MATERIAL AND METHODS

Effect of the Calcium on the determination of MIC of Daptomycin:

The influence of different blood culture media on minimal inhibitory concentration (MIC) determination, including the concentration of Calcium was first evaluated in double blind in two different laboratories using six clinical isolates and five reference strains of C. difficile, Staphylococcus aureus (ATCC 29213) and Enterococcus faecalis (ATCC 29212) were used as controls. The blood agar medium (Oxoid CA2057, BD) was used for all tests. The CaCl₂ stock solution (100 mM) was added to the media at a final concentration of 10 mM. The cultures were incubated at 37 °C for 24 hours. The MIC was determined by assessing the growth of bacteria on the agar by plate count.

Detection of the selection of resistant strains

CCs media media including 10 μg/L daptomycin were used for isolating resistant strains from stool specimens and nasal contents of dead animals. The MIC of daptomycin were systematically determined for each isolate during all the in vivo experiments.

RESULTS of in vivo experiments

Effect of a single dose of Vancomycin or Daptomycin

- The result of a single dose of both antibiotics, orally administered 24 hours after infection, failed to significantly alter the occurrence of C. difficile-induced colitis in hamsters and the occurrence of death (p=0.00007, Log Rank test) (Figures 1 and 2).
- However, a single of both of them was not sufficient to prevent death that invariably occurs within 7 to 20 days after those of untreated animals.
- We did not observe differences in the respective efficacies of Daptomycin and Vancomycin (as reference treatment), after the administration of a single dose that invariably occurs within 7 to 20 days after those of untreated animals.

Effect of different antibiotics on the determination of MIC of Daptomycin:

- Multiple doses of Daptomycin and Vancomycin started since the onset of CDI, cures animals from C. difficile-induced colitis in hamsters (P<0.0001, Log Rank Test, Figure 3). However, this therapeutic scheme led to early mortality not reported with the previous assay and recurrences occurred with both antibiotics.
- This clearly shows the interest to start an adapted treatment as soon as possible in order to prevent treatment failure or recurrence.
- No emergence of resistant strains or increase MICs of daptomycin has been detected during all the in vivo experiments.

Antibiotic efficacies following various scheme of multiple doses treatment

- Multiple doses of Daptomycin and Vancomycin started since the onset of CDI, cures animals from C. difficile-induced colitis in hamsters (P<0.0001, Log Rank Test, Figure 3). However, this therapeutic scheme led to early mortality not reported with the previous assay and recurrences occurred with both antibiotics.
- This clearly shows the interest to start an adapted treatment as soon as possible in order to prevent treatment failure or recurrence.
- No emergence of resistant strains or increase MICs of daptomycin has been detected during all the in vivo experiments.

CONCLUSIONS

- Daptomycin, a non-absorbed, rapidly bactericidal agent, appears to be an effective treatment to prevent or cure CDI.
- Further studies are needed to determine the impact of Daptomycin on the occurrence of death, as well as its impact on the recurrence of CDI.
- The results of this study suggest that Daptomycin may be a valuable therapeutic alternative to conventional treatments for CDI.

REFERENCES