**INSIGHTS Analysis Plan (Outline)**

1. Generate Severity Groups - Brandon
   1. Test BU model for
      1. Clinical vs. Quality of Life
      2. *If i is significant,* Quality of Life vs. Psychosocial
   2. *If i and/or ii are significant,* Integrate quality of life and/or psychosocial measures into severity scores. (MFA)
2. Descriptive Statistics:
   1. Overall
   2. Current Ulcer(s) vs. Past Ulcer(s) vs. No Ulcers
   3. Hydroxyurea vs. No Hydroxyurea
   4. Sickle Cell vs. Diabetes
   5. Severity Groups
3. Characterize Microbial Population (See Morgan and Huttenhower, 2012)
   1. Relative Abundance
   2. Functional Classification Enrichment (dependent on sequencing method… but could also do a variation of Gene Set Enrichment Analysis)
   3. Population Diversity
4. Compare Abundance and Diversity Measures Between:
   1. Current vs. Past vs. No Ulcer Groups
   2. Hydroxyurea vs. No Hydroxyurea
   3. Sickle Cell vs. Diabetes
   4. Severity Groups

**NEXT…**

1. Compare Clinical Measures Between:
   1. Current vs. Past vs. No Ulcer Groups
   2. Hydroxyurea vs. No Hydroxyurea
2. Compare Quality of Life/Psychosocial Measures Between:
   1. Current vs. Past vs. No Ulcer Groups
   2. Hydroxyurea vs. No Hydroxyurea
3. Compare Microbiome Measures Between:
   1. Current vs. Past vs. No Ulcer Groups
   2. Hydroxyurea vs. No Hydroxyurea
   3. Severity Groups
4. *Integrate Microbiome Measures into Severity Scores (?)*
5. Generate Polygenic Score (PGS) from Genome Sequence Date (curated gene interaction networks 🡪 talk to Niraj)
   1. *Integrate PGS into Severity Scores (?)*
6. Generate Coexpression Networks from RNA-Seq Data
   1. Compare with PGS
   2. Compare with Microbiome Measures
7. Evaluate Differential Coexpression Network Connectivity Between:
   1. Current vs. Past vs. No Ulcer Groups
   2. Hydroxyurea vs. No Hydroxyurea (if possible)
   3. Severity Groups (if possible)