



VALIDATING PCR QUANTIFICATION FOR PATHOGEN DETERMINATION IN CSOM WITH HIRA-TAN METHOD

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Introduction

- Pathogen identification has been challenging in Chronic Suppurative Otitis Media (CSOM). The recurrent nature, negative culture and misuse antibiotic in developing countries may conceal the true pathogen.¹
- The pathogen identification by quantitative PCR with human cell as the internal control (HIRA-TAN method) in Community Acquired Pneumonia (CAP) can be similar applied.²
- This study objective was to preliminary investigate the utility of HIRA-TAN method in CSOM pathogen identification.

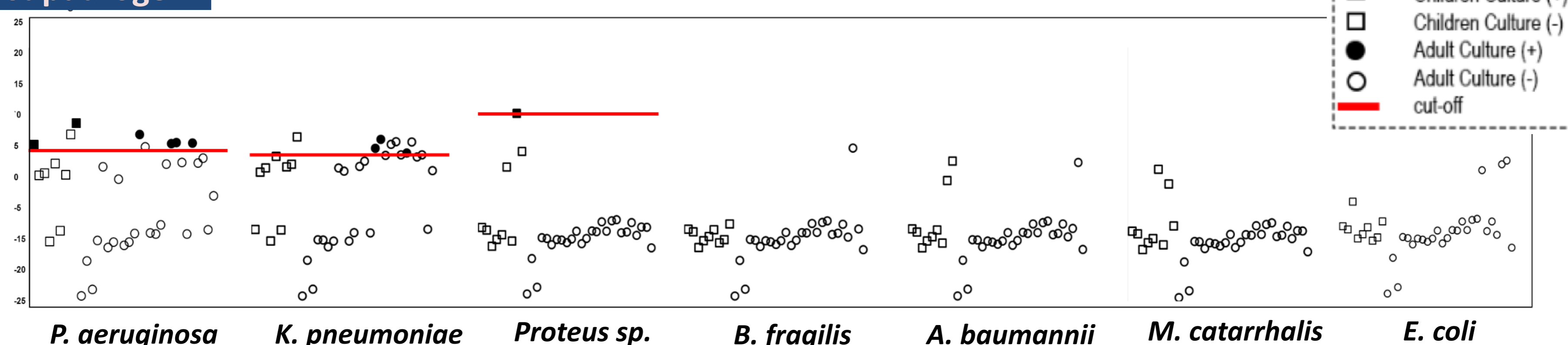
Material & Method

During December 2016 to January 2017, all CSOM patients in ENT outpatient clinic were enrolled. The otorrhea swab collection was managed for microbiological culture and HIRA-TAN method.

A multiplex TaqMan assay were performed with 16 common pathogens. The cycle threshold difference between pathogen and human (ΔC_t pathogen) was the index for defining the pathogen.

The ROC curve analyses was performed to validate the cut-off values.

ΔC_t pathogen



Result

Thirty-nine patients ranging 1.7 to 62 years old positive culture in 14 samples (35.9%) (Fig. 1).

The ΔC_t pathogen cut-off for *P. aeruginosa* were 3.33 (90%, 100%); *K. pneumoniae* were 1.71 (85%, 100%); and *Proteus sp.* were 8.29 (90%, 100%).

Multiple pathogen detected with negative culture result were *B. fragilis*, *A. baumannii*, *M. catarrhalis*, and *E. coli*.

Conclusion

The cut-off values for *P. aeruginosa*, *K. pneumoniae*, and *Proteus sp.*, from this study can be useful for similar study.

Multiple other pathogen detection and other cut-off values investigation require further larger study.

Reference

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Discussion

Most pathogens were detected by the PCR method omitting *M. morganii* → different finding between PCR & culture can be occurred.³

DNA within a specimen can be from nonviable bacteria and may be not clinically relevant. DNA from viable bacteria can be detected by PCR for weeks but DNA from nonviable bacteria do not persist > 1 day.^{4,5}

Positive PCR result indicating that viable but not culturable bacteria → can be existed in otitis media

Culture Result

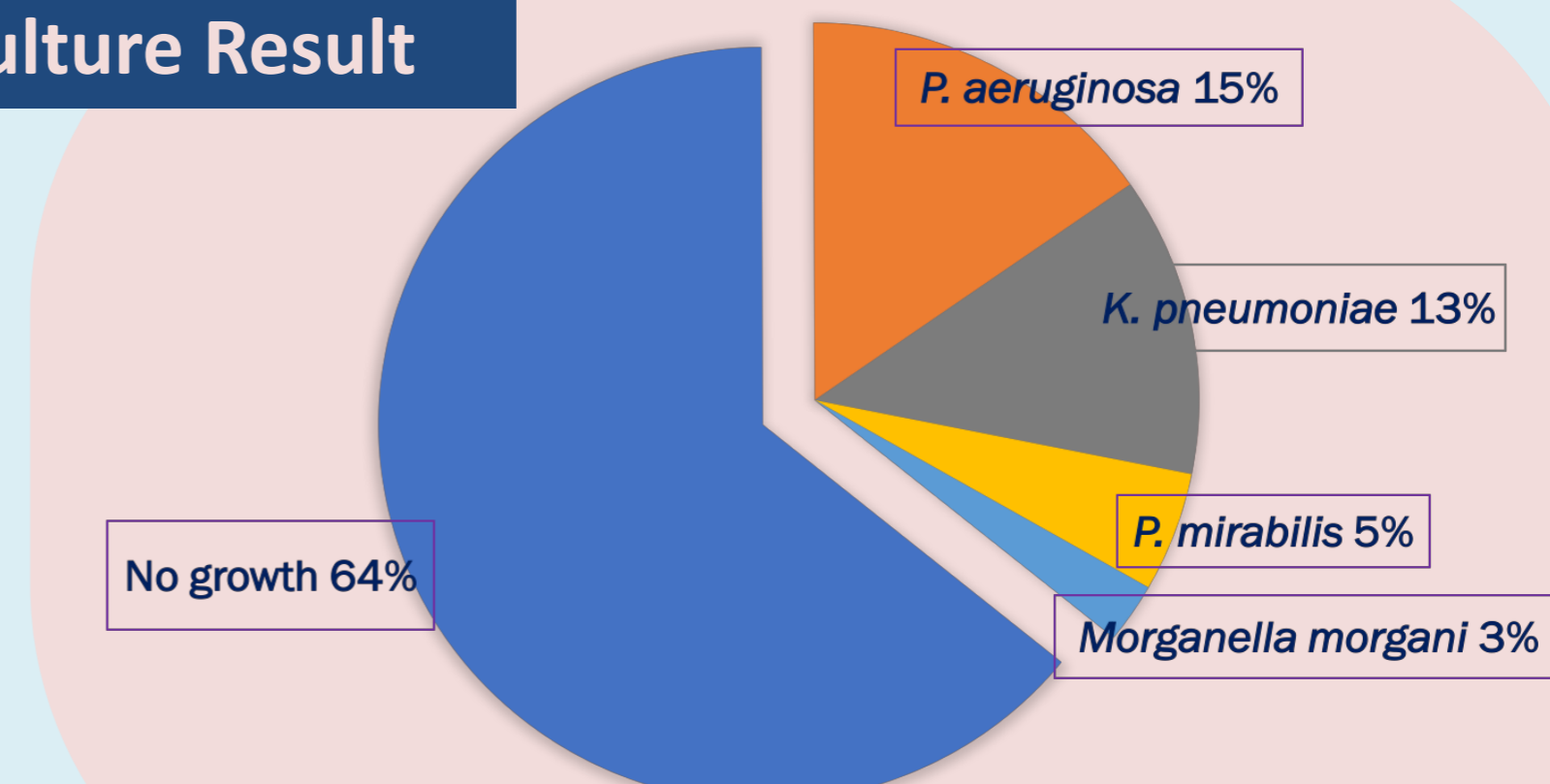


Fig. 1