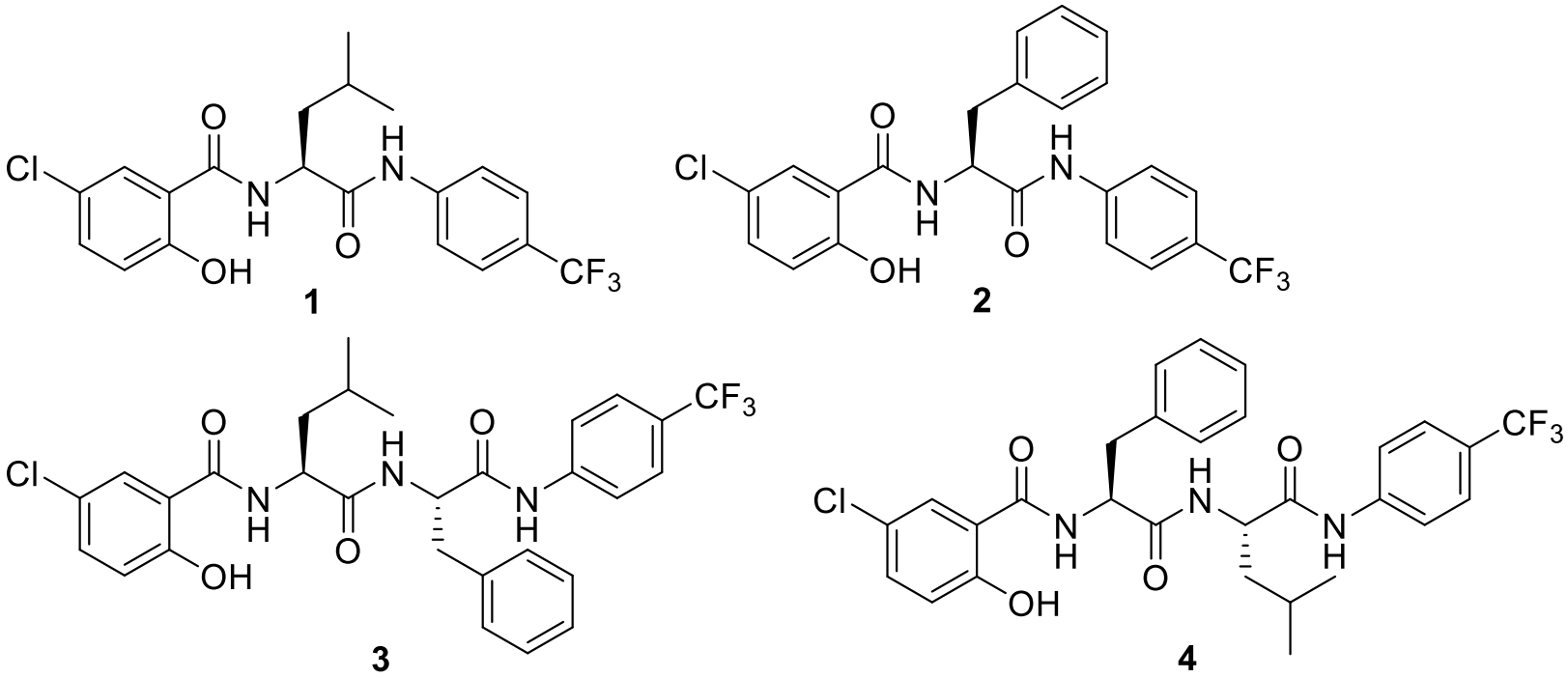
Graphical Abstract



Compounds 1 and 2:

S. aureus: MIC = 1.08 and 1.17 μM / MBC = 1.17 and 2.16 μM

MRSA strains: MIC = 0.07-2.16 μM / MBC = 0.07-17.28 μM

Abstract:

Antimicrobial resistance is still a serious global threat. *Staphylococcus aureus* is a common cause of severe infections in health facilities and the community. People with methicillin-resistant *S. aureus* (MRSA) are estimated to be 64% more likely to die than people with a non-resistant form of the infection. For that reason, the research and development of new active compounds is really needed.

Salicylamides are anti-infectious agents with a wide range of pharmacological effects, such us antiviral, antibacterial, antifungal and anthelminthic. Thus, derivatives of this group are very promising compounds.

A series of newly synthesized complex salicylamide derivatives was tested for their antimicrobial effect against methicillin-resistant *Staphylococcus aureus* (MRSA). *Staphylococcus aureus* ATCC 29213 as a control and three isolates of MRSA were used. The activity was assessed by the evaluation of minimum inhibitory concentration. Using the microdilution method with subcultivation of aliquots, the minimum bactericidal concentration was determined too. Ciprofloxacin and ampicillin were used as standard antibacterial drugs. Four most promising compounds from the series were chosen and are demonstrated in this contribution.

Keywords: antibacterial activity; Staphylococcus aureus; MRSA; salicylamide

Introduction

Testing of antistaphylococcal activity of newly synthesized compound is very important and needed because of severity of resistance and global dissemination of methicillin-resistant *S. aureus* (MRSA) strains.

Salicylamide derivatives are really promised compounds thanks to their wide spectrum of activity. Previous series of these structures was tested for its effect against VRE (vancomycin-resistant enterococci). Three of the investigated compounds showed strong bacteriostatic activity against VRE (0.199–25 μM) comparable to or more potent than ampicillin and ciprofloxacin[1].

New salicylamide-based compound were prepared and tested against three isolates of MRSA – MRSA 63718, MRSA SA 630 and MRSA SA 3202. Minimum inhibitory concentration (MIC) was assessed by microdilution method with microtitration plate. Each compound was diluted to concentration 256–0.008 μg/ml. The MIC was defined as the lowest concentration of the compound, at which no visible bacterial growth was observed. Minimum bactericidal concentration (MBC) was determined by subcultivation of aliquots from previous testing. Results are expressed in "μM" unit for higher informative value.

1. Pospíšilová, Š.; Michnová, H.; Kauerová, T.; Pauk, K.; Kollár, P.; Vinšová, J.; Imramovský, K.; Čížek, A.; Jampílek, J. *In vitro* activity of salicylamide derivatives against vancomycin-resistant enterococci. *Bioorg. Med. Chem. Lett.* **2018**, *28*, 2184-2188.

Results

Table 1: Minimum inhibitory (MIC) and bactericidal (MBC) concentration [µM];
S. aureus – *Staphylococcal aureus*, cpx – ciprofloxacin, amp - ampicillin

Compound	***S. aureus* ATCC 29213**		**MRSA 63718**		**MRSA SA 3202**		**MRSA SA 630**	
	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC
1	1.17	1.17	0.07	0.07	1.17	1.17	1.17	1.17
2	1.08	2.16	0.27	0.27	2.16	2.16	1.08	17.28
3	222	> 444	> 444	> 444	444	> 444	444	> 444
4	55.85	112	447	> 447	55.85	> 447	55.85	447
cpx	0.75	0.75	24.14	24.14	386.30	386.30	48.29	48.29
amp	1.43	2.86	>45.79	>45.79	>45.79	>45.79	>45.79	>45.79

Discussion

It was found previously that substitution of anilide ring by a lipophilic and electron-withdrawing moiety, such as CF_3, significantly increases antibacterial activity[1,2]. This substitution of the *para*-position seems to be the most advantageous, which was also confirmed in this case.

Based on the results, it is evident that elongation of the chain/insertion of another amino acid significantly decreased the activity (compound **4**) or it caused its loss (compound **3**).

The efficacy of compounds **1** and **2** against reference strain *S. aureus* ATCC 29213 was comparable with activity of ciprofloxacin and ampicillin. Excellent results were detected for the compound **1** and **2** against MRSA isolates, where the MIC is distinctly lower than the MIC of ciprofloxacin. These two structures demonstrate the influence of the CF_3 moiety. The compounds differ only by the presence of isobutyl and phenyl on the "diamide" skeleton, which, based on the results, has only a secondary effect on activity.

1. Pospíšilová, Š.; Michnová, H.; Kauerová, T.; Pauk, K.; Kollár, P.; Vinšová, J.; Imramovský, K.; Čížek, A.; Jampílek, J. *In vitro* activity of salicylamide derivatives against vancomycin-resistant enterococci. *Bioorg. Med. Chem. Lett.* **2018**, *28*, 2184-2188.
2. Zadražilová, I.; Pospíšilová, Š.; Pauk, K.; Imramovský, A.; Vinšová, J.; Čížek, A.; Jampílek, J. In Vitro Bactericidal Activity of 4- and 5-Chloro-2-hydroxy-*N*-[1-oxo-1- (phenylamino)alkan-2-yl]benzamides against MRSA. *Biomed Res. Int.* 2015, 2015, Article ID 349534, http://www.hindawi.com/journals/bmri/aa/349534/. [ISSN 2314-6133]

Conclusions

One of the ways to solve the problem of antibacterial resistance is the development of new effective drugs. In this contribution there are two potential compounds with high potency against MRSA.

Structure-activity relationships prove the importance of CF_3 moiety. In addition, a comparison of compounds **1**/**3** and **2**/**4** showed that the insertion of another amino acid and elongation of the chain resulted in loss of activity. These findings are important for synthesis of new optimized compounds.

(*S*)-5-Chloro-2-hydroxy-*N*-(4-methyl-1-oxo-1-{[4-(trifluoromethyl)phenyl]-amino}pentan-2-yl)benzamide (**1**) and (*S*)-5-chloro-2-hydroxy-*N*-(1-oxo-3-phenyl-1-{[4-(trifluoromethyl)phenyl]amino}propan-2-yl)benzamide (**2**) proved to be suitable for further investigation.

Acknowledgments

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