International Journal of Antimicrobial Agents 46 (2015) 28–32

Contents lists available at ScienceDirect

International Journal of Antimicrobial Agents

journal homepage: http://www.elsevier.com/locate/ijantimicag

Effect of dexamethasone on the efficacy of daptomycin in the therapy of experimental pneumococcal meningitis

M. Vivas[a,*], E. Force[a], F. Tubau[b], C. El Haj[a], J. Ariza[a], C. Cabellos[a]

[a] Laboratory of Experimental Infection, Infectious Diseases Department, IDIBELL-Hospital Universitari de Bellvitge, Feixa Llarga s/n, 08907 L'Hospitalet de Llobregat, Barcelona, Spain
[b] Microbiology Department, IDIBELL-Hospital Universitari de Bellvitge and CIBERES ISCIII, Feixa Llarga s/n, 08907 L'Hospitalet de Llobregat, Barcelona, Spain

ARTICLE INFO

Article history:
Received 10 October 2014
Accepted 28 January 2015

Keywords:
Steroids
Lipopeptide
CNS infection
Streptococcus pneumoniae

ABSTRACT

This study aimed to determine the effect of dexamethasone in combination with low-dose or high-dose daptomycin for the treatment of penicillin- and cephalosporin-resistant pneumococcal meningitis. Efficacy (ΔCFU/mL) and cerebrospinal fluid (CSF) levels of daptomycin at 15 mg/kg and 25 mg/kg were studied in a rabbit model of pneumococcal meningitis, comparing them with the same doses in combination with dexamethasone at 0.125 mg/kg every 12 h over a 26-h period against two different *Streptococcus pneumoniae* strains, HUB 2349 and ATCC 51916 with daptomycin minimum inhibitory concentrations (MICs) of 0.09 mg/L and 0.19 mg/L, respectively. Daptomycin levels in CSF were lower when dexamethasone was given concurrently. Against strain HUB 2349, therapeutic failure occurred with daptomycin 15 mg/kg + dexamethasone; daptomycin 25 mg/kg + dexamethasone was better at reducing bacterial counts than the lower dose throughout treatment. Against the highly cephalosporin-resistant ATCC 51916 strain, daptomycin 15 mg/kg + dexamethasone achieved a lower bacterial decrease than daptomycin 15 mg/kg alone, and therapeutic failure at 24 h occurred in the daptomycin 15 mg/kg + dexamethasone group. Addition of dexamethasone to a 25 mg/kg daptomycin dose did not affect the efficacy of daptomycin: it remained bactericidal throughout treatment. In conclusion, against the studied strains, low-dose (15 mg/kg/day) daptomycin is affected by concomitant use of dexamethasone: CSF levels are reduced and its bacterial efficacy is affected. At a higher daptomycin dose (25 mg/kg/day), however, the use of dexamethasone does not alter efficacy; the combination appears to be a good choice for the treatment of pneumococcal meningitis.

1. Introduction

Despite the efficacy of antibiotic therapy, the mortality rate of bacterial meningitis is still significant, ranging between 10% and 30% [1], and >30% of survivors develop long-term sequelae including hearing loss and neurological deficits [2].

The rapid bactericidal action of daptomycin makes it an attractive alternative for the treatment of multidrug-resistant pneumococcal meningitis. In addition, its non-bacteriolytic activity may represent an advantage even in cases of full β-lactam susceptibility.

Adjunctive treatment with corticosteroids has been recommended to decrease the inflammation and mortality in bacterial meningitis. Several clinical studies have demonstrated that dexamethasone is beneficial in the treatment of the condition, reducing mortality and improving the prognosis [1], but concomitant use of dexamethasone in combination with antibiotic therapy is still controversial [3].

Diverse clinical trials have demonstrated that adjunctive therapy with dexamethasone reduced severe hearing loss in children and reduced the risk of death and non-favourable sequelae in adults with bacterial meningitis [4,5]. However, it is well known that concomitant use of dexamethasone affects antibiotic penetration into the cerebrospinal fluid (CSF) across the blood–brain barrier and may be associated with failure of antibiotic therapy [6].

Two clinical studies showed that dexamethasone could affect the efficacy of vancomycin for the treatment of pneumococcal meningitis depending on the antibiotic concentrations in the CSF [7,8].

The interaction between dexamethasone and different antibiotics has been widely studied in experimental meningitis models. Syrogiannopoulos et al. assessed the effect of dexamethasone in

* Corresponding author. Tel.: +34 932 607 625/934 035 805; fax: +34 932 607 637.
E-mail address: mireiavivas@gmail.com (M. Vivas).

http://dx.doi.org/10.1016/j.ijantimicag.2015.01.014
0924-8579/

combination with ceftriaxone. CSF antibiotic levels were lower when dexamethasone was added and they found no differences in inflammation parameters [9].

Another experimental study by our group demonstrated that the use of dexamethasone resulted in therapeutic failure and a substantial decrease in ceftriaxone levels in the CSF, even at high ceftriaxone doses, in a cephalosporin-resistant strain of *Streptococcus pneumoniae* [10].

Addition of dexamethasone had a beneficial effect on the inflammatory response in an experimental study using rifampicin, rifampicin plus vancomycin, and ceftriaxone plus rifampicin and did not significantly interfere with antibiotic levels in the CSF [11,12].

Demonstrating that the efficacy of antibiotic therapy is not affected by concomitant use of dexamethasone is a matter of necessity, since the use of dexamethasone may represent a useful adjunctive therapy in cases of pneumococcal meningitis.

In an experimental animal model, we recently reported [13] that daptomycin may represent a good alternative for the treatment of penicillin- and cephalosporin-resistant strains of *S. pneumoniae* owing to its ability to sterilise CSF samples from the very beginning of treatment. The aim of this study was to test the effect of dexamethasone as standard adjunctive therapy given in combination with low-dose or high-dose daptomycin on the treatment of penicillin- and cephalosporin-resistant strains of *S. pneumoniae* in an experimental meningitis model.

2. Materials and methods

2.1. Bacterial strains

Two different strains of *S. pneumoniae* isolated from patients with meningitis were used: strain HUB 2349 is penicillin- and cephalosporin-resistant; and the ATCC 51916 strain (Tennessee 23F-4 clone) is intermediately penicillin-resistant and highly cephalosporin-resistant. Minimum inhibitory concentrations (MICs) and minimum bactericidal concentrations, respectively, were as follows: for strain HUB 2349, penicillin, 4 mg/L and 4 mg/L; ceftriaxone/cefotaxime, 2 mg/L and 4 mg/L; vancomycin, 0.25 mg/L and 0.25 mg/L; and daptomycin, 0.09 mg/L and 0.18 mg/L; and for the ATCC 51916 strain, penicillin, 0.12 mg/L and 0.25 mg/L; ceftriaxone/cefotaxime, 32 mg/L and 32 mg/L; vancomycin, 0.25 mg/L and 0.25 mg/L; and daptomycin, 0.19 mg/L and 0.38 mg/L.

2.2. In vivo studies

2.2.1. Meningitis model

The rabbit model of meningitis originally described by Dacey and Sande [14] was used with slight modifications. Young female New Zealand White rabbits were anaesthetised intramuscularly with 35 mg/kg ketamine (Ketolar®; Parke-Davis, Prat de Llobregat, Spain) and 5 mg/kg xylazine (Rompun®; Bayer AG, Leverkusen, Germany). Meningitis was induced with an intracisternal injection of 250 μL of a saline suspension containing 10^6 CFU/mL of inoculum. In rabbits infected with the HUB 2349 strain, therapy was started 18 h post-inoculation. In animals infected with the ATCC 51916 strain, therapy was initiated 40 h post-inoculation owing to the slow progression of meningitis [15].

Rabbits were anaesthetised using urethane (Sigma Chemical Co., St Louis, MO) at 1.75 g/kg subcutaneously and thiopental sodium (Tiopental®; B. Braun Medical S.A., Rubí, Spain) at 5 mg/kg intravenously. A blood sample was collected to assess secondary bacteraemia.

Animals were placed in a stereotactic frame and a baseline CSF sample was taken (0 h). A dose of dexamethasone (Fortecortin®; Merck, Barcelona, Spain) of 0.25 mg/24 h divided every 12 h (q12h) was given intravenously. Then, 10 min later antibiotic therapy was administered. CSF samples were taken after 2, 6, 24 and 26 h of therapy. Hydration was ensured throughout the experiment. Mortality was assessed at 26 h. Surviving animals were euthanised using a lethal dose of thiopental sodium at the end of each experiment.

2.2.2. Therapeutic groups

Intravenous antibiotic therapy was then administered for 26 h using one of the following therapy schedules: daptomycin at 15 mg/kg ($n = 8$ rabbits for each strain) given once daily; daptomycin 15 mg/kg given once daily plus dexamethasone ($n = 9$ rabbits for each strain) at 0.125 mg/kg q12h; daptomycin at 25 mg/kg ($n = 9$ rabbits for each strain) given once daily; and daptomycin 25 mg/kg given once daily plus dexamethasone ($n = 10$ rabbits for each strain) at 0.125 mg/kg q12h. Untreated control rabbits received saline. Part of the results have already been described in our previous study [13].

2.3. Sample processing

CSF samples were used to determine CSF white blood cell (WBC) counts, bacterial counts and antibiotic levels at peak and trough time points. For leukocyte counts, 10 μL of each sample was diluted 1:1 with Turk solution and read with a Neubauer chamber. Animals presenting ≥300 cells/mm^3 were included in the study groups. Serial ten-fold dilutions were made to determine bacterial counts at each time point. To avoid interference due to carryover of antimicrobial agent, an entire agar plate was used for each sample. The lowest bacterial concentration detectable was 10 CFU/mL. For the purpose of analysis, a value of 0.99 log CFU/mL was assigned to the first sterile culture, and a value of 0 log CFU/mL to subsequent ones. Changes in bacterial counts (Δlog CFU/mL) were calculated as the difference between bacterial concentrations at the start of therapy and at 2, 6, 24 and 26 h. Therapeutic failure was defined as an increase in bacterial concentration of ≥1 log CFU/mL compared with a previous count. A therapy was considered bactericidal when a reduction of 3 log CFU/mL was achieved. Samples were centrifuged at 5000 × *g* for 10 min and the supernatants were stored at −70 °C.

2.4. Antibiotic assays

Daptomycin concentrations were measured using the agar disk diffusion method [16] using *Micrococcus luteus* ATCC 9341 [linearity of assay (r^2) = 0.99; lower detection limit, 2 μg/mL] as the assay organism.

2.5. Statistical analysis

All bacterial counts are presented as log numbers of CFU per millilitre (mean ± standard deviation). Differences in bacterial counts between treated and untreated animals were evaluated for statistical significance using analysis of variance (ANOVA). An unpaired Student's *t*-test with Bonferroni correction was used to determine statistical significance. WBC counts were compared using the non-parametric Mann–Whitney and Wilcoxon tests. For all tests, differences were considered to be statistically significant when *P*-values were <0.05.