Effect of tebipenem on the normal gut microbiota of healthy adult population

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Introduction

- Oral tebipenem pivoxil hydrobromide is an investigational carbapenem antibiotic currently in development for the treatment of cUTI, including acute pyelonephritis

- Broad-spectrum antimicrobials, including carbapenems, may impair the gut microbiota with alterations dependent not only on anti-anaerobic properties but also parameters with marked interindividual variability

- Perturbation of gut microbiome may be associated with several health complications and diminished colonization resistance
  - Compositions
  - Load of each taxa
  - Relative abundance
  - Selection of resistance

- Evaluation of collateral damage is important
Introduction

- A greater understanding of how antibiotics alter the composition and function of the gut microbiome is of important clinical and societal utility
  - Extent of collateral damage on the gut microbiome
  - Rate of recovery to baseline microbiome composition after treatment

- We aimed to assess the potential ecological effects of oral tebipenem pivoxil hydrobromide on the gut microbiome of adult healthy population compared to amoxicillin-clavulanic acid (ClinicalTrials.gov Identifier: NCT04376554)

- Hypothesis:
  - The impact of oral tebipenem pivoxil hydrobromide on pertubations of the gut microbiome is comparable to that of amoxicillin-clavulanic acid.

- Quantitative culture and 16S rDNA metagenomics were used
Methods

- Phase 1, single-center, open-label, randomized, parallel-group, active-control study

- Healthy study subjects were randomized (1:1) to a treatment arm:
  - Oral tebipenem pivoxil hydrobromide 600mg q8h
  - Oral amoxicillin/clavulanate 500/125mg q8h

**Flowchart:**

```
Screened (N=49)  
  -----------------  
  Randomized (N=30)  
  |                |                |
  |  Amoxicillin-clavulanate (N=15) |  Tebipenem pivoxil hydrobromide (N=30) |
  |                |                |
  | Treatment      | Treatment       |
  | Completed (N=14)| Completed (N=15)|
  | Withdrew (N=1) | Withdrew (N=0) |
  |                |                |
  | Follow-Up      | Follow-Up       |
  | Entered (N=15) | Entered (N=15) |
  | Completed (N=15)| Completed (N=15) |

Failed screening (N=19)
```
Methods

Study Population, Sample Collection, and Lab analysis

A. Randomized - two group
   - Study group
     - Healthy Individual
     - treated with Tebipenem
   - Control group
     - treated with Amoxicillin-clavulanic acid

B. Data and sample collection
   - Fecal samples were collected at 9 sampling points:
     - Baseline, Day 3, Day 4, Day 7, Day 10, Day 14, Day 21,
     - Day 90, Day 180

C. Analysis
   - Phenotypic microbiota analysis
   - 16S rRNA metagenomic analysis
Results

- More pronounced reductions in CFU observed in tebipenem group as compared to AMOX/CLAV for:
  - **Enterobacterales**
    - Day4 (TukeyHSD; p-value=0.000433),
    - Day7 (TukeyHSD; p-value=0.000509)
    - Day10 (TukeyHSD; p-value=0.00902)
  - **Enterococcus spp.**
    - Day4 (TukeyHSD; p-value=0.0247),
    - Day7 (TukeyHSD; p-value=0.00027),
    - Day10 (TukeyHSD; p-value=0.00436), and
    - Day14 (TukeyHSD; p-value=0.00305)

- All measured genera showed recovery after 14 days in both groups (CFU counts reverted to baseline after treatment)

- Variable impact on *Bacteroides*, *Lactobacillus*, and *Bifidobacterium* (not significant)
Phenotypic microbiome: clinically important taxa

Tebipenem pivoxil hydrobromide group

Amoxicillin-clavulanate group

Enterobacterales

Enterococcus
Phenotypic microbiome: clinically important taxa

**Bacteroides**
- Tebipenem
- Amoxicillin-clavulanate

**Bifidobacterium**
- Tebipenem
- Amoxicillin-clavulanate

**Lactobacillus**
- Tebipenem
- Amoxicillin-clavulanate

**Candida**
- Tebipenem
- Amoxicillin-clavulanate
16S rDNA metagenomics

Tebipenem group
Between Day 1 and:
- Day 7 (TukeyHSD; p-value= 0.00489)
- Day 10 (TukeyHSD; p-value= 0.00123)
- Day 14 (TukeyHSD; p-value= 0.0205)

Amoxicillin-clavulanic acid group
Between Day 1 and:
- Day 7 (TukeyHSD; p-value=0.0522)

Tebipenem group
Between Day 1 and:
- Day4 (TukeyHSD; p-value=1.37E-6),
- Day7 (TukeyHSD; p-value=6.26E-4) and
- Day10 (TukeyHSD; p-value=3.83E-7).

Amoxicillin-clavulanic acid group
No significant difference
Differentially abundant taxa (16S rDNA)

**Enterobacterales**

**Enterococcus**

- Amoxicillin-clavulanate
- Tebipenem pivoxil hydrobromide
Differentially abundant taxa (16S rDNA)

**Bacteroides**

**Clostridiales**

**Lactobacillus**

**Bifidobacterium**

- Amoxicillin-clavulanate
- Tebipenem pivoxil hydrobromide
Recovery of the microbiome

- Beta-diversity analysis shows microbiome significantly impacted in both treatment groups with most subjects undergoing recovery after treatment.
- OTU composition of many samples belonging to the beginning of treatment were similar to follow-up samples (i.e., Day 21, Day 90, Day 180).

Principal coordinate analysis (PCoA) plot based on Bray-Curtis measure where each point represents a sample.
Selection or emergence of resistance

- Selection of *Candida spp.* relatively higher in tebipenem group
  - 40.0% in the tebipenem group vs. 11.8% in the amoxicillin-clavulanic acid group

- Selection of *Clostridioides difficile* was low
  - 2.2% in the tebipenem group vs. 0% in the amoxicillin-clavulanic acid group

- Selection of resistant strains of *Enterobacterales* was low in both treatment groups:
  - Cefotaxime resistance - 3.7% for tebipenem group vs. 17.0% for amoxicillin-clavulanate group
  - Meropenem resistance - 5.4% for tebipenem group vs. 1.48% for amoxicillin-clavulanate group
  - Tebipenem resistance - 4.4% for tebipenem group vs. 12.5% for amoxicillin-clavulanate group
  - Very low emergence of decreased susceptibility to tebipenem observed in tebipenem group (*E. coli*, n=1 and *Enterobacter bugandensis*, n=1)
Conclusion

- Effects of tebipenem were more pronounced against *Enterobacterales* and *Enterococcus* spp. compared to amoxicillin/clavulanate but reverted to baseline after 14-21 days
  - Quantitative culture showed more clear recovery compared to the 16S rDNA method

- No apparent impact on *Bifidobacterium* spp. or *Lactobacillus* spp. between treatment groups with minimal difference observed in *Bacteroides* spp.
  - Microbiome balance either unchanged or recovered

- Emergence of resistance low
  - Colonization with MDR pathogens during treatment period (diminished colonization resistance) less likely with intended outpatient use
  - 16S rDNA method provide no information regarding emergence of resistance
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