Finafloxacin as a novel prophylaxis for *Burkholderia pseudomallei* infection

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Introduction

Burkholderia pseudomallei is intrinsically resistant to many antimicrobial agents, most apparent in acidic conditions that may be encountered *in vivo*. Antibiotic treatment is lengthy and relapses of infection are seen. Finafloxacin is a novel C-8-cyano-fluoroquinolone developed by MerLion Pharmaceuticals that has been chemically manipulated to improve its antibacterial activity under acidic conditions, where other fluoroquinolones, including ciprofloxacin, are less well activated. It has good antimicrobial activity against a wide range of bacterial species including MDR Gram-negative pathogens, has been shown to be safe in Phase I and II clinical trials and, furthermore, to be more effective than ciprofloxacin in treating complicated urinary tract infections and pyelonephritis. Here we describe how we have used an established mouse model of melioidosis to determine the efficacy of finafloxacin against an aerosol challenge of *B. pseudomallei*.

Methods

Groups of 20 Balb/c mice were challenged with approximately 128 cfu of *B. pseudomallei* strain K96243 by the inhalational route using a Henderson apparatus. Mice received a human equivalent dose of finafloxacin (37.5 mg/kg every 8 hours orally), ciprofloxacin (30 mg/kg every 12 hours via intraperitoneal injection) or co-trimoxazole (240 mg/kg every 12 hours orally) initiated at 6 or 24 post-challenge for a period of 14 days. Intraperitoneal PBS or oral diluent (Tris buffer) were delivered following the same dosing regimen to control animals. The experiment was terminated at day 63, post mortems were performed and the spleen, liver and lungs were harvested and plated onto L agar plates to determine the bacterial load. Additionally, following 24 hours of antibiotic dosing and at cessation of antibiotic therapy (day 15), 5 mice from each group were culled and the bacterial load within the organs was determined.

Results and Discussion

All treatments significantly reduced the bacterial load in the spleen, liver and lungs ($p<0.05$) in comparison to controls when antibiotics were delivered for 24 hours. When therapy was initiated at 6 hours the spleens and livers of mice treated with finafloxacin were clear from detectable *B. pseudomallei* (Figure 1A). Treating with either finafloxacin or co-trimoxazole reduced the bacterial load in the lungs in comparison to ciprofloxacin ($p<0.05$).
When the antibiotic treatment was delayed until 24 hours, the mice treated with finafloxacin had lower levels of bacteria in the liver and spleen in comparison to ciprofloxacin ($p<0.05$), and a lower bacterial load in the lungs compared to both ciprofloxacin and co-trimoxazole ($p<0.05$) (Figure 1B).

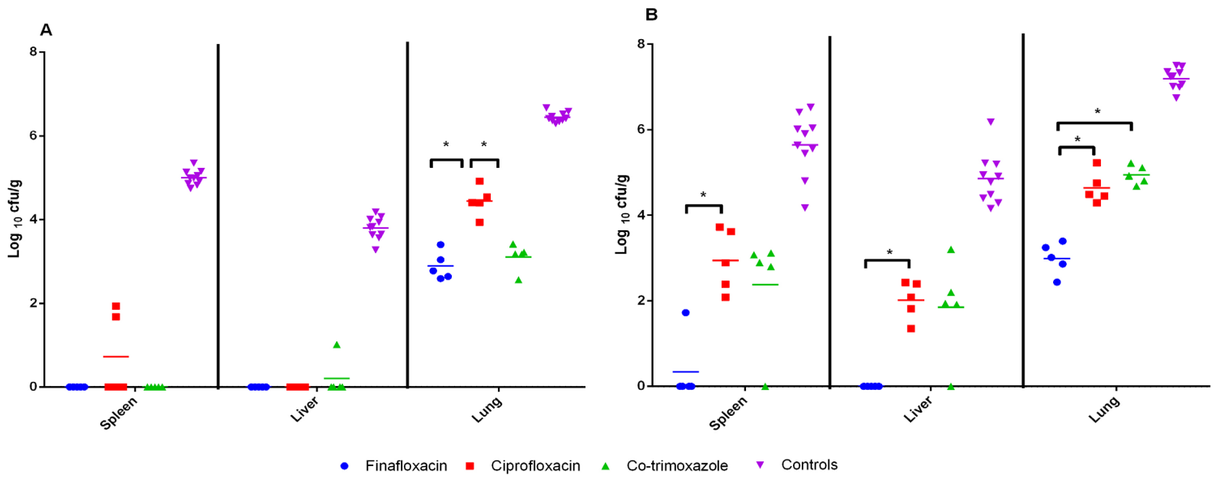

Figure 1. Bacterial load in organs at 24 hours following initiation of antibiotic treatment. Bacterial counts (CFU/g) in the spleen, liver and lungs of 5 mice per group at 30 (A) or 48 hours (B) post challenge. Each group received 24 hours treatment with finafloxacin), co-trimoxazole or ciprofloxacin. Control animals received intraperitoneal PBS or oral diluent. Statistical analysis was performed using Dunn's comparisons of a Mood's Median test (*$p<0.05$).

At cessation of therapy all organs harvested from mice treated with finafloxacin or co-trimoxazole were clear (bar one mouse that was treated with co-trimoxazole that was colonised in the lungs) (Figure 2). In comparison *B. pseudomallei* was recovered all animals treated with ciprofloxacin. When therapy was initiated at 6 hours, mice treated with finafloxacin or co-trimoxazole had less bacteria in the lungs and liver in comparison to mice treated with ciprofloxacin ($p<0.05$) (Figure 2A).

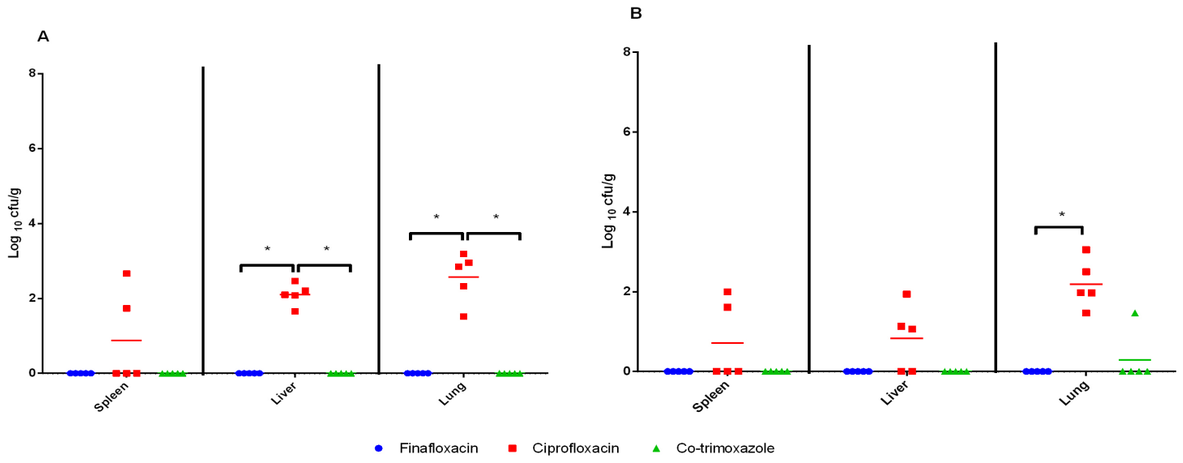

Figure 2. Bacterial load in organs at the end of antibiotic treatment. Bacterial counts (CFU/g) in the spleen, liver and lungs of 5 mice per group when treatment commenced at 6 (A) or 24 hours challenge (B) post challenge. Each group received 14 days of treatment with finafloxacin, co-trimoxazole or ciprofloxacin. Statistical analysis was performed using Dunn's comparisons of a Mood's Median test (*$p<0.05$).

When therapy was delayed until 24 hours there was a lower bacterial load in the lungs of mice treated with finafloxacin in comparison to ciprofloxacin ($p<0.05$) (Figure 2B). *B. pseudomallei* was recovered from all animals that had been treated with ciprofloxacin. At the end of the study, finafloxacin or co-trimoxazole delivered at 6 hours post-challenge increased survival when compared to ciprofloxacin ($p<0.0001$) (Figure 3A). When therapy was delayed to 24 hours finafloxacin or co-trimoxazole also increased survival when compared to ciprofloxacin ($p=0.004$ and $p=0.004$, respectively) (Figure 3B).

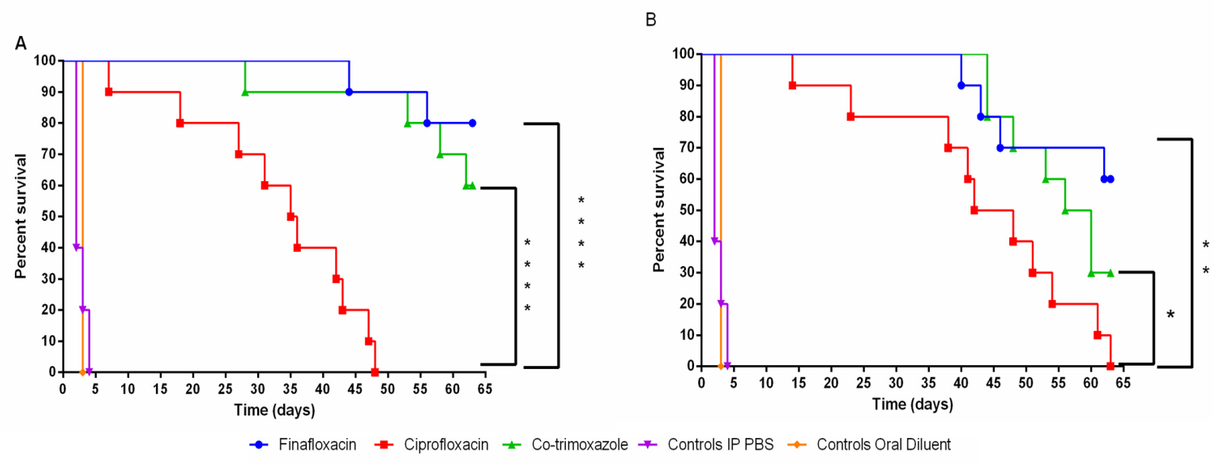

Figure 3. Percentage survival of mice following challenge with aerosolised *B. pseudomallei*. Mice were challenged with *B. pseudomallei* by the inhalational route and treated with oral finafloxacin, intraperitoneal ciprofloxacin or oral co-trimoxazole. Regimens were initiated at 6 (A) or 24 hours (B) post challenge. Control animals received intraperitoneal PBS or oral diluent. Statistical analysis was performed using the Mantel-Haenszel log rank test (*$p=0.043$, **$p=0.004$, ****$p<0.0001$).

Of the survivors, over 70% of animals treated with finafloxacin had cleared the infection (Figure 4A and B). *B. pseudomallei* was recovered from all bar one of the surviving animals that had been treated with co-trimoxazole.

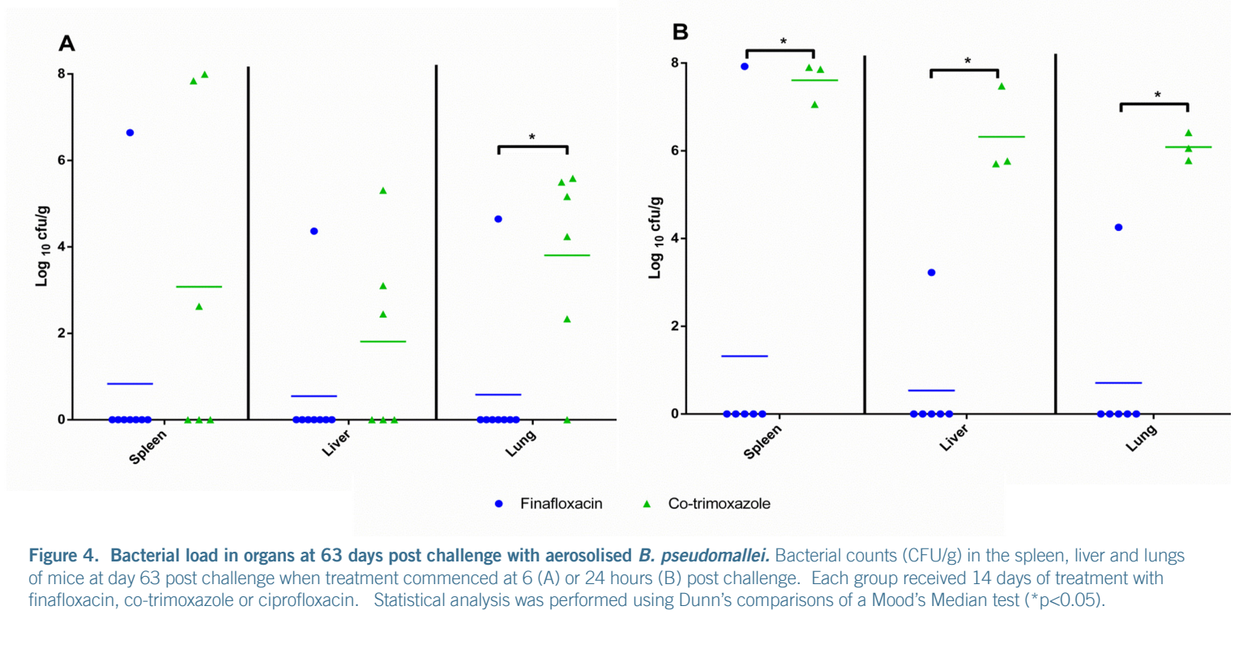

Figure 4. Bacterial load in organs at 63 days post challenge with aerosolised *B. pseudomallei*. Bacterial counts (CFU/g) in the spleen, liver and lungs of mice at day 63 post challenge when treatment commenced at 6 (A) or 24 hours (B) post challenge. Each group received 14 days of treatment with finafloxacin, co-trimoxazole or ciprofloxacin. Statistical analysis was performed using Dunn's comparisons of a Mood's Median test (*$p<0.05$).

Summary

Finafloxacin demonstrated improved efficacy, in comparison to ciprofloxacin or co-trimoxazole, when administered at 6 hours or 24 hours post challenge. Of the survivors, over 70% of animals treated with finafloxacin had cleared the infection. This study demonstrates the benefit of evaluating novel antibiotics, shown to be active against MDR Gram negative organisms, against intracellular pathogens such as *B. pseudomallei*. Further studies are warranted to further investigate the utility of finafloxacin for the treatment of melioidosis.

References