

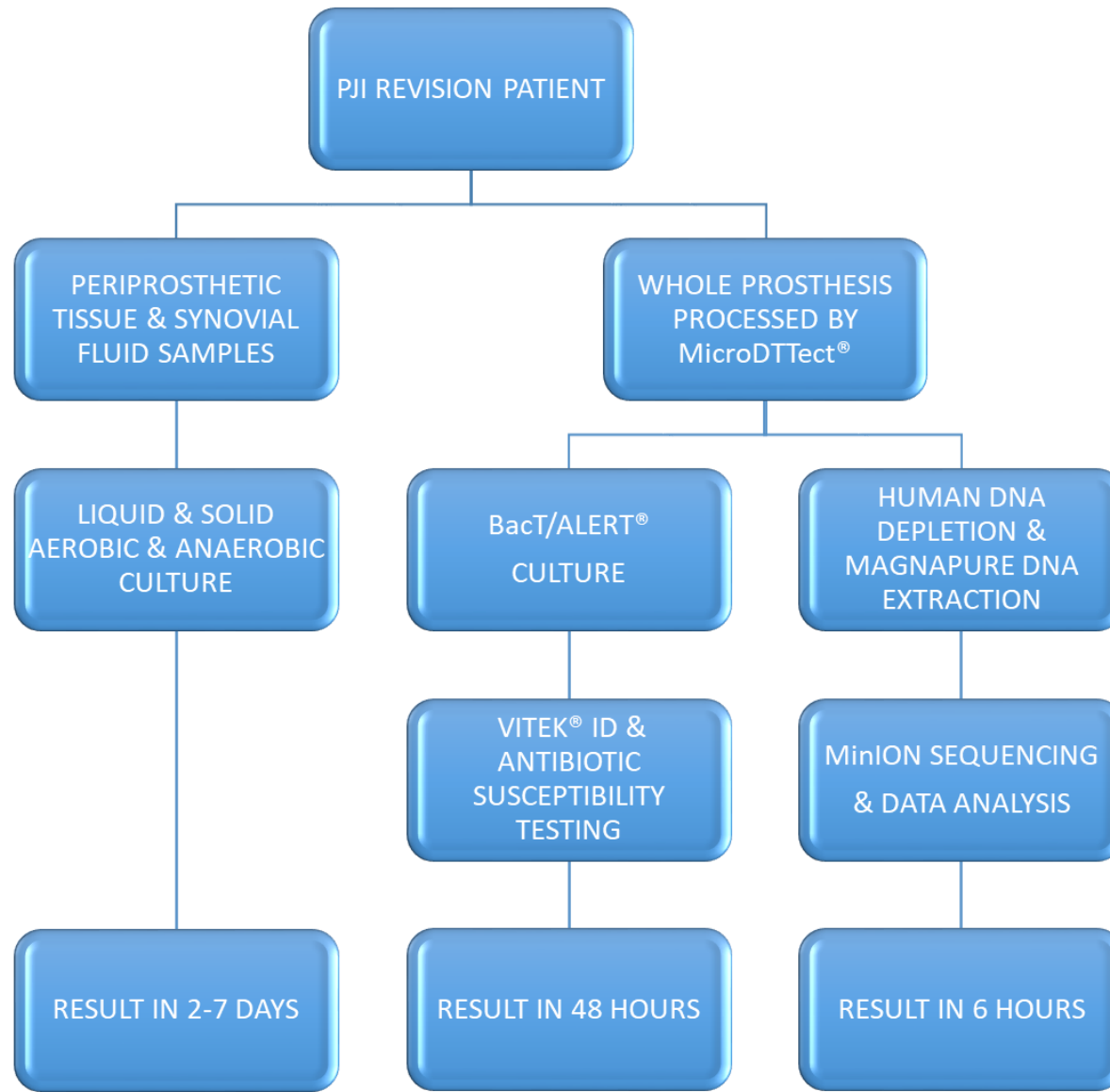
Investigating the utility of MicroDTect® and rapid metagenomic sequencing for the detection of prosthetic joint infection

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Introduction

- Gold standard for the diagnosis of prosthetic joint infection (PJI) in the UK is culture of multiple periprosthetic tissue and synovial fluid samples, which is time consuming and has poor sensitivity.
- Fastidious organisms can be difficult to culture and it is challenging to distinguish causative from commensal bacteria.
- We evaluated whole prosthesis culture using MicroDTTect® compared to routine culture.
- We developed a novel metagenomics pipeline for the diagnosis of PJI capable of pathogen detection within 6 hours.

Methods






Results

Sample No.	1	2	3	4	5	6	7	8	9	10	11	12	13
Routine Culture	negative	negative	<i>P. aeruginosa</i> , <i>C. tuberculostearicum</i>	negative	<i>C. acnes</i>	negative	<i>P. mirabilis</i>	negative	<i>S. lugdunensis</i>	negative	negative	negative	negative
MicroDTTect Fluid Culture	negative	negative	<i>P. aeruginosa</i>	negative	negative	negative	<i>P. mirabilis</i>	negative	<i>S. lugdunensis</i>	<i>S. epidermidis</i>	negative	negative	<i>S. epidermidis</i>
Metagenomics	negative	<i>S. pneumoniae</i>	negative	negative	negative	negative	<i>P. mirabilis</i>	negative	<i>S. lugdunensis</i>	negative	negative	negative	negative

Sample No.	14	15	16	17	18	19	20	21	22	23	24	25	26
Routine Culture	negative	negative	negative	negative	negative	<i>S. anginosus</i>	negative	<i>S. aureus</i> , <i>C. acnes</i>	<i>S. epidermidis</i>	<i>C. striatum</i> , <i>F. magna</i>	negative	<i>S. epidermidis</i> , <i>C. striatum</i>	<i>S. epidermidis</i>
MicroDTTect Fluid Culture	negative	negative	negative	<i>S. warnerii</i>	negative	<i>S. anginosus</i>	negative	<i>S. aureus</i>	<i>S. epidermidis</i>	<i>S. haemolyticus</i>	negative	<i>S. epidermidis</i>	<i>S. epidermidis</i>
Metagenomics	negative	negative	negative	<i>S. warnerii</i>	negative	<i>S. anginosus</i>	negative	<i>S. aureus</i>	<i>S. epidermidis</i>	negative	negative	<i>S. epidermidis</i>	<i>S. epidermidis</i>

Metagenomics	Sensitivity	Specificity
Compared to Routine Culture	50%	87.5%
Compared to MicroDTTect Culture	66.67%	92.86%

Full Concordance 
 Minor Discordance 
 Major Discordance 

Discordant Results

Compared to Routine Culture	MicroDTTect® Culture	Metagenomics
Full Concordance	18	19
Minor Discordance	3	2
Major Discordance	5	5

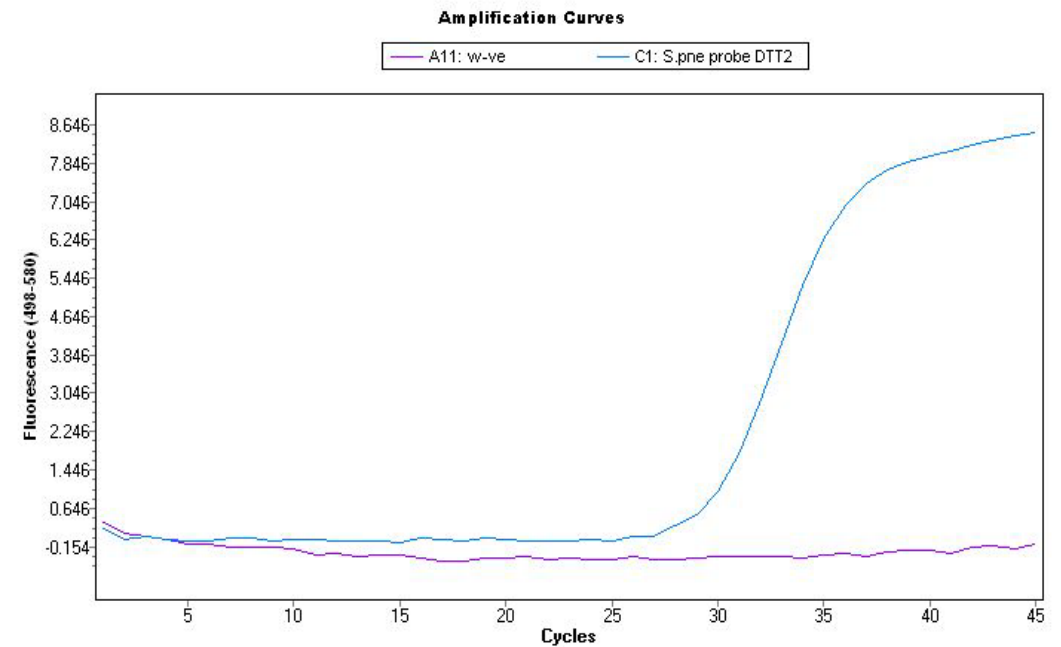
Sample	Organism	RC	DTT	MGS
2	<i>S. pneumoniae</i>			X
3	<i>P. aeruginosa</i>	X	X	
	<i>C. tuberculostearicum</i>	X		
5	<i>C. acnes</i>	X		
10	<i>S. epidermidis</i>		X	
13	<i>S. epidermidis</i>		X	
17	<i>S. warneri</i>		X	X
21	<i>S. aureus</i>	X	X	X
	<i>C. acnes</i>	X		
23	<i>C. striatum</i>	X		
	<i>F. magna</i>	X		
	<i>S. haemolyticus</i>		X	
25	<i>S. epidermidis</i>	X	X	X
	<i>C. striatum</i>	X		

Major Discordant Results – Sample 2

Sample	Organism	RC	DTT	MGS
2	<i>S. pneumoniae</i>			X

Sample tested with <i>S. pneumoniae</i> probe	qPCR Crossing Point (CP)
Negative Control	-
Sample 2	29.53

Taxon ⇅	Cumulative Reads ▾
Homo sapiens	29,584
Streptococcus pneumoniae	836
Escherichia coli	119
Clostridium perfringens	101
Citrobacter amalonaticus	37
Klebsiella pneumoniae	14
Cutibacterium acnes	13
Streptococcus mitis	12
Bacillus subtilis	12
Stenotrophomonas maltophilia	12
Moraxella osloensis	10
Comamonas testosteroni	8

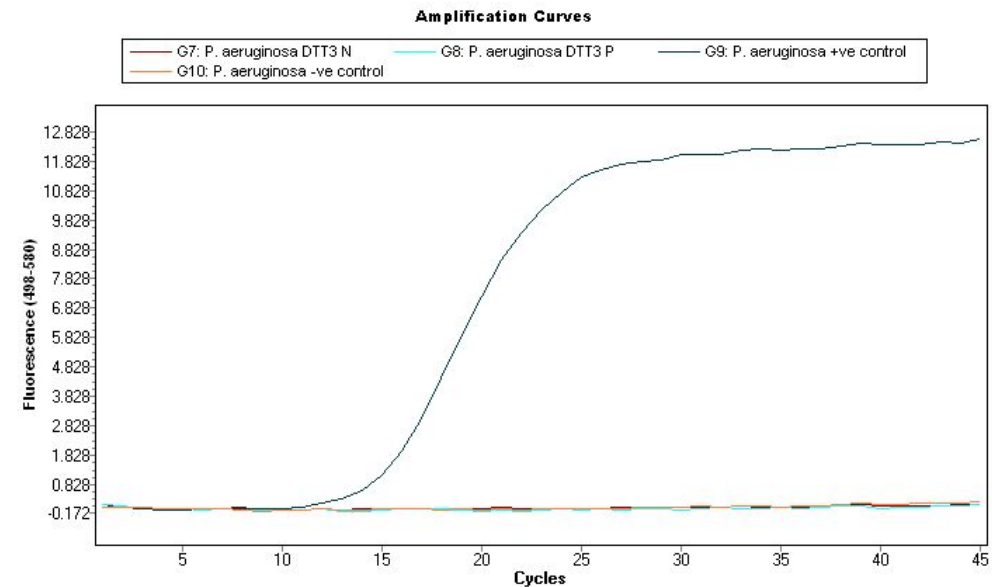


Major Discordant Results – Sample 3

Sample	Organism	RC	DTT	MGS
3	<i>P. aeruginosa</i>	X	X	
	<i>C. tuberculostearicum</i>	X		

Sample tested with <i>P. aeruginosa</i> probe	qPCR Crossing Point (CP)
Water Negative Control	-
<i>P. aeruginosa</i> DNA Positive Control	14.95
Sample 3 - Processed	-
Sample 3 - No Depletion	-

Taxon	Cumulative Reads
Homo sapiens	23,784
Escherichia coli	112
Clostridium perfringens	23
Klebsiella pneumoniae	17
Citrobacter amalonaticus	14
Bacillus subtilis	10
Raoultella ornithinolytica	9
Arthrobacter sp. IHBB 11108	7
Cutibacterium acnes	7
Stenotrophomonas maltophilia	6
Mucilaginibacter sp. PAMC 26640	4
Comamonas testosteroni	4



Conclusions

- Both MicroDTTect® culture and metagenomics have high specificity compared to culture and provide results within a considerably shorter timeframe, but low sensitivity.
- Discordant result analysis suggests that many of the organisms not detected by metagenomics are likely to be skin contaminants (e.g. *C. acnes*, *Corynebacterium*).
- Metagenomics can provide whole pathogen genome information in a total turnaround time of 6 hours.

Future Work

- Obtaining and testing more samples.
- qPCR analysis and patient follow-up to confirm the presence of pathogens in discordant results.

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