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# *In Vitro* Antibacterial Spectrum of BC-7013, a Novel Pleuromutilin Derivative for Topical Use in Humans


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## Abstract


**Background:** BC-7013 [14-O-[(3-Hydroxymethyl-phenylsulfanyl)-acetyl]-mutilin] is a novel semi-synthetic pleuromutilin derivative that inhibits prokaryotic protein synthesis. BC-7013 is in early stage of clinical development for the topical treatment of uncomplicated skin and skin structure infections (uSSSI). We assessed BC-7013 activity against a wide range of clinical isolates.

**Methods:** BC-7013 and comparators were susceptibility (S) tested by the CLSI broth microdilution methods against 1063 recent clinical isolates (mainly from 2006-2007) from the USA and Europe collected from the SENTRY Antimicrobial Surveillance Program.

**Results:** BC-7013 was the most active agent against *Staphylococcus* spp. and *Streptococcus* spp. including methicillin-resistant *S. aureus* (MRSA) ($MIC_{90}$, 0.03 µg/ml), community-acquired (CA-MRSA) ($MIC_{90}$, 0.03 µg/ml) and penicillin-resistant *Streptococcus pneumoniae* (PEN-R SPN) ($MIC_{90}$, 0.06 µg/ml). Against *S. aureus*, BC-7013 was significantly more potent than retapamulin (RET) and the other topical agents tested. BC-7013 was highly active against coagulase-negative staphylococci (CoNS) ($MIC_{90}$, 0.03-0.25 µg/ml), and beta-hemolytic streptococci including group A ($MIC_{90}$, 0.03 µg/ml) and B ($MIC_{90}$, 0.06 µg/ml). BC-7013 was fully active against isolates resistant (R) to the topical comparison agents.

**Conclusions:** BC-7013 exhibited potent antimicrobial activity against the most prevalent Gram-positive pathogens involved in uSSSI, being more active against staphylococci than antimicrobials currently available and inhibited growth of 100% of *S. aureus* at an MIC ≤0.03 µg/ml, including MRSA.

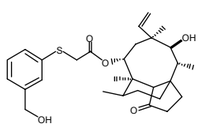

## Introduction

Gram-positive microorganisms, such as *Staphylococcus aureus,* are the causative agents of severe infections including skin and skin structure infections (SSSI). Over the past four decades methicillin-resistant *S. aureus* (MRSA) has spread throughout the world and become highly endemic in many geographic areas causing severe morbidity and mortality in hospitals worldwide. Community-acquired MRSA (in particular the highly virulent clone USA300) has become more and more prevalent since the beginning of this millennium and the development of resistances to systemic and topical agents dictate the need for novel antibiotics such as pleuromutilins.

Pleuromutilin-derived antibiotics are inhibitors of ribosomal protein synthesis interacting with the central part of domain V of the 23S rRNA at the ribosomal peptidyl transferase cavity and preventing the correct positioning of the CCA-ends of tRNAs for peptide transfer and translation initiation.[1-4, 6] One example for pleuromutilins is retapamulin[5] approved for topical treatment of impetigo, infected small lacerations and wounds.

In this study we present the antibacterial activity of the novel pleuromutilin derivative BC-7013 (Figure 1) in comparison to established topical antibiotics. BC-7013 is characterized by excellent activity against Gram-positive bacterial pathogens and is in early clinical development for the topical treatment of uSSSI.

**Figure 1: Structure of BC-7013**




## Methods

**Organism Collection:** The activity of BC-7013 was determined against bacterial pathogens causing uSSSI. Organisms included strains isolated from patients in either the United States (USA, 52.3%) or European (47.6%) medical centers (n=67) primarily during 2006 to 2007. Less commonly isolated species and those organisms with characterized resistance mechanism were included from 2005 (7.8%), 2004 (4.7%) and 2000-2003 (7.2%). Most of the isolates were from patients with bloodstream infections (63.1%), respiratory tract infections (29.6%) and skin and skin structure infections (5.6%).

Organisms examined included amongst others methicillin susceptible *S. aureus* (MSSA, 151), methicillin resistant *S. aureus* (MRSA, 102), community-acquired MRSA (CA-MRSA, 50), ß-hemolytic streptococci (152) including *S. pyogenes* (50), *S. agalactiae* (50).

**Susceptibility test methods:** MIC values were determined according to the CLSI guidelines (M07-A8, 2009 and M100-S19, 2009).[8] 96-well format panels were produced by JMI Laboratories using Mueller Hinton broth, cation adjusted MHB supplemented with 2-5% lysed horse blood for testing *Streptococcus* spp.. Interpretive criteria for comparator antibiotics were used as published by CLSI (M100-S19, 2009).[7]

Quality control was performed as recommended by CLSI using the following strains: *S. aureus* ATCC 29213 and *S. pneumoniae* ATCC 49619.

## Results

- The antibacterial activity of BC-7013 and comparator antibiotics is presented in Table 1 and the cumulative percentage of strains inhibited by BC-7013 is shown in Table 2.
- BC-7013 was very active against the 303 tested *S. aureus* isolates. With a $MIC_{50}$ value of 0.015 µg/ml and a $MIC_{90}$ value of 0.03 µg/ml. BC-7013 was four-fold more potent than retapamulin ($MIC_{90}$ of 0.12 µg/ml) and was the most potent compound tested against *S. aureus*. 100.0% of *S. aureus* strains were inhibited at ≤ 0.03 µg/ml.
- BC-7013 potency was the same for MRSA and MSSA. The rank order of potency for those agents with "on-scale" $MIC_{50}$ values against MSSA was: BC-7013 (0.03 µg/ml) > retapamulin, clindamycin and minocycline (0.12 µg/ml) > levofloxacin and fusidic acid (0.25 µg/ml) > vancomycin (1 µg/ml) > linezolid (2 µg/ml). Neomycin ($MIC_{90}$ ≤1.7 µg/ml) was also active against these isolates. MRSA isolates were much less susceptible to macrolides (16.7%), clindamycin (57.8%) and levofloxacin (8.8%) but fully susceptible to BC-7013. CA-MRSA resistant to macrolides were also fully susceptible to BC-7013 (MIC range 0.03-0.06 µg/ml.
- BC-7013 was also the most potent compound tested against CoNS. Among the oxacillin-resistant isolates, four out of the five linezolid-resistant strains had only slightly elevated BC-7013 MIC values (0.06 – 0.25 µg/ml). 14.3% of isolates had a high-level resistance to mupirocin (MIC >128 µg/ml) and higher MIC values were noted among the oxacillin-resistant CoNS isolates, all of them being fully susceptible to BC-7013.
- Against β-hemolytic streptococci including *S. pyogenes* and *S. agalactiae* BC-7013 was the most active compound too with MIC values ranging from 0.015 to 0.06 µg/ml. 100.0% of the isolates were inhibited at a BC-7013 concentration of 0.06 µg/ml. The *S. pyogenes* isolates were very susceptible to the comparator agents (4.0 – 6.0% macrolide resistance). Higher rates of macrolide resistance were noted among the other species with 15.4% for group C and G and 34.0% for *S. agalactiae* isolates.

**Table 1. Antibacterial spectrum of BC-7013 and topical comparator antibiotics tested against Gram-positive bacteria**

| Antimicrobial agent | $MIC_{50}$ | $MIC_{90}$ | Range | % susceptible/ resistant [a] |
|---|---|---|---|---|
| ***S. aureus* (303)** | | | | |
| BC-7013 | 0.015 | 0.03 | 0.008 – 0.03 | - / - |
| Retapamulin | 0.12 | 0.12 | 0.03 – 0.12 | - / - |
| Erythromycin | 16 | >16 | 0.12 – >16 | 48.8 / 51.2 |
| Clindamycin | 0.12 | >16 | 0.03 – >16 | 84.8 / 15.2 |
| Minocycline | 0.12 | 0.25 | 0.06 – 8 | 98.0 / 0.0 |
| Neomycin | ≤1.7 | 109 | ≤1.7 – 219 | - / - |
| Mupirocin | ≤4 | ≤4 | ≤4 – >128 | - / - |
| Fusidic Acid | 0.25 | 0.25 | 0.12 – >32 | - / - |
| **MSSA (151)** | | | | |
| BC-7013 | 0.015 | 0.03 | 0.015 – 0.03 | - / - |
| Retapamulin | 0.12 | 0.12 | 0.03 – 0.12 | - / - |
| Erythromycin | 0.5 | >16 | 0.12 – >16 | 86.8 / 13.2 |
| Clindamycin | 0.12 | 0.12 | 0.06 – >16 | 98.0 / 2.0 |
| Minocycline | 0.12 | 0.12 | 0.06 – 8 | 99.3 / 0.0 |
| Neomycin | ≤1.7 | ≤1.7 | ≤1.7 – 109 | - / - |
| Mupirocin | ≤4 | ≤4 | ≤4 | - / - |
| Fusidic Acid | 0.25 | 0.25 | 0.12 – 16 | - / - |
| **MRSA (102)** | | | | |
| BC-7013 | 0.015 | 0.03 | 0.008 – 0.03 | - / - |
| Retapamulin | 0.12 | 0.12 | 0.06 – 0.12 | - / - |
| Erythromycin | >16 | >16 | 0.12 – >16 | 16.7 / 83.3 |
| Clindamycin | 0.12 | >16 | 0.03 – >16 | 57.8 / 42.2 |
| Minocycline | 0.12 | 0.25 | 0.06 – 8 | 95.1 / 0.0 |
| Neomycin | 6.8 | 109 | ≤1.7 – 219 | - / - |
| Mupirocin | ≤4 | ≤4 | ≤4 – >128 | - / - |
| Fusidic Acid | 0.25 | 0.25 | 0.12 – >32 | - / - |
| **CA-MRSA (50)** | | | | |
| BC-7013 | 0.015 | 0.03 | 0.015 – 0.03 | - / - |
| Retapamulin | 0.12 | 0.12 | 0.06 – 0.12 | - / - |
| Erythromycin | >16 | >16 | >16 | 0.0 / 100.0 |
| Clindamycin | 0.12 | 0.12 | 0.06 – 0.12 | 100.0 / 0.0 |
| Minocycline | 0.06 | 0.25 | 0.06 – 0.25 | 100.0 / 0.0 |
| Neomycin | 109 | 109 | ≤1.7 – 219 | - / - |
| Mupirocin | ≤4 | ≤4 | ≤4 – >128 | - / - |
| Fusidic Acid | 0.25 | 0.25 | 0.12 – 0.25 | - / - |
| **CoNS (105)** | | | | |
| BC-7013 | 0.03 | 0.06 | 0.008 – >8 | - / - |
| Retapamulin | 0.06 | 0.5 | 0.015 – >8 | - / - |
| Erythromycin | >16 | >16 | 0.06 – >16 | 43.8 / 53.3 |
| Clindamycin | 0.12 | >16 | 0.03 – >16 | 80.0 / 16.2 |
| Minocycline | 0.12 | 0.5 | 0.03 – 0.5 | 100.0 / 0.0 |
| Neomycin | ≤1.7 | 6.8 | ≤1.7 – 109 | - / - |
| Mupirocin | ≤4 | >128 | ≤4 – >128 | - / - |
| Fusidic Acid | 0.25 | 16 | 0.06 – 32 | - / - |
| **ß-hemolytic streptococci [b] (152)** | | | | |
| BC-7013 | 0.03 | 0.03 | 0.015 – 0.06 | - / - |
| Retapamulin | 0.03 | 0.06 | 0.015 – 0.25 | - / - |
| Erythromycin | 0.06 | 8 | 0.015 – >16 | 80.9 / 17.8 |
| Clindamycin | 0.06 | 0.12 | ≤0.008 – >16 | 90.8 / 7.9 |
| Minocycline | 0.12 | 16 | 0.03 – >16 | - / - |
| Neomycin | 3.4 | 13.7 | ≤1.7 – 27.3 | - / - |
| Mupirocin | ≤4 | ≤4 | ≤4 | - / - |
| Fusidic Acid | 8 | 16 | 2 – 16 | - / - |
| ***S. pyogenes* (50)** | | | | |
| BC-7013 | 0.015 | 0.03 | 0.015 – 0.06 | - / - |
| Retapamulin | 0.03 | 0.06 | 0.015 – 0.06 | - / - |
| Erythromycin | 0.03 | 0.06 | 0.03 – 8 | 94.0 / 4.0 |
| Clindamycin | 0.06 | 0.06 | 0.06 | 100.0 / 0.0 |
| Minocycline | 0.06 | 0.12 | 0.03 – 8 | - / - |
| Neomycin | ≤1.7 | ≤1.7 | ≤1.7 | - / - |
| Mupirocin | ≤4 | ≤4 | ≤4 | - / - |
| Fusidic Acid | 4 | 8 | 2 – 8 | - / - |
| ***S. agalactiae* (50)** | | | | |
| BC-7013 | 0.03 | 0.06 | 0.015 – 0.06 | - / - |
| Retapamulin | 0.03 | 0.06 | 0.015 – 0.06 | - / - |
| Erythromycin | 0.06 | >16 | 0.03 – >16 | 66.0 / 34.0 |
| Clindamycin | 0.06 | >16 | 0.03 – >16 | 82.0 / 18.0 |
| Minocycline | 16 | 16 | 0.06 – >16 | - / - |
| Neomycin | 13.7 | 13.7 | ≤1.7 – 13.7 | - / - |
| Mupirocin | ≤4 | ≤4 | ≤4 | - / - |
| Fusidic Acid | 16 | 16 | 8 – 16 | - / - |

[a] Criteria as published by the CLSI [2008]. β-lactam susceptibility should be directed by the oxacillin test results.
[b] Includes: Group A *Streptococcus* (50 strains), Group B *Streptococcus* (50 strains), Group C *Streptococcus* (26 strains), and Group G *Streptococcus* (26 strains).

**Table 2. MIC frequency distributions of BC-7013 against 560 bacterial isolates**

| Organism (no. tested) | ≤0.004 | 0.008 | 0.015 | 0.03 | 0.06 | 0.12 | 0.25 | 0.5 | 1 | 2 | 4 | 8 | >8 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| *S. aureus* (303) | - | 0.3 | 77.6 | 100.0 | - | - | - | - | - | - | - | - | - |
| Oxacillin-susceptible (151) | - | - | 78.2 | 100.0 | - | - | - | - | - | - | - | - | - |
| Oxacillin-resistant (102) | - | 1.0 | 73.5 | 100.0 | - | - | - | - | - | - | - | - | - |
| CA-Oxacillin-resistant (50) | - | - | 84.0 | 100.0 | - | - | - | - | - | - | - | - | - |
| Coagulase-negative staphylococci (105) | - | 1.9 | 39.1 | 86.7 | 90.5 | 93.3 | 95.2 | 96.2 | 96.2 | 97.1 | 99.1 | 99.1 | 100.0 |
| Oxacillin-susceptible (50) | - | 4.0 | 44.0 | 96.0 | 98.0 | 98.0 | 98.0 | 98.0 | 98.0 | 100.0 | - | - | - |
| Oxacillin-resistant (55) | - | - | 34.6 | 78.2 | 83.6 | 89.1 | 92.7 | 94.6 | 94.6 | 94.6 | 98.2 | 98.2 | 100.0 |
| Beta-hemolytic streptococci (152) | - | - | 42.1 | 92.8 | 100.0 | - | - | - | - | - | - | - | - |

## Conclusions

- BC-7013 was the most active compound against common Gram-positive bacterial pathogens such as *S. aureus*, *S. pyogenes* and *S. agalactiae* when compared with other topical antibiotics.
- BC-7013 was highly active against *Staphylococcus* spp., including MRSA and CA-MRSA, and BC-7013 inhibited 100.0% of *S. aureus* at an MIC value ≤0.03 µg/ml. The overall potency of BC-7013 ($MIC_{90}$ of 0.03 – 0.06 µg/ml) against *S. aureus* and CoNS was 4- to 8-fold greater than retapamulin and was more active than the other tested agents.
- BC-7013 also exhibited potent activity against group A ($MIC_{90}$ of 0.03 µg/ml) and group B ($MIC_{90}$, 0.06 µg/ml) streptococci, being the next most common cause of uSSSI.
- All bacterial isolates resistant to macrolides, clindamycin, tetracyclines, mupirocin and fusidic acid were fully susceptible to BC-7013. As expected no cross-resistance was noted with these agents.
- Overall, BC-7013 has an excellent activity against Gram-positive skin pathogens being clearly superior to that of antibiotics currently in topical use to treat uSSSI.

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