

## Supporting Information

### A Pragmatic eLCR for Ultrasentive Detection of Methicillin-Resistant

#### *Staphylococcus Aureus* in Joint Synovial Fluid: Superior to qPCR

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**Table of content**

Gel electrophoresis.....Page S3

DNA extraction, sequencing and qPCR.....Page S3

Table S1.....Page S4

**Gel electrophoresis.** To prepare the hydrogel with a percentage of 3.5 %, 3.5 g of agarose, 100 mL 1× TBE buffer (Sangon Biotechnology) and 5 µL volume of 10000× Gelred dye (Biotium) were mixed together and heated for 2 min in microwave oven. The gel was then cooled to room temperature and soaked in a 1× TBE buffer prior to use. A mixture of sample (8 µL) with 6× loading buffer (2 µL) was added to 3.5 % agarose gel and underwent electrophoresis in 1× TBE at a constant voltage of 150 V for about 45 min at room temperature. After the electrophoresis, the gel was scanned using a FluorChem R system (ProteinSimple, San Jose, CA).

**DNA extraction, sequencing and qPCR.** The genomic DNA was extracted from the standard strain freshly cultured on blood agar plates and the joint synovial fluid of PJI patient using the Ezup Column Bacteria Genomic DNA Purification Kit (Sangon Biotechnology) according to the manufacturer's instructions.

PCR assay was carried out in a 25 µL reaction mixture containing 1 µL volume of 10mM dNTP mix, 1 µL volume of 10 µM forward and reverse primers, 2.5 µL volume of 10× Taq Buffer (with MgCl<sub>2</sub>), 1 U Taq enzyme and 2 µL volume of extracted DNA

template. Water was used as no-template control (NTC). The assay was performed in the PCR instrument (ABI Company Limited, USA) as follows: polymerase activation at 95 °C for 5 min, followed by 38 cycles of denaturation at 95 °C for 30 s, annealing at 58 °C for 30 s, elongation at 72 °C for 60 s, and final extension at 72 °C for 10 min. In order to confirm the sequence of the amplified *mecA* gene, the PCR products were sequenced using ABI 3730XL DNA sequencer (ABI, Life Technology, CA, USA). For further absolute determination of the copy number of *mecA* gene, DNA extracts were used as templates to quantify target gene in the SG Fast qPCR Master Mix (High Rox, Sangon Biotechnology) by qPCR (ABI StepOnePlus, Life Technology).

**Table S1.** Calculated copies of *mecA* gene

Clinical sample	Mean Ct value (n=3)	Mean copies (n=3)
Patient 1 (P1)	26.8734531	453.816925
Patient 2 (P2)	26.3219376	666.455505