Sensitive Molecular Identification of Pathogens causing Implant and Tissue Infections (ITI)

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Objectives

Prosthetic infection is the most severe complication in joint arthroplasty. The diagnostic procedure is time consuming and in many cases unrewarding. Microbial growth can be slowed or suspended if the pathogens are weakened by antimicrobial therapy. Molecular diagnostics is a reliable complement for optimizing conventional microbiology by detecting bacteria that do not grow in culture. We used compact sequencing, a combination of highly-sensitive Polymerase Chain Reaction (PCR) with hybcell based identification, to detect and identify pathogenic bacteria or fungi in clinical tissue and synovial fluid samples. These samples were tested in parallel to bacterial culture in combination with identification by MALDI-TOF. Our aim was to test the suitability of compact sequencing to detect and identify pathogens as an additional standard method to diagnose implant or tissue infections.

Method

52 samples were tested: 40 samples from joints (shoulder, elbow, hip and knee), 4 from ascites and 8 from other sources. Samples from infected tissue were divided into two aliquots. One was used for bacterial culturing on agar and in blood culture bottles for 6 weeks. The second aliquot was stored deep frozen (- 80° C) and later used for pathogen DNA extraction with SelectNA (Molzym, DE). The detection and identification of bacteria and fungi were done by compact sequencing based on hybcell ITI DNA xA (Cube Dx, AT).

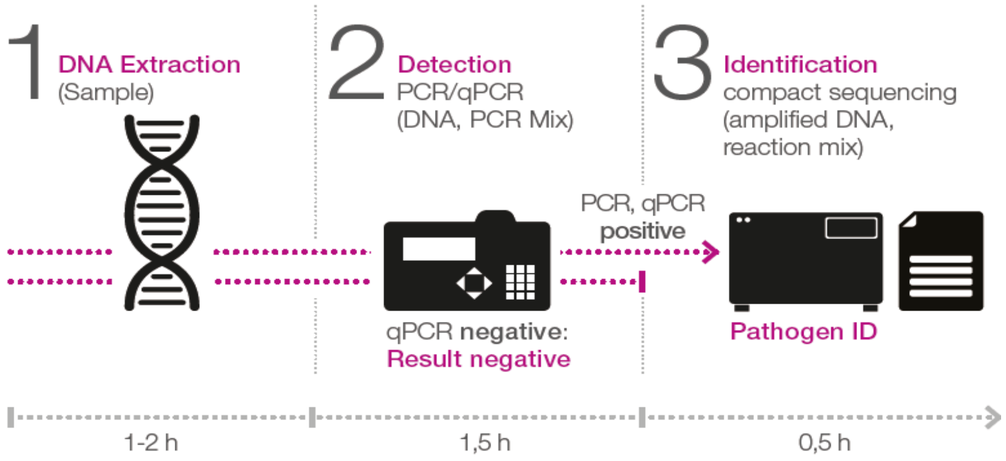
Results

The Cube Dx test shows a sensitivity of 93% and a specificity of 63%, a negative and positive predictive value of 96% and 50%, respectively and an accuracy of 71% in comparison to microbiological evaluation. The total number of samples testing positive for bacteria and fungi was higher for compact sequencing with 53% (28 of 52 samples) than for bacteriological culture with 29% (15 of 52 samples).

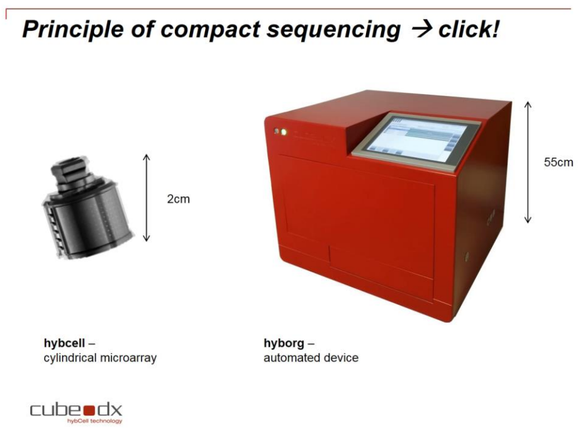
Conclusion

Our results demonstrate a good correlation between molecular and cultural detection. In 27% (14 of 52 samples) of all cases, molecular testing was more sensitive than culture. hybcell ITI DNA xA is well suited for use as an additional sensitive diagnostic tool for critical clinical samples in a routine lab.

No.	Source	Culture	hybcell ITI DNA xA
1	abdominal cavity	Candida albicans	Streptococcus anginosus; Klebsiella oxytoca, Candida albicans
2	abscess cavity	Pseudomonas aeruginosa, Enterococcus faecalis, Bacteroides fragilis, Bacteroides thetaiotaomicron	Bacteria pan
3	lymph node	Streptococcus salivarius	Streptococcus (genus)
4	secretion from urethra	Coagulase negative staphylococcae	Staphylococcus haemolyticus, Anaerococcus (genus), Corynebacterium (genus)
5	serom latissimus	neg	Nectriaceae
6	upper arm	Staphylococcus aureus	Staphylococcus aureus
7	upper arm	Staphylococcus aureus	Staphylococcus aureus
8	ascites	neg	Bacteria pan
9	ascites	neg	neg
10	ascites	neg	neg
11	elbow	Staphylococcus aureus	Staphylococcus aureus
12	elbow, bursa	neg	neg
13	hip joint	neg	neg
14	knee joint	Candida albicans	Enterococcus faecalis, Candida albicans
15	knee joint	Staphylococcus aureus	Staphylococcus aureus
16	knee joint	Staphylococcus aureus	Staphylococcus aureus
17	knee joint	Streptococcus pyogenes	Streptococcus pyogenes
18	knee joint	Streptococcus pyogenes	Streptococcus pyogenes
19	knee joint	Corynebacterium ssp.	neg
20	knee joint	neg	Anaerococcus (genus), Corynebacterium (genus)
21	knee joint	neg	neg
22	knee joint	neg	Candida (genus)
23	knee joint	neg	Streptococcus (genus)
24	knee joint	neg	Anaerococcus (genus), Corynebacterium (genus)
25	shoulder joint	Staphylococcus epidermidis	Staphylococcus (genus)
26	shoulder joint	neg	neg
27	knee joint	neg	neg
28	knee joint	neg	Staphylococcus aureus
29	knee joint	neg	Staphylococcus aureus
30	knee joint	neg	neg
31	knee joint	neg	neg
32	knee joint	neg	neg
33	knee joint	neg	neg
34	perikard	neg	neg
35	knee joint	Candida parapsilosis	Candida (genus), Candida dubliensis, Candida tropicalis
36	knee joint	neg	Streptococcus (genus)
37	hip joint	neg	neg
38	knee joint	neg	neg
39	shoulder joint	neg	neg
40	knee joint	neg	Bacteria pan
41	knee joint	neg	neg
42	knee joint	neg	Streptococcus (genus)
43	knee joint	neg	neg
44	knee joint	neg	Candida (genus)
45	knee joint	neg	neg
46	knee joint	neg	neg
47	shoulder joint	neg	neg
48	knee joint	neg	neg
49	aszites	neg	Bacteria pan
50	hip joint	neg	Streptococcus agalactiae
51	knee joint	neg	neg
52	shoulder joint	neg	neg



Workflow of hybcell ITI DNA xA.



hybcell \ Culture	pos.	neg.	Σ
pos.	14	14	**28**
neg.	1	23	**24**
Σ	**15**	**37**	**52**

Results and correlation of culture and hybcell.