Comparison of stool versus rectal swab samples from long-term acute care hospital (LTACH) patients for microbiota analysis

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Abstract

Background: Stool is the standard sample type for human gastrointestinal microbiota analysis. However, stool collection from in-patients can be challenging, making collection of rectal swabs a more feasible sampling approach. In this study, we compared the microbiota from stool and rectal swab samples to assess differences due to sample collection method.

Methods: Stool was collected into a sterile container immediately after a bowel movement. Subsequently, rectal swab samples were collected within 10 minutes, and at 3, 6, and 12-27 hours later. Rectal samples were collected using a Dacron swab that was inserted 1-2 cm past the anal verge, then transferred to liquid Stuart medium for transport (BBL). Samples were stored at 4°C for 1-26 hours, and then frozen at -80°C until analyzed. Total DNA was isolated using the Mo Bio PowerMag Soil DNA isolation kit. The V4 region of the 16S rRNA gene was PCR-amplified and sequenced using Illumina technology.

Results: Eight stool and 30 swab samples were collected from 8 in-patients at one LTACH. Inter-individual microbiota differences were assessed by analysis of molecular variance (AMOVA) of the Yue and Clayton dissimilarity index (θYC). Grouping samples by patient regardless of sample collection method demonstrated significant differences among all patients (p-values: <0.001-0.010) compared to no overall differences between all stool and swab samples collected from individual patients (p-value: 0.976). Overall, microbial communities were similar in stool and swab samples collected from a patient, although minor shifts in the relative abundance of dominant organisms were noted over the 12-27-hour window. The θYC dissimilarity index between stool and swab samples was significantly lower within individuals (median 0.17, IQR 0.17) than between individuals (median 0.93, IQR 0.12) (Wilcoxon test p-value: <0.0001) indicating minimal differences between stool and swab samples collected from the same individual over the sampling period.

Conclusion: Minor differences in the microbial community were observed between stool and rectal swab samples over a 12-27-hour period. Rectal swabs are a suitable alternative to assessing the stool microbiota when fresh stool material is difficult to obtain.

Methods

• Rectal swabs are routinely used in healthcare facilities to screen for carriage of multidrug-resistant organisms due to their ease of collection by healthcare staff compared to stool.
• Gastrointestinal microbiome research has been performed using freshly passed stool or intestinal mucosa samples.
• The use of rectal swabs to study the human gut microbiota has not been well established.

Results

• 8 stool samples and 30 swab samples were collected from 8 subjects.
• 6/8 (75%) of subjects were male with a median age of 55 years (IQR, 47-58).

Figure 1. Principal coordinates analysis (PCoA) of θYC distances between bacterial communities of stool and swab samples. Samples grouped by study subject (same color) regardless of collection method demonstrate significant differences between all subjects (AMOVA p-values: <0.001-0.029) compared to no overall difference between all stool and swab samples collected from individual subjects (AMOVA p-value: 0.976).

Figure 2. The θYC dissimilarity index between stool and swab samples was significantly lower within subjects (median 0.17, IQR 0.17) than between subjects (median 0.93, IQR 0.12) (Wilcoxon test p-value: <0.0001). This indicates that there were minimal differences present between stool and swab samples collected from the same subject over the sampling time period up to 27 hours after a bowel movement.

Conclusions

• Despite minor differences in the relative abundance between stool samples and rectal swabs, microbial communities were similar in these two sample types within subjects.
• Rectal swabs are a suitable alternative to stool samples for assessment of human gastrointestinal microbiota when fresh stool material is difficult to obtain.

References


Acknowledgements

• We would like to thank the study subjects, administration, medical staff, nursing staff, the infection preventionist at the LTACH and the Microbial Systems Laboratories at the University of Michigan.

This project is funded through the CDC Prevention Epicenters Program under Cooperative Agreement #US4C400016-0351.