Atypical Pyogenic Infections in Cats from an Institutionalized Hoarding Facility

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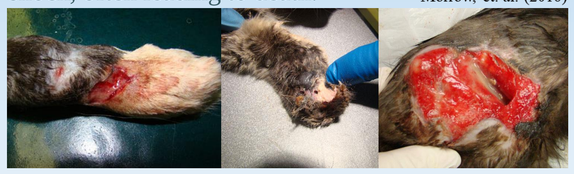
Frankie's Friends Cat Rescue

Morrow, et. al. (2016)

Introduction

Over 200 cats removed from an institutionalized hoarding facility (IHF) demonstrated severe, atypical pyogenic infections. Multiple pyogenic syndromes were documented, including cervical lymphadenitis with abscessation unassociated with any wounding in 51 cats, acute rhinitis with profuse purulent nasal discharge in 68 cats, and abscesses of the paws and carpal/tarsal regions in 82 cats. Many others exhibited septic arthritis with total joint destruction, necrotizing fasciitis, meningitis, otitis, and septic shock, often leading to death.

Morrow, et. al. (2016)



Clinical samples were collected and sent for culture and sensitivity to commercial laboratories. Culture showed the presence of Beta-hemolytic Streptococci in some samples, although many produced no growth and attempts at speciation were not consistent. Additional samples and deceased cats were stored at -20ºC for later identification of bacterial species through DNA sequencing. Initial results showed *Streptococcus canis* (*S. canis*) as the only bacterial species or dominant species identified and the only species present in the purulent material (Fig. 1). The objective of this study was to evaluate tissues from cats found dead in the freezers at the IHF and those that were removed from the IHF but died shortly thereafter through Next Generation Sequencing (NGS) of bacterial DNA and compare those data to sequences generated via Sanger sequencing.

Materials and Methods

Tissues were collected from the lungs, heart, liver, spleen, and kidneys of deceased cats found frozen at the IHF and those who died shortly after removal from the facility. DNA was extracted (Boom et. al., 1990). PCR with universal bacterial primers were used to amplify all bacterial DNA from the samples (Cardinale, M. et al) and products evaluated via gel electrophoresis. Cloning and Sanger sequencing were performed followed by use of the Basic Local Alignment Search Tool (BLAST) to determine the identity of the bacteria. Next Generation Sequencing was performed on the Illumina MiSeq platform, sequences were analyzed, and taxa were identified to the genus level via the UCLUST algorithm and Greengenes v. 13.8 database.

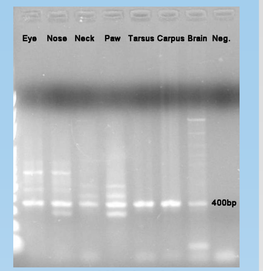


Figure 1 – 400bp band Sanger sequenced as *S. canis*. Morrow, et. al. (2016)

Results

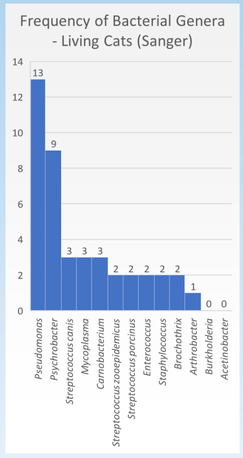


Figure 2: Sanger sequencing results in living cats showing relatively high frequency of *Pseudomonas, Psychrobacter, and Streptococcal spp.*

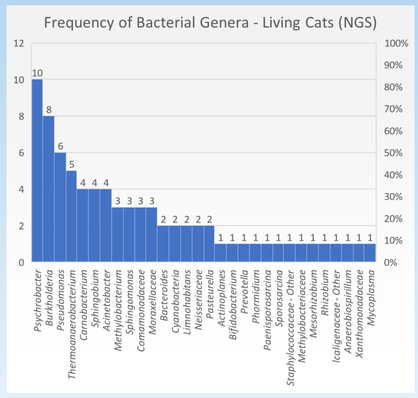


Figure 3: NGS results in living cats showing > 50% frequency of *Pseudomonas, Burkholderia*, and *Psychrobacter*.

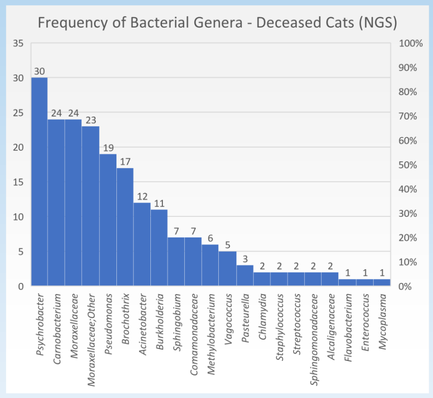


Figure 4: NGS results in living cats showing >50% frequency of *Pseudomonas, Burkholderia, Psychrobacter, Moraxellaceae* (undetermined genera), and *Brochothrix*.

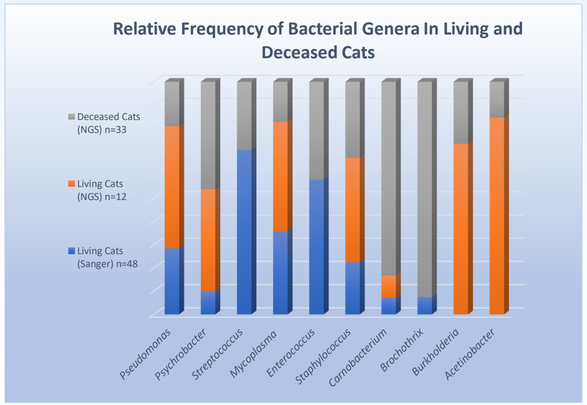


Figure 5: Relative frequency of select genera in living (Sanger and NGS) and deceased cats.

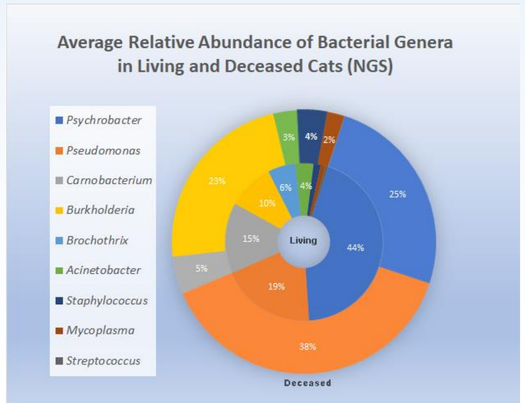


Figure 6: Relative abundance of select genera in living and deceased cats (NGS data only).

Conclusions

Streptococcus: Found only in living cats, with a higher frequency in the sequences obtained from Sanger compared to NGS of living cats (very low relative abundance).

- Bias in earlier sample collection possible - NGS cats were not specifically chosen.
- *Streptococcus* may be predominantly in peripheral regions, not the thoracic and abdominal organs.

Psychrobacter: Found in all groups of cats at high frequency - almost all of the NGS living and deceased cats. Also found at high relative abundance (44% of bacterial genera found in living cats and 25% in deceased cats). Unknown pathogenic potential.

Pseudomonas: Found in all groups of cats, with the highest frequency in NGS living cats. Also at a high relative abundance (19% in living cats and 38% in deceased cats). Known pathogenic potential.

Carnobacterium: Found mainly in NGS deceased cats. Unknown pathogenic potential.

Burkholderia: Found with relatively high frequency in NGS living cats. Associated with eye rupture (unpublished data). Found in internal organs in this study.

- Need to investigate if it was in the lungs and/or other organs and pathogenic potential in those organs.

Brochothrix and *Acetinobacter*: Low relative abundance. Unknown pathogenic potential.

References

Boom, R. C. J. A., Sol, C. J., Salimans, M. M., Jansen, C. L., Wertheim-van Dillen, P. M., & Van der Noordaa, J. P. M. E. (1990). Rapid and simple method for purification of nucleic acids. *Journal of clinical microbiology*, *28*(3), 495-503.

Cardinale, M., Brusetti, L., Quatrini, P., Borin, S., Puglia, A. M., Rizzi, A., ... & Daffonchio, D. (2004). Comparison of different primer sets for use in automated ribosomal intergenic spacer analysis of complex bacterial communities. *Appl. Environ. Microbiol.*, *70*(10), 6147-6156.

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