Anti-Microbial activity of Ofloxacin and its derivatives

Abstract
The Anti-microbial activity of a fluoroquinolone, levofloxacin and its derivatives was tested against numerous Gram positive and Gram negative bacteria. Antimicrobial susceptibility of levofloxacin and its derivatives was tested by the disc diffusion technique. Zone inhibition of Levofloxacin and its derivatives was analyzed at different concentrations (5, 10, 20 ppm).

Keywords: Ofloxacin, Anti-Microbial, Zone inhibition, Levofloxacin, Gram positive, Gram negative bacteria

1 Introduction
Levofloxacin is a synthetic chemotherapeutic antibiotic of the fluoroquinolone drug class. Levofloxacin is a chiral fluorinated carboxyquinolone. Investigation of ofloxacin, an older drug that is the racemic mixture, found that the L form [the (-)-(S) enantiomer] is more active. This specific component is levofloxacin. Levofloxacin is the L-isomer of the racemate ofloxacin, a quinolone antimicrobial agent. In chemical terms, levofloxacin, a chiral fluorinated carboxyquinolone, is the pure (-)-(S)-enantiomer of the racemic drug substance ofloxacin. The chemical name is (-)-(S)-9fluoro-2,3-dihydro-3-methyl-10-(4-methyl-1-piperazinyl)-7-oxo-7H-pyrido[1,2,3- de]-1,4benzoazine-6-carboxylic acid hemihydrate. The empirical formula is $C_{18}H_{20}FN_3O_4 \cdot \frac{1}{2} H_2O$, and the molecular weight is 370.38g

Figure 1: Structure of Levofloxacin
2 Synthesis of Levofloxacin and its derivatives

![Synthetic pathway of Levofloxacin derivatives]

Figure 2: Synthetic pathway of Levofloxacin derivatives

3 Anti-microbial activity of Ofloxacin and its derivatives

The antimicrobial susceptibility of all the derivatives was tested by the disc diffusion technique developed by Bauer et al. For this purpose 50 ppm stock solution of levofloxacin and its derivatives were prepared. The stock solution was diluted to 3 different concentrations i.e. 5, 10 and 20 ppm. Commercially available filter paper discs were soaked in the prepared drug and derivatives solution, dried and applied on the surface of solid culture media (Nutrient Agar), which had been streaked with standardized bacterial inoculums and incubated at 37°C for 24 h. This method is based on the determination of an inhibited zone proportional to the bacterial susceptibility to the antimicrobial present in the disk. The results were compared with the parent against 11 different strains of Gram positive (Staphylococcus aureus, Bacillus subtilis, Streptococcus pneumoniae, Corynebacterium hoffmannii) and Gram negative organisms (Klebsiella pneumoniae, Proteus mirabilis, Shigella flexneri, Escherichia coli, Pseudomonas areuginosa, Citrobacter species, and Salmonella typi).

When the aryl group is phenyl $b$ it was fairly active against Staphylococcus aureus and Shigella flexneri. Introduction of OH group to aryl amine $c$ and $e$ showed respectable activity against Proteus mirabilis and Shigella flexneri respectively.

Among these compounds, compound $d$ bearing amino group on the phenyl substituent, exhibited the lowest potency against Gram-negative Pseudomonas areuginosa and greater potency against Gram-positive Staphylococcus aureus. When an aromatic ring was introduced to the phenyl substituent $f$ significant enhancements of potency against Bacillus subtilis, Klebsiella pneumoniae and Escherichia coli were achieved. It is also worthwhile to point out that formation of hydrazide $a$ at 6 position exhibited the highest activity against all the Gram-positive strains tested, more potent than reference agent. It is
proposed that in the terms of structure-activity relationship, the antibacterial activity profile against Gram-negative bacteria was modulated and enhanced by the phenyl attachment via amide linkage at the 6-position of the levofloxacin molecule. The overall activity profile of compounds (a-f) against microorganisms revealed that there is remarkable difference in zone of inhibition values as compared to parent (Table 1). In the terms of structure activity relationship, the antibacterial activity profile against all bacterium was altered by addition of amino group in levofloxacin molecule. The alteration of substitution in amines made marked differences in activity. It seems that expansion of activity is due to better interaction of molecule with target enzymes or for penetration into these bacteria.

<table>
<thead>
<tr>
<th>S. No</th>
<th>Microorganisms</th>
<th>Levofoxacin</th>
<th>a</th>
<th>b</th>
<th>c</th>
<th>d</th>
<th>e</th>
<th>f</th>
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<tbody>
<tr>
<td></td>
<td></td>
<td>(5 ppm)</td>
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<td>(20 ppm)</td>
<td>(5 ppm)</td>
<td>(10 ppm)</td>
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<tr>
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<td>11 16 25</td>
<td>12 16 27</td>
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<td>13 15 28</td>
<td>9 13 19</td>
<td>11 14 25</td>
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<td>14 17 20</td>
<td>16 20 22</td>
<td>16 20 21</td>
<td>16 20 22</td>
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<td>15 21 32</td>
<td>12 19 23</td>
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<td>14 18 24</td>
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<tr>
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<td>11 13 17</td>
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a = Phenyl hydrazine, b = Aniline, c = 3’ amino phenol, d = Phenylene diamine, e = O’ amino phenol, f = Alpha naphthylamine.

Table 1: Zone (mm) of inhibition of Levofloxacin and its derivatives at different concentrations (5, 10, 20 ppm).

4 Results
The Anti-microbial Activity was analyzed, therefore resulting in positive results i.e. Hydrazide a exhibited the highest activity against all the Gram-positive strains, Phenyl b it was fairly active against Staphylococcus aureus and Shigella flexneri. Aryl amine c and e showed respectable activity against Proteus mirabilis and Shigella flexneri. Compound d bearing amino group on the phenyl substituent, exhibited the lowest potency against Gram-negative Pseudomonas aeruginosa and greater potency against Gram-positive Staphylococcus aureus. Phenyl substituent f significant enhancements of potency against Bacillus subtilis, Klebsiella pneumoniae and Escherichia coli were achieved.
5 Conclusion

Levofloxacin derivatives viz, Phenyl hydrazine, Aniline, 3’amino phenol, phenyl diamine, O’amino phenol and Alpha naphthylamine were synthesized successfully and possess potent anti-bacterial activity against numerous Gram positive and Gram negative bacteria.

6 References


