

Clinical Candidate AA139 - Efficacy Against ESBL Producing *E. coli* in the Murine Urinary Tract Infection Model

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Abstract

Introduction

Arenicin-3 was originally isolated from the lugworm *Arenicola marina* and structural analysis revealed a β -hairpin structure harboring 4 arginine residues and two disulfide bridges. AA139, the result of extensive lead optimization. AA139, exhibits potent *in vitro* activity against the Gram negative GAIN act pathogens and was shown to have potent *in vivo* efficacy against an ESBL producing strain of *E. coli* in the mouse urinary tract infection (UTI) model.

Method

Starting Day -4 before inoculation mice were given drinking water with 5% glucose. On day 0 mice were anaesthetized and inoculated in the bladder with 50 μ L bacterial suspension (1×10^9 CFU/ml) of *E. coli*. On day 1 and 2, mice (n=6) were treated IV with vehicle or with twice-daily administrations of 5 mg/kg AA139 or 40 mg/kg meropenem. Urine samples were taken on days 1 to 3 and on day 3 mice were sacrificed and urine, kidneys and bladder were collected for CFU counts.

Results

In the vehicle group the CFU levels in urine remained unchanged throughout the 3-day study period. At day 3, CFU levels in the bladder of the vehicle group were also in accordance with the model. Spreading of bacteria to the kidney, which in this model typically occurs in about 30-70% of mice, was 50% in this study. Overall results of the study are reported in the table:

| Mean colony counts of <i>E. coli</i> DSA 443 in mice treated with vehicle, AA139 or meropenem | | | | | |
|---|-------|--------|--------|---------|---------|
| Treatment | Urine | | | Bladder | Kidneys |
| | Day 1 | Day 2 | Day 3 | Day 3 | Day 3 |
| Vehicle | 6.61 | 5.35 | 5.70 | 5.28 | 2.25 |
| AA139 (5 mg/kg) | 6.39 | 3.00* | 1.62** | 3.62** | 1.41 |
| Meropenem (40 mg/kg) | 5.40 | 2.30** | 1.25** | 3.52** | 1.64 |

Asterisks indicate that value is significantly lower than for the vehicle group. *p<0.01; **p<0.001 (one-way ANOVA, Dunnett's multiple comparison test)

Conclusions

AA139 showed potent bioload reduction and significantly reduced the bacterial count in urine, bladder and kidneys at levels comparable to meropenem dosed at 40 mg/kg

Background

Arenicin-3 was isolated from the marine lugworm *Arenicola marina*; it contains 21 natural amino acid residues constrained in an amphipathic beta hairpin structure by two disulfide bridges between Cys3, Cys20 and Cys7, Cys16. Four positively charged arginines, and 9 hydrophobic residues contribute to the amphipathic characteristics of the peptide.

In this study we present the efficacy of clinical candidate AA139 in a first UTI infection model caused by ESBL producing *E. coli* DSA443.

Method

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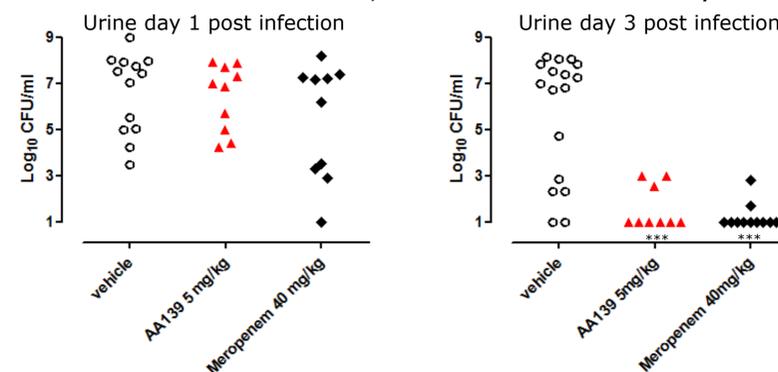


Figure 1. Bacterial load in the urine before treatment (Day 1) and after last treatment (Day 3) with AA139 or meropenem after infection with *E. coli*. ***: p<0.001

Results-Continued

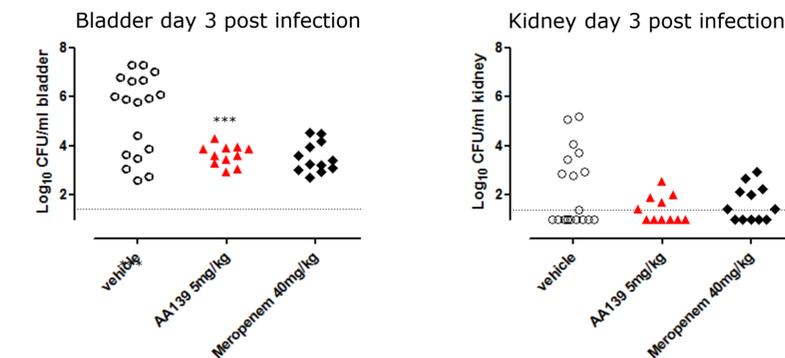


Figure 2. Bacterial counts (log10 CFU/ml) in the bladder and kidneys on Day 3 after infection with *E. coli* DSA 443 and twice daily treatment with AA139 at 5 mg/kg or meropenem at 40 mg/kg i.v. Significant; ***: p<0.001.

Conclusion

- ESBL producing strain *E. coli* DSA443 performed in this model of Urinary Tract Infection in the mouse as expected from literature with a persistent colonization of urine, bladder and kidney up to day three.
- AA139 showed potent bioload reduction and significantly reduced the bacterial count in urine, bladder and kidneys at levels comparable to meropenem dosed at 40 mg/kg