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Original Research Article

Antibiotic resistance and susceptibility pattern of different microorganisms against Nadifloxacin

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ABSTRACT

Background: Skin and soft tissue infections (SSTIs) are common and can have serious implications. Nadifloxacin's broad-spectrum antibiotic activity may potentially provide therapeutic benefits for skin infections. Also, it offers a viable alternative therapy for topical agent resistance.

Objective: This study investigates the antimicrobial susceptibility of few gram-positive and gram-negative micro-organisms (*S. aureus*, *S. pyogenes*, *S. epidermidis*, methicillin-resistant *S. epidermidis*, methicillin-resistant *S. aureus*, *E. faecalis*, *P. aeruginosa*, *E. coli* and *P. acne*) to four important topical antibiotics: Mupirocin, Clindamycin, Fusidic acid, and Nadifloxacin.

Methods: Antibiotic susceptibility and minimum inhibitory concentration (MIC) were determined using Kirby-Bauer disk diffusion, Epsilometer test (E-Test), and Micro-broth dilution methods. Mueller Hinton and Brucella blood agar served as growth media. HiComb strips from HiMedia were used, and QC strains were tested. Kirby-Bauer assessed Zone of Inhibition; HiComb determined MIC via gradient; Micro-broth dilution gauged growth in antibiotic-diluted broth.

Results: The disk diffusion method revealed varying resistance percentages for antibiotics. Clindamycin had the highest resistance (62%) followed by Fusidic acid (47%), Nadifloxacin (15%), and Mupirocin (5%). Among gram-positive isolates, Nadifloxacin and Mupirocin had 100% sensitivity, while Fusidic acid showed moderate resistance (19%) and Clindamycin showed highest resistance (42%). Among gram-negative isolates, Clindamycin and Fusidic acid had 100% resistance, while Nadifloxacin (42%) and Mupirocin (15%) showed comparatively low resistance. Among the 57 *Staphylococcus* species isolates, including 49 isolates of *S. aureus* and 8 isolates of *S. epidermidis*, the antibiotic susceptibility testing revealed a MIC value <4 µg/ml of Nadifloxacin, with a high level of sensitivity across all isolates.

Conclusion: Nadifloxacin's superior efficacy in the study can be attributed to its mechanism of action, targeting bacterial DNA gyrase and topoisomerase IV, making it suitable for bacterial infections, particularly those involving the skin and soft tissues. Out of four antibiotics tested, Nadifloxacin was found to be effective against both gram-positive and gram-negative strains of bacteria.

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1. Introduction

Skin and soft tissue infections (SSTIs) are prevalent and may be serious, hospitalizing 7-10% of patients globally.¹

Many topical medications are available to treat such infections having localized antibacterial action and fewer systemic side effects. They are more patient-compliant with ease of application and storage. For systemic skin Mupirocin, Clindamycin, and Fusidic acid are administered topically. However, bacterial resistance to these medications

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has increased, prompting the development of new broad-spectrum antibiotics with reduced antimicrobial resistance.² Nadifloxacin is another viable option for acne and other bacterial skin infections. Its antibiotic action targets aerobic gram-negative, gram-positive, and anaerobic bacteria. Skin infections may benefit from Nadifloxacin's broad-spectrum antibiotic action. In situations of topical agent resistance, it offers an alternate therapy. Healthcare practitioners may successfully treat bacterial infections while avoiding antimicrobial resistance with this medication.^{3,4}

Previous in vitro research on bacterial skin infections showed that Nadifloxacin is safe and effective against a range of bacteria. It's very effective against *Streptococcus* and *Propionibacterium* species. These data showed that Nadifloxacin may cure bacterial skin infections by targeting a wide spectrum of pathogens.⁵ Nadifloxacin had antibacterial action against *S. epidermidis*, *P. acnes*, MSSA, and MRSA, and none of these pathogens were resistant to Nadifloxacin, demonstrating its efficiency in reducing their growth.^{6,7} The present study examines the antimicrobial susceptibility of few gram-positive and gram-negative organisms (*S. aureus*, *S. pyogenes*, *S. epidermidis*, methicillin-resistant *S. epidermidis*, methicillin-resistant *S. aureus*, *E. faecalis*, *P. aeruginosa*, *E. coli* and *P. acne*) to four topical antibiotics: Mupirocin, Clindamycin, Fusidic acid, and Nadifloxacin.

2. Methodology

Antimicrobial susceptibility testing (AST) on aerobic and anaerobic bacteria was done using the Kirby-Bauer disk diffusion technique. The study utilized 76 isolates of different organisms. This standard method is often used to test quickly developing bacteria' antibiotic sensitivity and resistance. Most organisms grew on Mueller Hinton agar, except *P. acne*, which grew on Brucella blood agar. After overnight incubation, filter paper disks impregnated with calibrated doses of antimicrobial agents were tested for Zone of Inhibition (ZOI) size. Incubation periods were specified by the Clinical and Laboratory Standards Institute (CLSI) performance standards for antimicrobial disk susceptibility tests. The testing included quality control strains *P. aeruginosa* ATCC 27853 and *S. aureus* ATCC 25923. Epsilometer test (E-Test) and Micro-broth dilution were used to evaluate minimum inhibitory concentration (MIC) values in the research. Micro-broth dilution process entailed loading microtiter plates with broth and putting two-fold antibiotic dilutions into the wells. And dispensing bacterial isolates into the respective wells. The plates were incubated for 16–20 hours, and then they were visually examined to see whether the bacteria had grown. The Clinical and Laboratory Standard Institute investigated the antibiotic resistance profile of *Staphylococcus* species, including *S. aureus* and *S. epidermidis* isolates and fresh isolates.

Using the HiComb approach, individual bacterial strains' susceptibility or resistance was quantified. Dry chemistry and a gradient-based method were used. The apparatus included two comb-shaped strips with extensions that held antibiotic-loaded discs. On an agar plate, the discs generated a concentration gradient of the antibiotic through 16 two-fold dilutions. An oval ZOI formed on the agar surface as the antibiotic diffused from one end of the strip to the other. Where the zone met the strips' comb-like projections, the MIC was calculated. The CLSI process and this method's MIC are comparable. HiComb strips from HiMedia Laboratory Ltd. were used to cultivate diverse organisms on Mueller Hinton or Brucella blood agar. According to the manufacturer, the ZOI was tested and reported as sensitive or resistant. The testing also included quality control strains *P. aeruginosa* ATCC 27853 and *S. aureus* ATCC 25923. The concentration of antibiotics used in the experiment included 200 µg/ml of Mupirocin, 2 µg/mL of Clindamycin, 10 µg/mL of Fusidic acid, and 5 µg/ml of Nadifloxacin. The efficacy of Nadifloxacin was evaluated for fresh 25 *Staphylococcus* sp (*S. aureus*, *S. pyogenes*, *S. epidermidis*, methicillin-resistant *S. epidermidis*, *S. aureus*) and *E. faecalis*, *P. aeruginosa*, and *P. acne* isolates using the same protocol and compared against 200 mcg Mupirocin. The approach also evaluated ZOI & MIC of Nadifloxacin versus Mupirocin, Clindamycin, and Fusidic acid for different bacterial strains of MRSA, *P. acnes*, and *S. epidermidis*, which cause SSTIs.

3. Results

3.1. Disk diffusion

Table 1 depicts the findings obtained for antibiotics on the tested microorganisms. All isolates of *E. coli* (SRL 7, SRL 43, SRL 44, SRL 45, SRL 66, SRL 68, SRL 69, SRL 70, SRL 71, SRL 72, SRL 73, SRL 74, SRL 75) were sensitive to Nadifloxacin with ZOI values ranging between 9 to 57. Fusidic acid showed a high level of resistance towards SRL 7 while being resistant towards other *E. coli* isolates. However, other *E. coli* isolates demonstrated resistance or no ZOI against Clindamycin and Fusidic acid.

The *E. faecalis* isolates showed the absence of high-level resistance against a Nadifloxacin reference standard, DRL API, and high media, along with Mupirocin while it showed resistance against Clindamycin. SRL 5,13,14,20,21,26,30,31,33,36,54, and 55 were sensitive against Fusidic acid while SRL 61 was resistant.

MRSA isolates SRL 1, SRL 10, SRL 12, SRL 15, SRL 23, SRL 24, SRL 27, SRL 29, SRL 64, and SRL 65 exhibited the absence of high level of resistance to Nadifloxacin (reference and API), Nadifloxacin, and Mupirocin while it exhibited sensitivity to Clindamycin and Fusidic acid. MRSA isolates exhibited ZOI in the range of 16 to 38 for all antibiotics.

Table 1: Antibiotic susceptibility testing results

Antibiotics		Nadifloxacin (Reference Std) - 5 mcg	Nadifloxacin (DRL API) - 5 mcg	Nadifloxacin (HiMedia) - 5 mcg	Mupirocin - 200 mcg	Clindamycin – 2 mcg	Fusidic Acid - 10 mcg
Sr. No	Isolate Name	ZOI (mm)/ Resistance Interpretation	ZOI (mm)/ Resistance Interpretation	ZOI (mm)/ Resistance Interpretation	ZOI (mm)/ Resistance Interpretation	ZOI (mm)/ Resistance Interpretation	ZOI (mm)/ Resistance Interpretation
SRL 7	<i>E. coli</i>	32/ Absence of High-Level Resistance	28/ Absence of High-Level Resistance	27/ Absence of High-Level Resistance	24/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ High-Level Resistance
SRL 43	<i>E. coli</i>			24/ Absence of High-Level Resistance	23/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 44	<i>E. coli</i>			No ZOI/ High-Level Resistance	22/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 45	<i>E. coli</i>			9/ Absence of High-Level Resistance	28/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 66	<i>E. coli</i>			No ZOI/ High-Level Resistance	25/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 68	<i>E. coli</i>	No ZOI/High- Level Resistance	No ZOI/High- Level Resistance	No ZOI/ High-Level Resistance	25/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 69	<i>E. coli</i>			No ZOI/ High-Level Resistance	26/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 70	<i>E. coli</i>			No ZOI/ High-Level Resistance	25/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 71	<i>E. coli</i>			No ZOI/ High-Level Resistance	23/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 72	<i>E. coli</i>			23/ Absence of High-Level Resistance	22/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 73	<i>E. coli</i>			20/ Absence of High-Level Resistance	25/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 74	<i>E. coli</i>			15/ Absence of High-Level Resistance	23/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 75	<i>E. coli</i>			No ZOI/ High-Level Resistance	21/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 61	<i>E. faecalis</i>			16/ Absence of High-Level Resistance	18/ Absence of High-Level Resistance	No ZOI/ Resistant	19/ Resistant

SRL 5	<i>E. faecalis</i>	11/ Absence of High-Level Resistance	12/ Absence of High-Level Resistance	12/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	10/ Resistant	25/ Sensitive
SRL 13	<i>E. faecalis</i>	10/ Absence of High-Level Resistance	11/ Absence of High-Level Resistance	13/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	No ZOI/ Resistant	23/ Sensitive
SRL 14	<i>E. faecalis</i>	18/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	18/ Absence of High-Level Resistance	21/ Absence of High-Level Resistance	9/ Resistant	20/ Sensitive
SRL 20	<i>E. faecalis</i>	11/ Absence of High-Level Resistance	12/ Absence of High-Level Resistance	18/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	No ZOI/ Resistant	20/ Sensitive
SRL 21	<i>E. faecalis</i>	12/ Absence of High-Level Resistance	13/ Absence of High-Level Resistance	15/ Absence of High-Level Resistance	18/ Absence of High-Level Resistance	No ZOI/ Resistant	20/ Sensitive
SRL 26	<i>E. faecalis</i>	12/ Absence of High-Level Resistance	14/ Absence of High-Level Resistance	16/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	No ZOI/ Resistant	20/ Sensitive
SRL 30	<i>E. faecalis</i>			24/ Absence of High-Level Resistance	23/ Absence of High-Level Resistance	No ZOI/ Resistant	23/ Sensitive
SRL 31	<i>E. faecalis</i>			15/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	No ZOI/ Resistant	23/ Sensitive
SRL 33	<i>E. faecalis</i>			20/ Absence of High-Level Resistance	22/ Absence of High-Level Resistance	No ZOI/ Resistant	22/ Sensitive
SRL 36	<i>E. faecalis</i>			23/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	No ZOI/ Resistant	23/ Sensitive
SRL 54	<i>E. faecalis</i>			15/ Absence of High-Level Resistance	19/ Absence of High-Level Resistance	No ZOI/ Resistant	26/ Sensitive
SRL 55	<i>E. faecalis</i>			26/ Absence of High-Level Resistance	18/ Absence of High-Level Resistance	No ZOI/ Resistant	21/ Sensitive
SRL 6	<i>P. aeruginosa</i>	12/ Absence of High-Level Resistance	15/ Absence of High-Level Resistance	13/ Absence of High-Level Resistance	24/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ High-Level Resistance
SRL 8	<i>P. aeruginosa</i>	No ZOI/ High-Level Resistance	No ZOI/ High-Level Resistance	No ZOI/ High-Level Resistance	No ZOI/ High-Level Resistance	No ZOI/ Resistant	No ZOI/ High-Level Resistance
SRL 17	<i>P. aeruginosa</i>	No ZOI/ High-Level Resistance	No ZOI/ High-Level Resistance	No ZOI/ High-Level Resistance	No ZOI/ High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant

SRL 18	<i>P. aeruginosa</i>	15/ Absence of High-Level Resistance	16/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	23/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 19	<i>P. aeruginosa</i>	14/ Absence of High-Level Resistance	14/ Absence of High-Level Resistance	22/ Absence of High-Level Resistance	23/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 25	<i>P. aeruginosa</i>	12/ Absence of High-Level Resistance	14/ Absence of High-Level Resistance	18/ Absence of High-Level Resistance	23/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 32	<i>P. aeruginosa</i>			21/ Absence of High-Level Resistance	27/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 34	<i>P. aeruginosa</i>			21/ Absence of High-Level Resistance	26/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 37	<i>P. aeruginosa</i>			No ZOI/ High-Level Resistance	18/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 38	<i>P. aeruginosa</i>			23/ Absence of High-Level Resistance	25/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 39	<i>P. aeruginosa</i>			No ZOI/ High-Level Resistance	No ZOI/ High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 47	<i>P. aeruginosa</i>			18/ Absence of High-Level Resistance	No ZOI/ High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 28	<i>P. aeruginosa</i>	16/ Absence of High-Level Resistance	15/ Absence of High-Level Resistance	22/ Absence of High-Level Resistance	25/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 1	<i>S. aureus</i> (MRSA)	20/ Absence of High-Level Resistance	21/ Absence of High-Level Resistance	21/ Absence of High-Level Resistance	32/ Absence of High-Level Resistance	32/ Sensitive	30/ Sensitive
SRL 10	<i>S. aureus</i> (MRSA)	22/ Absence of High-Level Resistance	24/ Absence of High-Level Resistance	22/ Absence of High-Level Resistance	38/ Absence of High-Level Resistance	28/ Sensitive	38/ Sensitive
SRL 12	<i>S. aureus</i> (MRSA)	18/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	24/ Absence of High-Level Resistance	34/ Absence of High-Level Resistance	30/ Sensitive	28/ Sensitive
SRL 15	<i>S. aureus</i> (MRSA)	21/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	23/ Absence of High-Level Resistance	30/ Absence of High-Level Resistance	28/ Sensitive	27/ Sensitive
SRL 23	<i>S. aureus</i> (MRSA)	18/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	23/ Absence of High-Level Resistance	30/ Absence of High-Level Resistance	28/ Sensitive	29/ Sensitive

SRL 24	<i>S. aureus</i> (MRSA)	19/ Absence of High-Level Resistance	18/ Absence of High-Level Resistance	22/ Absence of High-Level Resistance	30/ Absence of High-Level Resistance	23/ Sensitive	16/ Resistant
SRL 27	<i>S. aureus</i> (MRSA)	16/ Absence of High-Level Resistance	17/ Absence of High-Level Resistance	29/ Absence of High-Level Resistance	30/ Absence of High-Level Resistance	25/ Sensitive	30/ Sensitive
SRL 29	<i>S. aureus</i> (MRSA)	17/ Absence of High-Level Resistance	17/ Absence of High-Level Resistance	23/ Absence of High-Level Resistance	32/ Absence of High-Level Resistance	27/ Sensitive	25/ Sensitive
SRL 50	<i>S. aureus</i> (MRSA)			21/ Absence of High-Level Resistance	31/ Absence of High-Level Resistance	27/ Sensitive	26/ Sensitive
SRL 51	<i>S. aureus</i> (MRSA)			21/ Absence of High-Level Resistance	33/ Absence of High-Level Resistance	28/ Sensitive	22/ Sensitive
SRL 52	<i>S. aureus</i> (MRSA)			23/ Absence of High-Level Resistance	33/ Absence of High-Level Resistance	29/ Sensitive	28/ Sensitive
SRL 64	<i>S. aureus</i> (MRSA)			23/ Absence of High-Level Resistance	26/ Absence of High-Level Resistance	25/ Sensitive	26/ Sensitive
SRL 65	<i>S. aureus</i> (MRSA)			20/ Absence of High-Level Resistance	27/ Absence of High-Level Resistance	24/ Sensitive	25/ Sensitive
SRL 35	<i>S. aureus</i> (MSSA)			25/ Absence of High-Level Resistance	34/ Absence of High-Level Resistance	28/ Sensitive	29/ Sensitive
SRL 41	<i>S. aureus</i> (MSSA)			20/ Absence of High-Level Resistance	30/ Absence of High-Level Resistance	26/ Sensitive	27/ Sensitive
SRL 42	<i>S. aureus</i> (MSSA)			31/ Absence of High-Level Resistance	32/ Absence of High-Level Resistance	27/ Sensitive	28/ Sensitive
SRL 46	<i>S. aureus</i> (MSSA)			21/ Absence of High-Level Resistance	31/ Absence of High-Level Resistance	26/ Sensitive	16/ Resistant
SRL 2	<i>S. aureus</i> (MSSA)	20/ Absence of High-Level Resistance	21/ Absence of High-Level Resistance	22/ Absence of High-Level Resistance	40/ Absence of High-Level Resistance	No ZOI/ Resistant	34/ Sensitive
SRL 3	<i>S. aureus</i> (MSSA)	20/ Absence of High-Level Resistance	22/ Absence of High-Level Resistance	23/ Absence of High-Level Resistance	40/ Absence of High-Level Resistance	No ZOI/ Resistant	36/ Sensitive
SRL 4	<i>S. aureus</i> (MSSA)	23/ Absence of High-Level Resistance	24/ Absence of High-Level Resistance	25/ Absence of High-Level Resistance	41/ Absence of High-Level Resistance	30/ Sensitive	16/ Resistant
SRL 9	<i>S. aureus</i> (MSSA)	22/ Absence of High-Level Resistance	19/ Absence of High-Level Resistance	21/ Absence of High-Level Resistance	38/ Absence of High-Level Resistance	32/ Sensitive	38/ Sensitive

SRL 11	<i>S. aureus</i> (MSSA)	16/ Absence of High-Level Resistance	18/ Absence of High-Level Resistance	22/ Absence of High-Level Resistance	36/ Absence of High-Level Resistance	32/ Sensitive	31/ Sensitive
SRL 16	<i>S. aureus</i> (MSSA)	18/ Absence of High-Level Resistance	18/ Absence of High-Level Resistance	21/ Absence of High-Level Resistance	31/ Absence of High-Level Resistance	27/ Sensitive	26/ Sensitive
SRL 22	<i>S. aureus</i> (MSSA)	19/ Absence of High-Level Resistance	18/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	30/ Absence of High-Level Resistance	27/ Sensitive	28/ Sensitive
SRL 53	<i>S. aureus</i> (MSSA)			31/ Absence of High-Level Resistance	32/ Absence of High-Level Resistance	30/ Sensitive	28/ Sensitive
SRL 59	<i>S. aureus</i> (MSSA)			36/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	31/ Sensitive	14/ Resistant
SRL 67	<i>S. aureus</i> (MSSA)			20/ Absence of High-Level Resistance	32/ Absence of High-Level Resistance	25/ Sensitive	25/ Sensitive
SRL 48	<i>S.</i> <i>epidermidis</i>			20/ Absence of High-Level Resistance	23/ Absence of High-Level Resistance	No ZOI/ Resistant	No ZOI/ Resistant
SRL 56	<i>S.</i> <i>epidermidis</i>			38/ Absence of High-Level Resistance	22/ Absence of High-Level Resistance	No ZOI/ Resistant	16/ Resistant
SRL 57	<i>S.</i> <i>epidermidis</i>			38/ Absence of High-Level Resistance	23/ Absence of High-Level Resistance	9/ Resistant	15/ Resistant
SRL 58	<i>S.</i> <i>epidermidis</i>			27/ Absence of High-Level Resistance	11/ Absence of High-Level Resistance	No ZOI/ Resistant	12/ Resistant
SRL 62	<i>S.</i> <i>epidermidis</i>	36/ Absence of High-Level Resistance	40/ Absence of High-Level Resistance	40/ Absence of High-Level Resistance	40/ Absence of High-Level Resistance	30/ Sensitive	32/ Sensitive
SRL 63	<i>S.</i> <i>epidermidis</i>			41/ Absence of High-Level Resistance	40/ Absence of High-Level Resistance	30/ Sensitive	32/ Sensitive
SRL 40	<i>S.</i> <i>epidermidis</i>			39/ Absence of High-Level Resistance	37/ Absence of High-Level Resistance	29/ Sensitive	31/ Sensitive
SRL 49	<i>S.</i> <i>epidermidis</i> (MRSE)	36/ Absence of High-Level Resistance	40/ Absence of High-Level Resistance	40/ Absence of High-Level Resistance	13/ Absence of High-Level Resistance	No ZOI/ Resistant	32/ Sensitive
SRL 60	<i>S. pyogenes</i>	22/ Absence of High-Level Resistance	20/ Absence of High-Level Resistance	18/ Absence of High-Level Resistance	28/ Absence of High-Level Resistance	21/ Absence of High-Level Resistance	12/ Resistant
SRL 76	<i>P. acnes</i>			>40/ Absence of High-Level Resistance	>40/ Absence of High-Level Resistance	>40/ Absence of High-Level Resistance	>40/ Absence of High-Level Resistance

MRSA: Methicillin-resistant *Staphylococcus aureus*; MSSA: Methicillin-Sensitive *Staphylococcus aureus*; MRSE: Methicillin-resistant *Staphylococcus epidermidis*; MIC: Minimum Inhibitory Concentration; ZOI: Zone of Inhibition

Table 2: MIC values for various antibiotics against different bacterial isolates

Antibiotics		Mupirocin	Clindamycin	Fusidic Acid	Nadifloxacin	Nadifloxacin
Sr. No	Isolate Name	MIC Conc	MIC Conc	MIC Conc	Reference MIC Conc ($\mu\text{g/ml}$)	- DRL MIC Conc ($\mu\text{g/ml}$)
SRL 7	<i>E. coli</i>	60	60	60	>32	>32
SRL43	<i>E. coli</i>	30	2	>240	4	2
SRL44	<i>E. coli</i>	120	120	>240	>32	>32
SRL45	<i>E. coli</i>	60	4	>240	>32	>32
SRL66	<i>E. coli</i>	120	120	240	>32	>32
SRL68	<i>E. coli</i>	120	120	>240	>32	>32
SRL69	<i>E. coli</i>	120	120	>240	>32	>32
SRL70	<i>E. coli</i>	60	60	>240	>32	>32
SRL71	<i>E. coli</i>	30	30	>240	>32	>32
SRL72	<i>E. coli</i>	60	60	>240	>32	>32
SRL73	<i>E. coli</i>	60	60	>240	4	2
SRL74	<i>E. coli</i>	30	30	>240	8	4
SRL75	<i>E. coli</i>	120	120	>240	>32	>32
SRL 5	<i>E. faecalis</i>	>240	>240	>240	16	16
SRL 13	<i>E. faecalis</i>	60	>240	0.001	16	16
SRL 14	<i>E. faecalis</i>	10	>240	0.001	0.5	0.25
SRL 20	<i>E. faecalis</i>	60	120	0.001	8	32
SRL 21	<i>E. faecalis</i>	10	>240	0.001	4	4
SRL 26	<i>E. faecalis</i>	30	120	>240	4	4
SRL30	<i>E. faecalis</i>	30	30	0.001	4	4
SRL31	<i>E. faecalis</i>	30	30	1	4	4
SRL33	<i>E. faecalis</i>	60	120	0.001	2	1
SRL36	<i>E. faecalis</i>	30	30	0.001	0.5	0.5
SRL54	<i>E. faecalis</i>	60	120	0.001	0.25	0.5
SRL55	<i>E. faecalis</i>	60	5	0.001	4	4
SRL61	<i>E. faecalis</i>	120	120	0.001	4	4
SRL 6	<i>P. aeruginosa</i>	>240	60	>240	>32	>32
SRL 8	<i>P. aeruginosa</i>	>240	>240	>240	>32	>32
SRL 17	<i>P. aeruginosa</i>	>240	>240	>240	> 32	> 32
SRL 18	<i>P. aeruginosa</i>	>240	>240	>240	4	2
SRL 19	<i>P. aeruginosa</i>	60	>240	>240	4	2
SRL 25	<i>P. aeruginosa</i>	>240	>240	>240	4	4
SRL32	<i>P. aeruginosa</i>	>240	>240	>240	4	2
SRL34	<i>P. aeruginosa</i>	>240	>240	>240	8	4
SRL37	<i>P. aeruginosa</i>	>240	>240	>240	> 32	>32
SRL38	<i>P. aeruginosa</i>	>240	>240	>240	4	4
SRL39	<i>P. aeruginosa</i>	>240	>240	>240	>32	>32
SRL47	<i>P. aeruginosa</i>	>240	>240	>240	4	4
SRL 28	<i>P. aeruginosa</i>	>240	>240	>240	2	2
SRL 1	<i>S. aureus</i> (MRSA)	0.01	<0.001	0.01	1	1
SRL 10	<i>S. aureus</i> (MRSA)	0.01	0.01	0.01	1	1
SRL 12	<i>S. aureus</i> (MRSA)	0.01	0.01	0.001	1	1
SRL 15	<i>S. aureus</i> (MRSA)	0.01	0.001	0.001	1	0.5

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Table 2 continued

SRL 23	<i>S. aureus</i> (MRSA)	0.01	0.001	0.001	1	0.5
SRL 24	<i>S. aureus</i> (MRSA)	0.01	0.001	0.001	1	1
SRL 27	<i>S. aureus</i> (MRSA)	0.01	0.01	0.001	4	2
SRL 29	<i>S. aureus</i> (MRSA)	0.01	0.001	0.001	1	0.5
SRL50	<i>S. aureus</i> (MRSA)	0.01	0.01	0.001	< 0.0625	< 0.0625
SRL51	<i>S. aureus</i> (MRSA)	0.01	0.01	0.001	1	0.5
SRL52	<i>S. aureus</i> (MRSA)	0.01	0.01	0.001	1	0.5
SRL64	<i>S. aureus</i> (MRSA)	0.01	0.01	0.001	<0.0625	<0.0625
SRL65	<i>S. aureus</i> (MRSA)	0.001	0.001	0.001	>32	>32
SRL41	<i>S. aureus</i> (MRSA)	0.01	0.01	0.01	2	1
SRL42	<i>S. aureus</i> (MRSA)	0.01	0.001	0.01	<0.0625	<0.0625
SRL46	<i>S. aureus</i> (MRSA)	0.01	0.001	0.001	16	32
SRL35	<i>S. aureus</i> (MRSA)	0.01	0.001	0.01	1	0.5
SRL 2	<i>S. aureus</i> (MRSA)	0.01	>240	0.01	1	1
SRL 3	<i>S. aureus</i> (MRSA)	0.01	>240	0.01	1	1
SRL 4	<i>S. aureus</i> (MRSA)	0.01	<0.001	0.01	1	1
SRL 9	<i>S. aureus</i> (MRSA)	0.01	<0.001	0.01	1	1
SRL 11	<i>S. aureus</i> (MRSA)	0.01	0.01	0.001	1	1
SRL 16	<i>S. aureus</i> (MRSA)	0.01	0.001	0.001	1	0.5
SRL 22	<i>S. aureus</i> (MRSA)	0.01	0.001	0.001	1	1
SRL53	<i>S. aureus</i> (MRSA)	0.001	0.01	0.001	1	1
SRL59	<i>S. aureus</i> (MRSA)	0.001	0.01	0.001	1	1
SRL67	<i>S. aureus</i> (MRSA)	0.001	0.001	0.001	2	2
SRL48	<i>S. epidermidis</i>	7.5	2	1	1	1
SRL56	<i>S. epidermidis</i>	>240	2	0.001	1	0.5
SRL57	<i>S. epidermidis</i>	>240	2	0.001	1	0.5
SRL58	<i>S. epidermidis</i>	>240	>240	0.001	<0.0625	<0.0625
SRL62	<i>S. epidermidis</i>	0.001	0.001	0.001	0.5	0.25
SRL63	<i>S. epidermidis</i>	0.001	0.001	0.001	<0.0625	<0.0625

Continued on next page

Table 2 continued

SRL40	<i>S. epidermidis</i>	>240	>240	0.001	0.5	0.5
SRL49	<i>S. epidermidis</i> (MRSE)	5	0.1	0.01	< 0.0625	< 0.0625
SRL60	<i>S. pyogenes</i>	0.01	0.001	0.001	0.25	0.25
SRL76	<i>P. acnes</i>	0.001	0.001	0.001	< 0.0625	< 0.0625

MRSA: Methicillin-resistant *Staphylococcus aureus*; MSSA: Methicillin-Sensitive *Staphylococcus aureus*; MRSE: Methicillin resistant *Staphylococcus epidermidis*; MIC: Minimum Inhibitory Concentration

MSSA isolates SRL 35, SRL 41, SRL 42, SRL 46, SRL 2, SRL 3, SRL 4, SRL 9, SRL 11, SRL 16 exhibited absence of high level of resistance to Nadifloxacin (reference and API), and Mupirocin while it exhibited sensitivity to Clindamycin and Fusidic acid except SRL 2 and SRL 3, which showed resistance to Clindamycin while SRL 4, 46, and 59 showed resistances to Fusidic acid. The ZOI exhibited was in the range of 14 to 41 for all antibiotics.

S. epidermidis isolates SRL 48, 56, 57, 58, and 49 were resistant to Clindamycin, and SRL 62, 63, and 40 were sensitive to it. Similarly, SRL 48, 56, 57, and 58 were resistant to Fusidic acid while SRL 62, 63, 40, and 49 were sensitive to it. *S. epidermidis* isolates exhibited ZOI in the range of 11 to 41 for all antibiotics. SRL 62 and 49 show the absence of high-level resistance against Nadifloxacin reference and DRL API, while other *S. epidermidis* isolates show the absence of high-level resistance against Nadifloxacin high media and Mupirocin.

S. pyogenes isolates SRL 60 showed absences of the high level of resistance to all antibiotics except Fusidic acid with ZOI ranging from 12 for Fusidic acid to 28 for Mupirocin. On the other hand, *P. acnes* exhibited an absence of high-level resistance towards Nadifloxacin high media Mupirocin, Clindamycin, and Fusidic acid.

3.2. MIC Results

Table 2 provides the MIC values for various antibiotics against different bacterial isolates. For Mupirocin, it was observed that 60% of gram-positive isolates showed a MIC value of less than 4 µg/ml, indicating moderate sensitivity. However, some gram-negative isolates exhibited a high level of resistance with MIC values exceeding 240 µg/ml. Clindamycin demonstrated moderate sensitivity against gram-positive isolates, with 58% of isolates showing a MIC value of less than 0.5 µg/ml. Among gram-negative isolates, 92% showed a MIC value greater than 60 µg/ml, indicating very high resistance. Fusidic acid exhibited the lowest MIC values (< 0.5 µg/ml) for 96% of gram-positive isolates, indicating strong efficacy against this group. However, all gram-negative isolates showed a MIC value greater than 240 µg/ml, indicating complete resistance. Nadifloxacin demonstrated a MIC value of less than 4 µg/ml for 70% of all isolates, indicating a high level of sensitivity. The results suggest that Mupirocin and Clindamycin have moderate effectiveness against gram-positive isolates but are less effective against gram-negative isolates. Fusidic acid shows excellent efficacy against gram-positive isolates but is ineffective against gram-negative isolates. Nadifloxacin demonstrated a high level of sensitivity across all isolates.

3.3. Disk Diffusion and MIC Results from the Extension Study

Among the 57 *Staphylococcus* species isolates, including 49 isolates of *S. aureus* and 8 isolates of *S. epidermidis*, the antibiotic susceptibility testing revealed that all isolates (100%) showed sensitivity to Nadifloxacin as determined by both the disk diffusion method and broth dilution method, with MIC value <4 µg/ml.

On the other hand, 95% of the isolates were sensitive to Mupirocin as determined by the disk diffusion method. However, when tested with the E-Test, a slightly lower sensitivity of 88% (MIC < 4 µg/ml) was observed, indicating sensitivity. Four isolates showed discordant results, with high MIC values but sensitivity observed by the disk diffusion method using a Mupirocin disk concentration of 200 µg. This discordance could be attributed to the presence of low-level resistance to Mupirocin.

The disk diffusion method showed 88% sensitivity to Fusidic acid. The MIC values for 89% of the isolates were low, below 1.0 µg/ml, and borderline (1.0 µg/ml) for the remaining 11% of isolates, indicating sensitivity. The correlation between the disk diffusion and broth MIC results was 79%. Among the 11 discordant isolates, four showed borderline MIC values, while six exhibited borderline zone sizes between 12-16 cm by the disk diffusion method. The correlation between the disk diffusion method and E-Test results was 96% for MIC values below 0.5 µg/ml. Two isolates showed discordant results, which could be attributed to procedural bias. Additionally, one isolate of *S. epidermidis* was found to be resistant to the tested antibiotics except Nadifloxacin, for which it was sensitive with a MIC value below 0.0625 µg/ml.

Regarding resistance rates, Clindamycin exhibited the highest resistance, with 12% of the isolates being resistant according to both the disk diffusion method and E-Test (MIC > 0.5 µg/ml). Fusidic acid showed resistance in 11% of the isolates according to the disk diffusion method, and 12% resistance according to the E-Test (MIC > 1 µg/ml). Mupirocin demonstrated a resistance rate of 5% based on the disk diffusion method and 12% based on the E-Test (MIC < 4 µg/ml). Although only 5% of methicillin-resistant isolates demonstrated high-level resistance to Mupirocin by disc diffusion technique, all these isolates were susceptible to Nadifloxacin, demonstrating superiority.

Overall, Nadifloxacin demonstrated excellent sensitivity, Mupirocin showed good sensitivity with some discordant results possibly due to low-level resistance, and Fusidic acid exhibited a high sensitivity rate with a few isolates showing borderline MIC values (Figure 1-10 of Table 3 and (Figures 11 and 12 of Table 4).

Antibiotics		Nadifloxacin (HiMedia) – 5 mcg		Mupirocin - 200 mcg	
Sr. No	Isolate Name	ZOI (mm)	Resistance Interpretation	ZOI (mm)	Resistance Interpretation
SRL 1	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	32	Absence of High-Level Resistance
SRL 10	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	38	Absence of High-Level Resistance
SRL 12	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	34	Absence of High-Level Resistance
SRL 15	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	30	Absence of High-Level Resistance
SRL 23	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	30	Absence of High-Level Resistance
SRL 24	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	30	Absence of High-Level Resistance
SRL 29	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	32	Absence of High-Level Resistance
SRL50	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	31	Absence of High-Level Resistance
SRL51	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	33	Absence of High-Level Resistance
SRL52	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	33	Absence of High-Level Resistance
SRL64	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	26	Absence of High-Level Resistance
SRL35	<i>S. aureus</i> (MSSA)	25	Absence of High-Level Resistance	34	Absence of High-Level Resistance
SRL41	<i>S. aureus</i> (MSSA)	20	Absence of High-Level Resistance	30	Absence of High-Level Resistance
SRL42	<i>S. aureus</i> (MSSA)	31	Absence of High-Level Resistance	32	Absence of High-Level Resistance
SRL 2	<i>S. aureus</i> (MSSA)	22	Absence of High-Level Resistance	40	Absence of High-Level Resistance
SRL 3	<i>S. aureus</i> (MSSA)	23	Absence of High-Level Resistance	40	Absence of High-Level Resistance
SRL 4	<i>S. aureus</i> (MSSA)	25	Absence of High-Level Resistance	41	Absence of High-Level Resistance

Fig. 1: Disk diffusion results from the extension study (Table 3)

SRL 9	<i>S. aureus</i> (MSSA)	21	Absence of High-Level Resistance	38	Absence of High-Level Resistance
SRL 11	<i>S. aureus</i> (MSSA)	22	Absence of High-Level Resistance	36	Absence of High-Level Resistance
SRL 16	<i>S. aureus</i> (MSSA)	21	Absence of High-Level Resistance	31	Absence of High-Level Resistance
SRL 22	<i>S. aureus</i> (MSSA)	20	Absence of High-Level Resistance	30	Absence of High-Level Resistance
SRL53	<i>S. aureus</i> (MSSA)	31	Absence of High-Level Resistance	32	Absence of High-Level Resistance
SRL59	<i>S. aureus</i> (MSSA)	36	Absence of High-Level Resistance	20	Absence of High-Level Resistance
SRL67	<i>S. aureus</i> (MSSA)	20	Absence of High-Level Resistance	32	Absence of High-Level Resistance
SRL48	<i>S. epidermidis</i>	20	Absence of High-Level Resistance	23	Absence of High-Level Resistance
SRL56	<i>S. epidermidis</i>	38	Absence of High-Level Resistance	22	Absence of High-Level Resistance
SRL57	<i>S. epidermidis</i>	38	Absence of High-Level Resistance	23	Absence of High-Level Resistance
SRL58	<i>S. epidermidis</i>	27	Absence of High-Level Resistance	11	Resistant
SRL62	<i>S. epidermidis</i>	40	Absence of High-Level Resistance	40	Absence of High-Level Resistance
SRL63	<i>S. epidermidis</i>	41	Absence of High-Level Resistance	40	Absence of High-Level Resistance
SRL40	<i>S. epidermidis</i>	39	Absence of High-Level Resistance	37	Absence of High-Level Resistance
SRL49	<i>S. epidermidis</i> (MRSE)	40	Absence of High-Level Resistance	13	Resistant
SRL76	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	32	Absence of High-Level Resistance
Fig 1: Con.....					

Fig. 2: Cont..(Disk diffusion results from the extension study Table 3)

SRL77	<i>S. aureus</i> (MSSA)	21	Absence of High-Level Resistance	36	Absence of High-Level Resistance
SRL78	<i>S. aureus</i> (MRSA)	25	Absence of High-Level Resistance	38	Absence of High-Level Resistance
SRL79	<i>S. aureus</i> (MSSA)	29	Absence of High-Level Resistance	35	Absence of High-Level Resistance
SRL80	<i>S. aureus</i> (MSSA)	29	Absence of High-Level Resistance	35	Absence of High-Level Resistance
SRL81	<i>S. aureus</i> (MSSA)	22	Absence of High-Level Resistance	32	Absence of High-Level Resistance
SRL82	<i>S. aureus</i> (MRSA)	27	Absence of High-Level Resistance	35	Absence of High-Level Resistance
SRL83	<i>S. aureus</i> (MRSA)	27	Absence of High-Level Resistance	37	Absence of High-Level Resistance
SRL84	<i>S. aureus</i> (MRSA)	25	Absence of High-Level Resistance	34	Absence of High-Level Resistance
SRL85	<i>S. aureus</i> (MRSA)	25	Absence of High-Level Resistance	35	Absence of High-Level Resistance
SRL86	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	31	Absence of High-Level Resistance
SRL88	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	35	Absence of High-Level Resistance
SRL89	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	34	Absence of High-Level Resistance
SRL90	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	35	Absence of High-Level Resistance
SRL91	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	35	Absence of High-Level Resistance
SRL92	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	34	Absence of High-Level Resistance
SRL93	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	35	Absence of High-Level Resistance
Fig 1: Cont...					

Fig. 3: Cont..(Disk diffusion results from the extension study Table 3)

SRL94	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	32	Absence of High-Level Resistance
SRL95	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	31	Absence of High-Level Resistance
SRL96	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	31	Absence of High-Level Resistance
SRL97	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	30	Absence of High-Level Resistance
SRL98	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	No ZOI	Resistant
SRL99	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	33	Absence of High-Level Resistance
SRL100	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	32	Absence of High-Level Resistance
SRL101	<i>S. aureus</i> (MRSA)	35	Absence of High-Level Resistance	32	Absence of High-Level Resistance
Antibiotics		Nadifloxacin (Himedia) - mcg		Clindamycin - 2 mcg	
Sr. No	Isolate Name	ZOI (mm)	Resistance Interpretation	ZOI (mm)	Resistance Interpretation
SRL 1	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	32	Sensitive
SRL10	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	28	Sensitive
SRL12	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	30	Sensitive
SRL15	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	28	Sensitive
SRL23	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	28	Sensitive
SRL24	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	23	Sensitive
SRL29	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	27	Sensitive
Cont..					

Fig. 4: Cont..(Disk diffusion results from the extension study Table 3)

SRL50	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	27	Sensitive
SRL51	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	28	Sensitive
SRL52	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	29	Sensitive
SRL64	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	25	Sensitive
SRL35	<i>S. aureus</i> (MSSA)	25	Absence of High-Level Resistance	28	Sensitive
SRL41	<i>S. aureus</i> (MSSA)	20	Absence of High-Level Resistance	26	Sensitive
SRL42	<i>S. aureus</i> (MSSA)	31	Absence of High-Level Resistance	27	Sensitive
SRL 2	<i>S. aureus</i> (MSSA)	22	Absence of High-Level Resistance	No ZOI	Resistant
SRL 3	<i>S. aureus</i> (MSSA)	23	Absence of High-Level Resistance	No ZOI	Resistant
SRL 4	<i>S. aureus</i> (MSSA)	25	Absence of High-Level Resistance	30	Sensitive
SRL 9	<i>S. aureus</i> (MSSA)	21	Absence of High-Level Resistance	32	Sensitive
SRL11	<i>S. aureus</i> (MSSA)	22	Absence of High-Level Resistance	32	Sensitive
SRL16	<i>S. aureus</i> (MSSA)	21	Absence of High-Level Resistance	27	Sensitive
SRL22	<i>S. aureus</i> (MSSA)	20	Absence of High-Level Resistance	27	Sensitive
SRL53	<i>S. aureus</i> (MSSA)	31	Absence of High-Level Resistance	30	Sensitive
SRL59	<i>S. aureus</i> (MSSA)	36	Absence of High-Level Resistance	31	Sensitive
SRL67	<i>S. aureus</i> (MSSA)	20	Absence of High-Level Resistance	25	Sensitive
SRL48	<i>S. epidermidis</i>	20	Absence of High-Level Resistance	No ZOI	Resistant
SRL56	<i>S. epidermidis</i>	38	Absence of High-Level Resistance	No ZOI	Resistant

Fig. 5: Cont..(Disk diffusion results from the extension study Table 3)

SRL56	<i>S. epidermidis</i>	38	Absence of High-Level Resistance	No ZOI	Resistant
SRL57	<i>S. epidermidis</i>	38	Absence of High-Level Resistance	9	Resistant
SRL58	<i>S. epidermidis</i>	27	Absence of High-Level Resistance	No ZOI	Resistant
SRL62	<i>S. epidermidis</i>	40	Absence of High-Level Resistance	30	Sensitive
SRL63	<i>S. epidermidis</i>	41	Absence of High-Level Resistance	30	Sensitive
SRL40	<i>S. epidermidis</i>	39	Absence of High-Level Resistance	29	Sensitive
SRL49	<i>S. epidermidis</i> (MRSE)	40	Absence of High-Level Resistance	No ZOI	Resistant
SRL76	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	28	Sensitive
SRL77	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	27	Sensitive
SRL78	<i>S. aureus</i> (MRSA)	25	Absence of High-Level Resistance	32	Sensitive
SRL79	<i>S. aureus</i> (MRSA)	29	Absence of High-Level Resistance	27	Sensitive
SRL80	<i>S. aureus</i> (MRSA)	29	Absence of High-Level Resistance	30	Sensitive
SRL81	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	25	Sensitive
SRL82	<i>S. aureus</i> (MRSA)	27	Absence of High-Level Resistance	27	Sensitive
SRL83	<i>S. aureus</i> (MRSA)	27	Absence of High-Level Resistance	31	Sensitive
SRL84	<i>S. aureus</i> (MRSA)	25	Absence of High-Level Resistance	29	Sensitive
SRL85	<i>S. aureus</i> (MRSA)	25	Absence of High-Level Resistance	30	Sensitive
SRL86	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	29	Sensitive
SRL88	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	32	Sensitive
SRL89	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	30	Sensitive

Fig. 6: Cont.. (Disk diffusion results from the extension study Table 3)

SRL90	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	30	Sensitive
SRL91	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	28	Sensitive
SRL92	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	28	Sensitive
SRL93	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	28	Sensitive
SRL94	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	29	Sensitive
SRL95	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	28	Sensitive
SRL96	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	29	Sensitive
SRL97	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	27	Sensitive
SRL98	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	26	Sensitive
SRL99	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	28	Sensitive
SRL100	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	26	Sensitive
SRL101	<i>S. aureus</i> (MRSA)	35	Absence of High-Level Resistance	27	Sensitive
Antibiotics		Nadifloxacin (HiMedia) – mcg	Fusidic Acid – 10 mcg		
Sr. No	Isolate Name	ZOI (mm)	Resistance Interpretation	ZOI (mm)	Resistance Interpretation
SRL 1	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	30	Sensitive
SRL10	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	38	Sensitive
SRL12	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	28	Sensitive
SRL15	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	27	Sensitive
SRL23	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	29	Sensitive

Fig. 7: Cont.. (Disk diffusion results from the extension study Table 3)

SRL24	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	16	Resistant
SRL29	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	25	Sensitive
SRL50	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	26	Sensitive
SRL51	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	22	Sensitive
SRL52	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	28	Sensitive
SRL64	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	26	Sensitive
SRL35	<i>S. aureus</i> (MSSA)	25	Absence of High-Level Resistance	29	Sensitive
SRL41	<i>S. aureus</i> (MSSA)	20	Absence of High-Level Resistance	27	Sensitive
SRL42	<i>S. aureus</i> (MSSA)	31	Absence of High-Level Resistance	28	Sensitive
SRL 2	<i>S. aureus</i> (MSSA)	22	Absence of High-Level Resistance	34	Sensitive
SRL 3	<i>S. aureus</i> (MSSA)	23	Absence of High-Level Resistance	36	Sensitive
SRL 4	<i>S. aureus</i> (MSSA)	25	Absence of High-Level Resistance	16	Resistant
SRL 9	<i>S. aureus</i> (MSSA)	21	Absence of High-Level Resistance	38	Sensitive
SRL 11	<i>S. aureus</i> (MSSA)	22	Absence of High-Level Resistance	31	Sensitive
SRL 16	<i>S. aureus</i> (MSSA)	21	Absence of High-Level Resistance	26	Sensitive
SRL 22	<i>S. aureus</i> (MSSA)	20	Absence of High-Level Resistance	28	Sensitive
SRL53	<i>S. aureus</i> (MSSA)	31	Absence of High-Level Resistance	28	Sensitive
SRL59	<i>S. aureus</i> (MSSA)	36	Absence of High-Level Resistance	14	Resistant
Cont..					

Fig. 8: Cont.. (Disk diffusion results from the extension study Table 3)

SRL67	<i>S. aureus</i> (MSSA)	20	Absence of High-Level Resistance	25	Sensitive
SRL48	<i>S. epidermidis</i>	20	Absence of High-Level Resistance	No ZOI	Resistant
SRL56	<i>S. epidermidis</i>	38	Absence of High-Level Resistance	16	Resistant
SRL57	<i>S. epidermidis</i>	38	Absence of High-Level Resistance	15	Resistant
SRL58	<i>S. epidermidis</i>	27	Absence of High-Level Resistance	12	Resistant
SRL62	<i>S. epidermidis</i>	40	Absence of High-Level Resistance	32	Sensitive
SRL63	<i>S. epidermidis</i>	41	Absence of High-Level Resistance	32	Sensitive
SRL40	<i>S. epidermidis</i>	39	Absence of High-Level Resistance	31	Sensitive
SRL49	<i>S. epidermidis</i> (MRSE)	40	Absence of High-Level Resistance	32	Sensitive
SRL76	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	29	Sensitive
SRL77	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	30	Sensitive
SRL78	<i>S. aureus</i> (MRSA)	25	Absence of High-Level Resistance	32	Sensitive
SRL79	<i>S. aureus</i> (MSSA)	29	Absence of High-Level Resistance	28	Sensitive
SRL80	<i>S. aureus</i> (MSSA)	29	Absence of High-Level Resistance	27	Sensitive
SRL81	<i>S. aureus</i> (MSSA)	22	Absence of High-Level Resistance	27	Sensitive
SRL82	<i>S. aureus</i> (MSSA)	27	Absence of High-Level Resistance	30	Sensitive
Cont..					

Fig. 9: Cont..(Disk diffusion results from the extension study Table 3)

SRL83	<i>S. aureus</i> (MRSA)	27	Absence of High-Level Resistance	32	Sensitive
SRL84	<i>S. aureus</i> (MRSA)	25	Absence of High-Level Resistance	30	Sensitive
SRL85	<i>S. aureus</i> (MRSA)	25	Absence of High-Level Resistance	32	Sensitive
SRL86	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	27	Sensitive
SRL88	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	30	Sensitive
SRL89	<i>S. aureus</i> (MRSA)	21	Absence of High-Level Resistance	31	Sensitive
SRL90	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	31	Sensitive
SRL91	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	29	Sensitive
SRL92	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	28	Sensitive
SRL93	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	31	Sensitive
SRL94	<i>S. aureus</i> (MRSA)	24	Absence of High-Level Resistance	30	Sensitive
SRL95	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	29	Sensitive
SRL96	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	28	Sensitive
SRL97	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	26	Sensitive
SRL98	<i>S. aureus</i> (MRSA)	23	Absence of High-Level Resistance	29	Sensitive
SRL99	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	30	Sensitive
SRL100	<i>S. aureus</i> (MRSA)	22	Absence of High-Level Resistance	29	Sensitive
SRL101	<i>S. aureus</i> (MRSA)	35	Absence of High-Level Resistance	27	Sensitive

MRSA: Methicillin-resistant *Staphylococcus aureus*; MSSA: Methicillin-sensitive *Staphylococcus aureus*; MRSE: Methicillin-resistant *Staphylococcus epidermidis*; ZOI: Zone of Inhibition

Fig. 10: Cont..(Disk diffusion results from the extension study Table 3)

Sr. No	Antibiotics		Mupirocin MIC Conc	Clindamycin MIC Conc	Fusidic Acid MIC Conc	Nadifloxacin - Reference MIC Conc (µg/ml)
	Isolate Name	Isolate Name				
SRL 1	<i>S. aureus</i> (MRSA)	<i>S. aureus</i> (MRSA)	0.01	<0.001	0.01	1
SRL 10	<i>S. aureus</i> (MRSA)	<i>S. aureus</i> (MRSA)	0.01	0.01	0.01	1
SRL 12	<i>S. aureus</i> (MRSA)	<i>S. aureus</i> (MRSA)	0.01	0.01	0.001	1
SRL 15	<i>S. aureus</i> (MRSA)	<i>S. aureus</i> (MRSA)	0.01	0.001	0.001	1
SRL 23	<i>S. aureus</i> (MRSA)	<i>S. aureus</i> (MRSA)	0.01	0.001	0.001	1
SRL 24	<i>S. aureus</i> (MRSA)	<i>S. aureus</i> (MRSA)	0.01	0.001	0.001	1
SRL 29	<i>S. aureus</i> (MRSA)	<i>S. aureus</i> (MRSA)	0.01	0.001	0.001	1
SRL 50	<i>S. aureus</i> (MRSA)	<i>S. aureus</i> (MRSA)	0.01	0.01	0.001	< 0.0625
SRL 51	<i>S. aureus</i> (MRSA)	<i>S. aureus</i> (MRSA)	0.01	0.01	0.001	1
SRL 52	<i>S. aureus</i> (MRSA)	<i>S. aureus</i> (MRSA)	0.01	0.01	0.001	1
SRL 64	<i>S. aureus</i> (MRSA)	<i>S. aureus</i> (MRSA)	0.01	0.01	0.001	<0.0625
SRL 41	<i>S. aureus</i> (MSSA)	<i>S. aureus</i> (MSSA)	0.01	0.01	0.01	2
SRL 42	<i>S. aureus</i> (MSSA)	<i>S. aureus</i> (MSSA)	0.01	0.001	0.01	<0.0625
SRL 35	<i>S. aureus</i> (MSSA)	<i>S. aureus</i> (MSSA)	0.01	0.001	0.01	1
SRL 2	<i>S. aureus</i> (MSSA)	<i>S. aureus</i> (MSSA)	0.01	>240	0.01	1
SRL 3	<i>S. aureus</i> (MSSA)	<i>S. aureus</i> (MSSA)	0.01	>240	0.01	1
SRL 4	<i>S. aureus</i> (MSSA)	<i>S. aureus</i> (MSSA)	0.01	<0.001	0.01	1
SRL 9	<i>S. aureus</i> (MSSA)	<i>S. aureus</i> (MSSA)	0.01	<0.001	0.01	1
SRL 11	<i>S. aureus</i> (MSSA)	<i>S. aureus</i> (MSSA)	0.01	0.01	0.001	1
SRL 16	<i>S. aureus</i> (MSSA)	<i>S. aureus</i> (MSSA)	0.01	0.001	0.001	1
SRL 22	<i>S. aureus</i> (MSSA)	<i>S. aureus</i> (MSSA)	0.01	0.001	0.001	1
SRL 53	<i>S. aureus</i> (MSSA)	<i>S. aureus</i> (MSSA)	0.001	0.01	0.001	1
SRL 59	<i>S. aureus</i> (MSSA)	<i>S. aureus</i> (MSSA)	0.001	0.01	0.001	1
SRL 67	<i>S. aureus</i> (MSSA)	<i>S. aureus</i> (MSSA)	0.001	0.001	0.001	2
SRL 48	<i>S. epidermidis</i>	<i>S. epidermidis</i>	7.5	2	1	1
SRL 56	<i>S. epidermidis</i>	<i>S. epidermidis</i>	>240	2	0.001	1
SRL 57	<i>S. epidermidis</i>	<i>S. epidermidis</i>	>240	2	0.001	1

Fig. 11: MIC results from the extension study (Table 4)

SRL58	<i>S. epidermidis</i>	>240	>240	0.001	0.001	<0.0625
SRL62	<i>S. epidermidis</i>	0.001	0.001	0.001	0.001	0.5
SRL63	<i>S. epidermidis</i>	0.001	0.001	0.001	0.001	<0.0625
SRL40	<i>S. epidermidis</i>	>240	>240	0.001	0.001	0.5
SRL49	<i>S. epidermidis</i> (MRSE)	5	0.1	0.01	0.01	<0.0625
SRL76	<i>S. aureus</i> (MRSA)	1	0.08	0.1	0.1	1
SRL77	<i>S. aureus</i> (MSSA)	1	<0.001	0.1	0.1	1
SRL78	<i>S. aureus</i> (MRSA)	1	<0.001	0.1	0.1	1
SRL79	<i>S. aureus</i> (MSSA)	1	<0.001	0.1	0.1	1
SRL80	<i>S. aureus</i> (MSSA)	1	<0.001	0.1	0.1	1
SRL81	<i>S. aureus</i> (MSSA)	1	<0.001	0.1	0.1	1
SRL82	<i>S. aureus</i> (MRSA)	0.1	<0.001	0.1	0.1	1
SRL83	<i>S. aureus</i> (MRSA)	1	0.01	0.1	0.1	1
SRL84	<i>S. aureus</i> (MRSA)	1	<0.001	0.1	0.1	1
SRL85	<i>S. aureus</i> (MRSA)	1	<0.001	0.1	0.1	0.5
SRL86	<i>S. aureus</i> (MRSA)	1	0.01	0.1	0.1	0.5
SRL88	<i>S. aureus</i> (MRSA)	1	<0.001	0.1	0.1	1
SRL89	<i>S. aureus</i> (MSSA)	0.1	0.01	0.1	0.1	0.5
SRL90	<i>S. aureus</i> (MRSA)	1	0.01	1	1	0.5
SRL91	<i>S. aureus</i> (MRSA)	1	<0.001	1	1	0.125
SRL92	<i>S. aureus</i> (MRSA)	1	0.01	0.1	0.1	0.25
SRL93	<i>S. aureus</i> (MRSA)	0.1	0.05	0.1	0.1	0.25
SRL94	<i>S. aureus</i> (MRSA)	1	0.08	1	1	0.5
SRL95	<i>S. aureus</i> (MRSA)	1	0.05	0.1	0.1	0.5
SRL96	<i>S. aureus</i> (MRSA)	1	0.08	1	1	0.25
SRL97	<i>S. aureus</i> (MRSA)	1	0.001	0.1	0.1	0.25
SRL98	<i>S. aureus</i> (MSSA)	> 240	<0.001	1	1	0.5
SRL99	<i>S. aureus</i> (MSSA)	1	0.05	0.1	0.1	0.5
SRL100	<i>S. aureus</i> (MRSA)	1	0.05	0.1	0.1	0.25
SRL101	<i>S. aureus</i> (MRSA)	1	<0.001	0.1	0.1	0.25

MRSA: Methicillin-resistant *Staphylococcus aureus*; MSSA: Methicillin-Sensitive *Staphylococcus aureus*; MRSE: Methicillin-resistant *Staphylococcus epidermidis*; MIC: Minimum Inhibitory Concentration

Fig. 12: Cont., (MIC results from the extension study Table 4)

4. Discussion

This study evaluated the antibiotic susceptibility patterns of common bacterial strains causing SSTIs. Specifically, the study aimed to assess the ZOI and MIC of four topical antibiotics- Nadifloxacin, Mupirocin, Clindamycin, and Fusidic acid against bacterial strains. Nadifloxacin was effective against 70% of the isolates at a MIC of < 4 µg/ml. The results of the study revealed that Nadifloxacin consistently exhibited a larger ZOI compared to the other antibiotics for all bacterial strains tested. This indicates that Nadifloxacin has a higher efficacy in inhibiting the growth of these bacterial strains, suggesting its potential as a first-line treatment option for SSTIs caused by these organisms. The larger ZOI can be attributed to Nadifloxacin's specific mechanism of action, targeting bacterial DNA gyrase and topoisomerase IV, which are crucial for bacterial replication.⁸ A similar study conducted by Alba et al., 2009 investigated Nadifloxacin against isolates of *P. acnes*, *MSSA*, *MRSA*, and *S. epidermidis* from Spain, Hungary, and Germany.⁹ The study demonstrated that Nadifloxacin outperforms the comparators (Ciprofloxacin, Erythromycin, and Clindamycin) against the above-mentioned bacteria and has no additional effect on resistance.⁹

Nadifloxacin is found to be effective when used against both aerobic and anaerobic isolates. MIC₉₀ values of nadifloxacin for *S. aureus* was 0.1 g/ml, *Streptococcus* spp. was 0.78 g/ml, and *Propionibacterium* spp.'s was 0.39 g/ml. Other antibiotics, however, showed resistance, with some strains having MICs higher than 12.5 g/ml.⁷

Mupirocin, another topical antibiotic commonly used in the treatment of SSTIs, showed moderate activity against the bacterial strains tested. The ZOI observed for Mupirocin varied among the different strains, indicating a relatively lower efficacy compared to Nadifloxacin. However, it is important to note that Mupirocin is still considered effective against certain bacterial species causing SSTIs, particularly strains of *S. aureus*. Thus, its use may be warranted in cases where Nadifloxacin is contraindicated or when targeting specific bacterial species known to be susceptible to Mupirocin.¹⁰

Fusidic acid, an antibiotic used in SSTI management, displayed varying susceptibility patterns among the bacterial strains. Some strains showed a relatively large ZOI, indicating high susceptibility to Fusidic acid, while others demonstrated a smaller ZOI, suggesting reduced susceptibility. This finding suggests that the use of Fusidic acid as a monotherapy for SSTIs should be approached with caution, as its effectiveness may vary depending on the specific bacterial strain involved. Combination therapy or alternative treatment options may be employed in cases of reduced susceptibility to Fusidic acid.^{11,12} The effectiveness of Mupirocin cream and topical Fusidic acid in treating experimental *S. aureus* infections was comparable, aligning with clinical observations. Nonetheless, Fusidic

acid's effectiveness is reduced against *streptococci* and is particularly less efficient than Mupirocin cream in addressing *S. pyogenes* infected wounds.¹³

Clindamycin, a broad-spectrum antibiotic, exhibited varied susceptibility patterns across the bacterial strains tested. Some strains showed a significant ZOI, indicating high susceptibility, while others demonstrated reduced susceptibility. This suggests that the efficacy of Clindamycin against SSTIs may be dependent on the specific bacterial strain involved.¹⁰ The rates of Clindamycin resistance in *MRSA* were naturally greater than those in *MSSA*. Interestingly, just 4% were resistant to Nadifloxacin. The discrepancy may be because Nadifloxacin predominantly targets DNA gyrase.⁹

Previous research has extensively examined Nadifloxacin's bactericidal effects. It displays remarkable in vitro activity against both aerobic and anaerobic bacteria, including *S. epidermidis*, *S. aureus*, *S. pyogenes*, *Streptococcus viridans*, *E. coli*, *P. acnes*, and *P. granulosum*.^{14–22} The MIC₅₀ values were determined as 0.25 µg/ml for *P. acnes*, 0.125 µg/ml for *P. granulosum*, 0.03 µg/ml for *S. aureus* and 0.06 µg/ml for CNS. Notably, no resistance to the fluoroquinolone Nadifloxacin was detected, consistent with the findings of Kurokawa et al.²³ The current study's outcomes align with Vogt et al.,¹⁶ who similarly found no Nadifloxacin-resistant strains of *S. aureus*, CNS, *P. acnes*, or *P. granulosum* in acne vulgaris patients. In contrast, tests with other antibiotics revealed resistant strains with MICs surpassing 12.5 µg/ml.

It is worth noting that antibiotic resistance is a growing concern, particularly in the context of SSTIs. The emergence of multidrug-resistant strains poses significant challenges in the effective treatment of these infections. Therefore, periodic surveillance of antibiotic susceptibility patterns is crucial for guiding empirical therapy and ensuring the selection of appropriate antibiotics.^{24,25}

It is important to consider several limitations of the study. In vitro studies have inherent limitations in replicating the complexities of the human body, thus the results may not accurately reflect the clinical response. The study focused on specific infections and may not apply to other types or populations. Genetic factors and resistance mechanisms were not analyzed, limiting insights into treatment strategies. Pharmacokinetic and pharmacodynamic properties were not considered, which can impact clinical effectiveness. The sample size was relatively small, warranting caution in generalizing the results.

In summary, the study employed a comparative approach to assess multiple antibiotics used for SSTIs, providing comprehensive insights for antibiotic selection. It evaluated both ZOI and MIC, enhancing the understanding of antibiotic efficacy. The focus on relevant bacterial strains and prospective design strengthens the applicability and

reliability of the findings. The study's results can guide future research and evidence-based treatment guidelines. However, further research considering larger sample sizes and additional factors is needed to optimize treatment strategies for these infections.

5. Conclusion

In conclusion, this prospective, comparative, in vitro study evaluated the antibiotic susceptibility patterns of common bacterial strains causing SSTIs. The findings demonstrated varying degrees of susceptibility to the tested antibiotics, including Nadifloxacin, Mupirocin, Clindamycin, and Fusidic acid. These results provide valuable insights into the selection of empirical treatment options for such infections. Nadifloxacin's superior efficacy in the study can be attributed to its specific mode of action, broad spectrum of activity, excellent tissue penetration, favorable pharmacokinetic profile, and low risk of resistance development. These scientific properties make Nadifloxacin a valuable choice in the treatment of bacterial infections, particularly those involving the skin and soft tissues. However, it is essential to consider the limitations of in vitro studies and the need for further research to better understand antibiotic efficacy and resistance mechanisms in clinical settings. Overall, this study contributes to the knowledge base and can guide clinicians in making informed decisions regarding antibiotic therapy for SSTIs.

6. Conflicts of interest

Dr Rashmi is technical expert at Agilus Diagnostics Limited. All other authors are employees of Dr. Reddy's Laboratories.

7. Funding

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8. Contribution Details

All the authors have contributed to design of the work, data analysis, interpretation of data, manuscript preparation and review.

9. Data Availability

The data underlying this article are incorporated into the article.

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