METRC
Major Extremity Trauma Research Consortium

Microbial interspecies associations in open fracture and subsequent infection using Next Generation Sequencing

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BACKGROUND

Multispecies interactions and polymicrobial biofilm formation (on foreign material as well as bone/soft tissue) are poorly understood but are increasingly thought to be prognostically important. Co-occurring microbes can mutually influence behavior, physiology and environment in profound ways that can directly impact outcomes. Interactions can modulate virulence factor expression, antibiotic sensitivity, bacterial dispersal, bacterial adhesion, foraging, reproduction, chemical warfare and defense.

PURPOSE AND HYPOTHESIS

Aim: Evaluate the co-occurrence or clustering of microbial taxa in open fracture and subsequent fracture related infection wounds using data derived from more sensitive NGS.
Hypothesis: Microbial species that tend to co-occur are likely to demonstrate synergistic and potentially clinically relevant inter-microbial interactions

MATERIALS AND METHODS

This is a secondary analysis of patients enrolled in the METRC Bioburden Study with polymicrobial NGS results (n = 94 total, 35 baseline open fracture patients and 53 follow-up infection or nonunion patients). Composite specimens were obtained after prep/drape at beginning of definative fixation procedure and at any follow-up surgery for a complication. NGS performed using 16S rRNA hypervariable region ith V1-V3 primers. Number of co-occurrences in this cohort was totaled to create a heat map.

Figure 1: Baseline definitive fixation open fracture wound microbial species co-occurrence heatmap

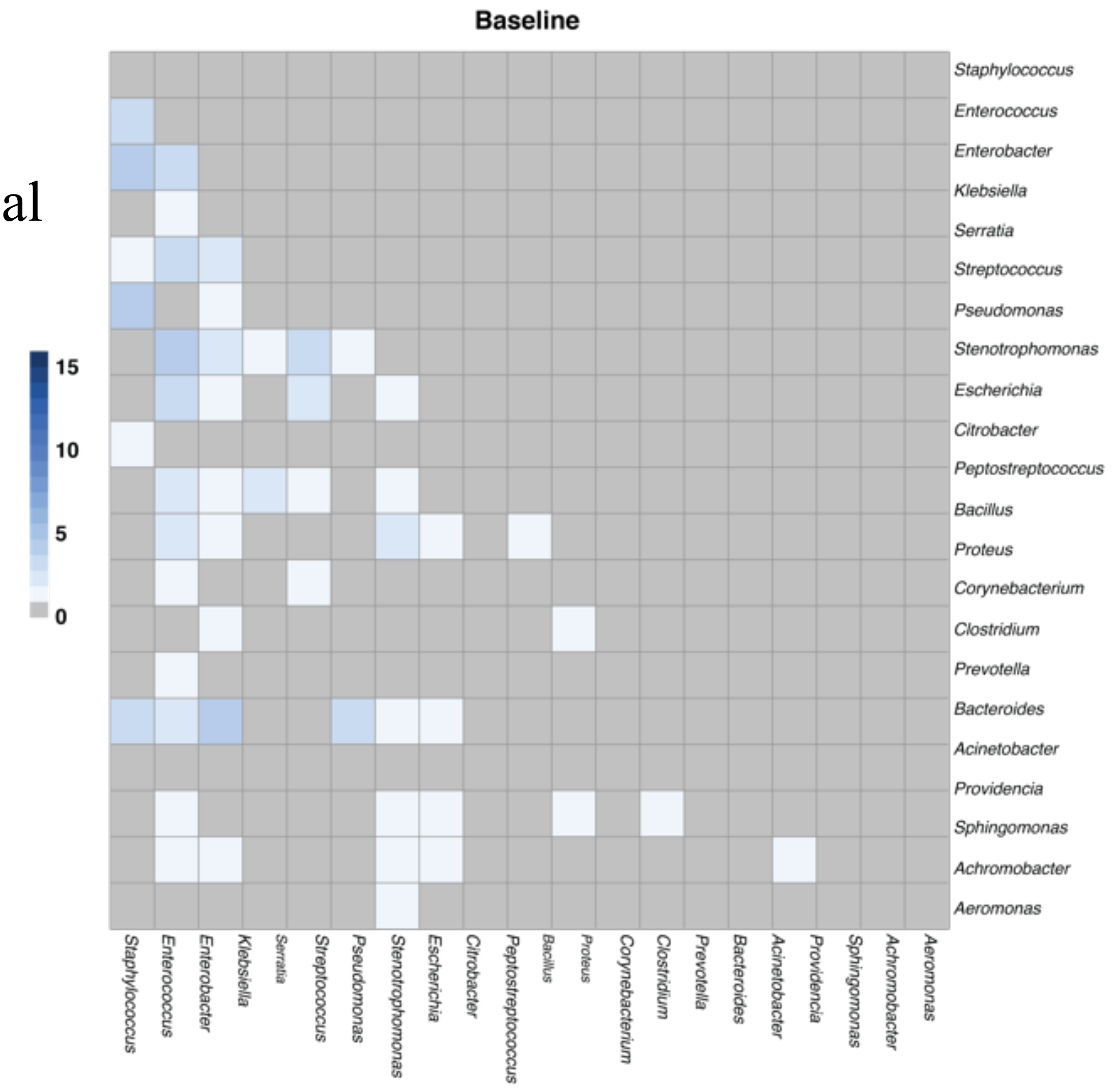


Figure 2: Follow-up infection/nonunion wound microbial species co-occurrence heatmap

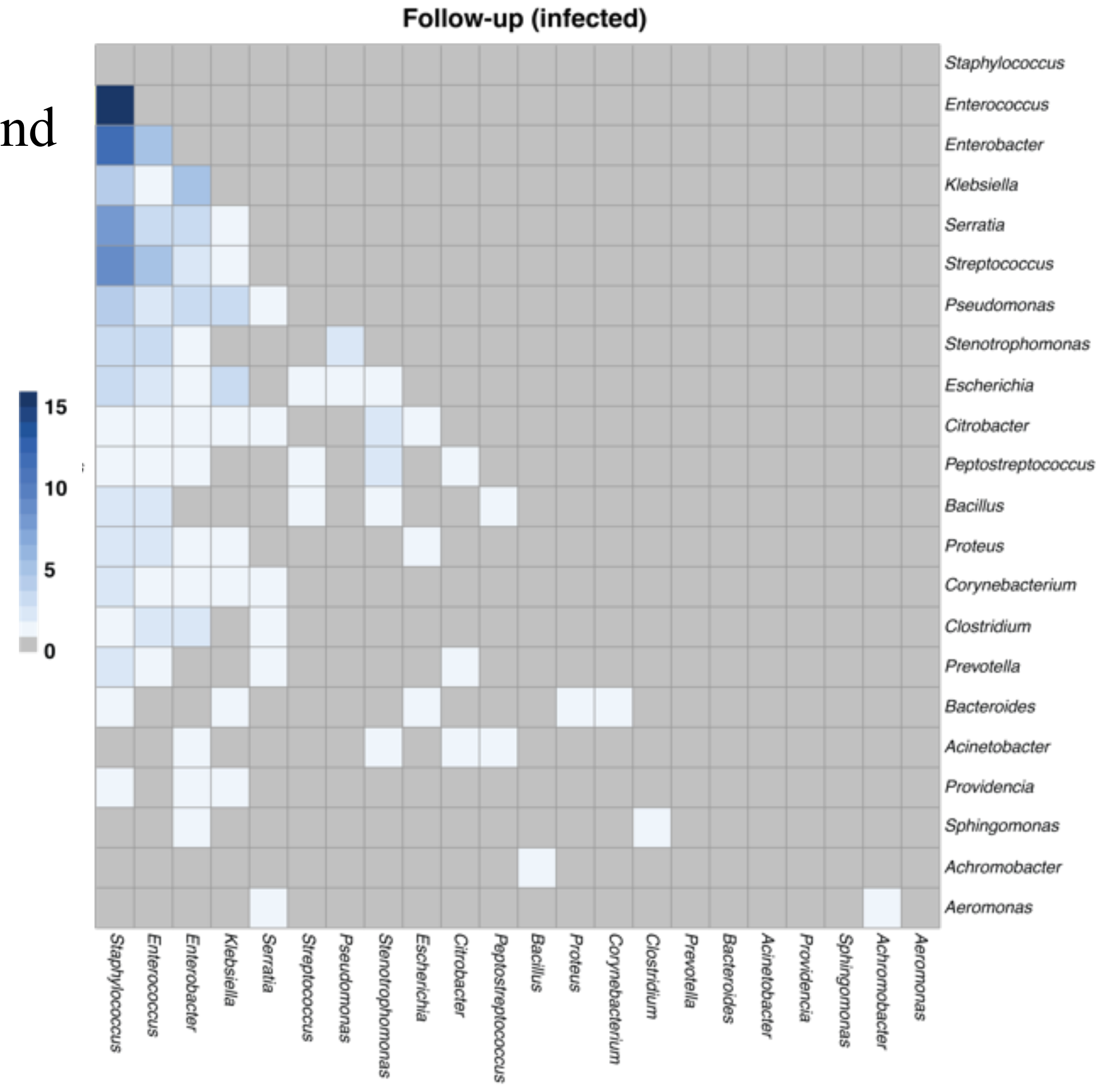
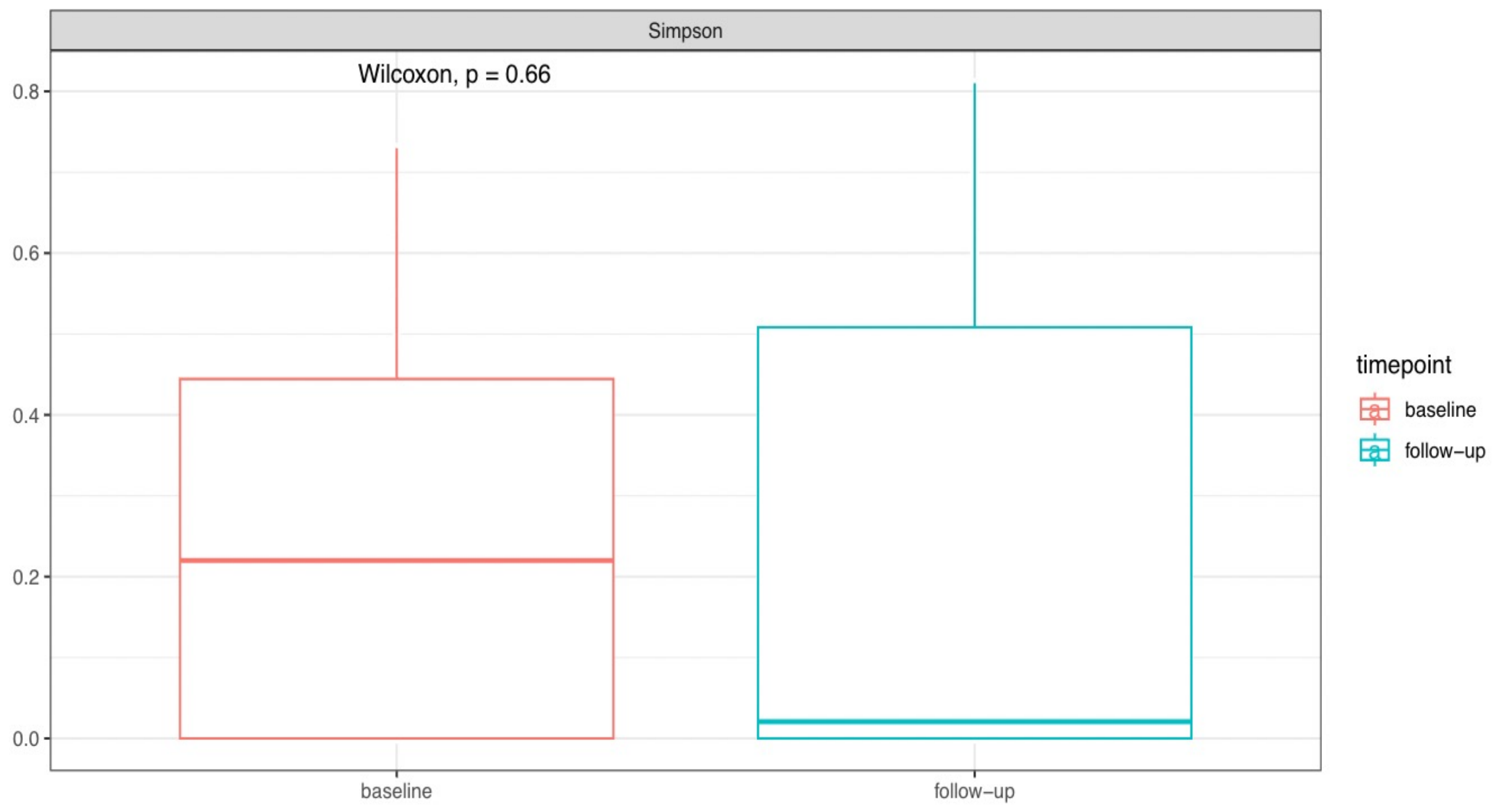


Figure 3: Simpson Diversity at baseline and follow-up



RESULTS

Among polymicrobial open fracture wounds, there were no microbe species pairs that co-occurred together in more than 4 patients. However, there were many low-frequency (51 unique) co-occurring pairs of species (Fig. 1). In follow-up specimens collected in nonunion or infection, there were four dominant co-occurring microbial species that were each seen in 10-15 patients (Staphylococcus-Enterococcus, Staphylococcus-Enterobacter, Staphylococcus-Serratia and Staphylococcus-Streptococcus) (Fig. 2). There is a trend from higher Simpson Diversity in baseline specimens to lower Simpson Diversity in follow-up specimens, which reflects the idea that the community is becoming dominated by a few species and demonstrating decreased evenness (Fig. 3).

CONCLUSIONS

The present study indicates that that among polymicrobial specimens, there was a conversion from a diverse array of polymicrobial species in baseline open fractures to more dominant Staphylococcus-Enterococcus, Staphylococcus-Enterobacter, Staphylococcus-Serratia or Staphylococcus-Streptococcus co-occurring species. These frequent microbial co-occurrences in patients with infection likely represent an evolutionary or survival benefit associated with these microorganisms. Understanding and disrupting these antimicrobial relationships could result in improvement in both prevention and treatment for infection.

INVESTIGATOR AFFILIATIONS

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