E-1328

In-vitro Activity of LTX-109 against Vancomycin Intermediate (VISA), Vancomycin Resistant (VRSA) and Mupirocin Susceptible, Low and High Level Resistant Strains of *Staphylococcus aureus*

[1]Louis D. Saravolatz, MD, [1]Joan Pawlak, BS, [1]Leonard B. Johnson, MD, [1]Mohammad G. Fakih, MD, [2]Anders Fugelli, PhD, and [2]Wenche M. Olsen, PhD

[1]St John Hospital and Medical Center, Detroit, Michigan and [2]Lytix Biopharma AS, Oslo Norway

Contact:
Louis D. Saravolatz, MD
19251 Mack Avenue, Suite 335
Grosse Pointe Woods, MI 48236
Phone: 313-343-3362
Fax: 313-343-7784
E-mail: louis.saravolatz@stjohn.org

Abstract

Background: LTX-109 is a novel synthetic topical antimicrobial peptidomimetic drug with a unique membrane lysing mode of action causing ultra –rapid membrane disruption. Its potential clinical application includes decolonization of carriers of *Staphylococcus aureus* (SA). We evaluated its activity against different strains of SA including those with different susceptibilities to mupirocin and vancomycin.

Methods: We performed microdilution susceptibility testing using Mueller-Hinton broth to determine minimum inhibitory concentrations (MIC) of LTX-109 and mupirocin against 109 MRSA (methicillin-resistant SA), 33 VISA (vancomycin intermediate SA), and13 VRSA (vancomycin resistant SA). Of these isolates there were 139 mupirocin susceptible (MS), 6 mupirocin low level resistant (MLLR) and 10 mupirocin high level resistant (MHLR) isolates. Minimal bactericidal concentrations (MBC) were also determined according to CLSI guidelines.

Results: LTX-109 demonstrated similar activity with an MIC range of 2-4 mg/L for VISA, VRSA, and mupirocin sensitive (MS), MLLR and MHLR strains.

Conclusions: LTX-109 demonstrated good activity against VISA, VRSA and mupirocin sensitive, and low and high level resistant strains of *Staphylococcus aureus*. Clinical studies are ongoing to determine its efficacy in eradication of nasal staphylococcal colonization and as a treatment for Gram positive skin infections.

Introduction

- LTX-109 is an investigational antimicrobial drug with a novel membrane-lysing mechanism of action. This action is based on the biological principle of innate immune effectors, lytic peptides. LTX-109 has rapid bactericidal lytic activity.
- Preclinical models have demonstrated that LTX-109 is bactericidal, being equally effect against wild-type as well as drug-resistant bacteria such as MRSA. Because of its unique membrane-lysing mechanism of action, there is no cross-resistance to other drugs.
- LTX-109 is in development as a treatment for bacterial skin infections and nasal decolonization of MRSA. LTX-109 has undergone a comprehensive nonclinical safety and toxicology program. There has been the successful completion of two clinical studies with LTX-109, one for nasal decolonization of MRSA/MSSA and one for treatment of Gram positive skin infections. Topical application of LTX-109 to the anterior nares or on to infected skin is tolerated and safe, with a negligible systemic uptake.

PURPOSE:

In this study we evaluated the *in vitro* activity of LTX-109 against a collection of *Staphylococcus aureus* isolates, including those with different susceptibilities to mupirocin and vancomycin.

Methods

- A collection of 155 strains of *Staphylococcus aureus* were selected for evaluation. The MRSA (n=109) strains were isolated from patients admitted to St. John Hospital and Medical Center Detroit, MI. VISA (n=33) strains and VRSA (n=13) strains were obtained through the Network on Antimicrobial Resistance in *Staphylococcus aureus* (NARSA) program: supported under NIAID/NIH contract # HHSN272200700055C.
- Microdilution tests using cation adjusted Mueller-Hinton broth were used to determine the minimal inhibitory concentration (MIC) of LTX-109 and mupirocin (MUP). MIC's were determined in accordance with Clinical and Laboratory Standard Institute (CLSI) guidelines and MIC's were read visually as the lowest drug concentration well with no visible bacterial growth. *Staphylococcus aureus* ATCC 29213 and *Enterococcus faecalis* ATCC 29212 were used to monitor quality control for the antibiotics. The minimal bactericidal concentrations (MBC) for all the isolates were determined according to CLSI guidelines.
- Isolates with a mupirocin MIC of 8 to 256 ug/ml were identified as having low-level resistance (LLR), an MIC of greater than or equal to 512ug/ml were identified as high-level resistance (HLR). Isolates with a MIC of less than or equal to 4ug/ml were identified as susceptible (S).

Results

Table 1: Activity of LTX-109 and Mupirocin against all study isolates.

All Isolates (n=155)	MIC (mg/L) Range	MIC 50	MIC 90	MBC (mg/L) Range	MBC 50	MBC 90
LTX-109	2--4	4	4	2--8	4	4
MUP	0.03-->512	0.12	8	0.5-->512	16	256
All Isolates MUP S (n=139)						
LTX-109	2--4	4	4	2--8	4	4
MUP	0.03--0.5	0.12	0.25	0.5--32	8	32
All Isolates MUP LLR (n=6)						
LTX-109	2--4	2	4	2--4	4	4
MUP	8--32	16	32	256-->512	>512	>512
All Isolates MUP HLR (n=10)						
LTX-109	2--4	4	4	4	4	4
MUP	>512	>512	>512	>512	>512	>512

Table 2: Activity of LTX-109 and Mupirocin against all MRSA isolates.

MRSA (n=109)	MIC (mg/L) Range	MIC 50	MIC 90	MBC (mg/L) Range	MBC 50	MBC 90
LTX-109	2--4	2	4	2--8	4	4
MUP	0.06-->512	0.12	0.25	4-->512	16	32
MRSA MUP S (n=102)						
LTX-109	2--4	2	4	2--8	4	4
MUP	0.06--0.25	0.12	0.25	4--32	16	32
MRSA MUP LLR (n=1)						
LTX-109	4	4	4	4	4	4
MUP	32	32	32	>512	>512	>512
MRSA MUP HLR (n=6)						
LTX-109	2--4	2	4	4	4	4
MUP	>512	>512	>512	>512	>512	>512

Results

Table 3: Activity of LTX-109 and Mupirocin against VISA isolates.

VISA (n=33)	MIC (mg/L) Range	MIC 50	MIC 90	MBC (mg/L) Range	MBC 50	MBC 90
LTX-109	2--4	4	4	2--8	4	4
MUP	0.03-->512	0.25	>512	0.5-->512	8	>512
VISA MUP S (n=27)						
LTX-109	2--4	4	4	2--8	4	4
MUP	0.03--0.5	0.25	0.25	0.5--32	8	16
VISA MUP LLR (n=2)						
LTX-109	2	2	2	2--4	2	4
MUP	16	16	16	256-->512	256	>512
VISA MUP HLR (n=4)						
LTX-109	2--4	2	4	4	4	4
MUP	>512	>512	>512	>512	>512	>512

Table 4: Activity of LTX-109 and Mupirocin against VRSA isolates.

VRSA (n=13)	MIC (mg/L) Range	MIC 50	MIC 90	MBC (mg/L) Range	MBC 50	MBC 90
LTX-109	2--4	4	4	2--4	4	4
MUP	0.06--32	0.25	16	0.5-->512	4	>512
VRSA MUP S (n=10)						
LTX-109	4	4	4	4	4	4
MUP	0.06--0.25	0.12	0.25	0.5--16	4	16
VRSA MUP LLR (n=3)						
LTX-109	2	2	2	2	2	2
MUP	8--32	16	32	>512	>512	>512

Conclusion

- LTX-109 demonstrated bactericidal activity against all isolates tested and *in vitro* activity did not vary based upon resistance to other classes of drugs including mupirocin, methicillin, vancomycin, daptomycin, or linezolid.
- The topical formulation of LTX-109 has 20,000 mcg/ml which is far in excess of the MBC's of 2-8 mcg/ml.
- LTX-109 demonstrated excellent *in vitro* bactericidal activity against *S.aureus* making it a potential candidate drug for the treatment of staphylococcal infections.
- The drug has been tested in Phase I and two Phase I/IIa trials with good tolerance, minimal systemic bioavailability and has demonstrated Proof of Concept in decolonization of nasal MRSA / MSSA. Further Phase II studies are planned to demonstrate efficacy in larger patient populations.