PRECLINICAL EVALUATION OF PHENOTYPIC mariAST® MRSA RUO TEST AGAINST ROUTINE MRSA SCREENING METHODS IN HOSPITAL SETTING

Petri Antikainen[1], Roosa Savolainen[2], Suvi-Sirkku Kaukoranta[2], Jori Soukka[1], Janne O. Koskinen[1]

[1]ArcDia International Ltd, Turku, Finland; [2]Department of Clinical Microbiology, Vaasa Central Hospital, Vaasa, Finland

Introduction

mariAST® (ArcDia International Ltd, Finland; www.arcdia.com) is the only published pipeline platform that allows rapid and real-time phenotypic antimicrobial susceptibility testing (AST) directly from clinical samples containing commensal flora. mariAST® combines in-well culture and separation-free detection of live bacteria with specific antibody reagents in real-time. It is based on the same separation-free ArcDia™ TPX fluorescence technique for bioaffinity assays as the multianalyte mariPOC® identification test system **(Figure 1).**

mariAST® MRSA RUO (Research Use Only) test has shown to detect MRSA directly from screening samples in hours with sensitivity down to 1 CFU/well (50 CFU/ml). Performance of the test was studied against MRSA screening methods used routinely in Vaasa Central Hospital (Finland).

Figure 1. mariPOC® test system for rapid and multianalyte identification of pathogens

Materials & Methods

Primary MRSA screening in Vaasa Central Hospital is done by selective chromogenic culture (CHROMagar™ MRSA). The presence of resistance gene (*mecA* or *mecC*) from MRSA suspected colonies is confirmed by a PCR. The susceptibility of MRSAs against cefoxitin is examined by disk diffusion testing.

Leftover MRSA culture samples (N=667) from routine screening were tested with the mariAST® MRSA RUO test. Majority of the samples were nasal, pharyngeal and perineum swabs which were pooled into growth medium prior to mariAST® analysis (N=533). Others were e.g. swab samples from wounds, groins and axilla. Before mariAST® analysis, the swab samples in gel transport tubes were stored in +4 °C usually not more than overnight.

For automated mariAST® analysis, the swab samples were suspended into TSB medium (2.4 ml) and analyzed in batches once a day. In the mariAST® test, the growth of *Staphylococcus aureus* in the presence of cefoxitin (4 mg/l) antibiotic is monitored (**Figure 2**) in 20 µl volume without pure culture. Because of very suboptimal temperature (+26 °C) in the primary mariAST® analysis, MRSA positive samples stored at -20 °C were reanalyzed retrospectively in controlled temperature at +35 °C.

Results

Routine screening detected 43 MRSA positive samples with cefoxitin inhibition zone diameters ranging from six to 19 mm (EUCAST cefoxitin breakpoint R<22 mm). In the prospective analysis, the mariAST® test detected 32 (74%) culture positive samples with median time of eight hours. All missed MRSA samples showed low bacterial counts on culture plates.

In the retrospective analysis at +35 °C, further six samples were detected as MRSA positives leading to overall sensitivity of 88% (38/43). The optimized temperature reduced median time for positive results by 2–3 hours. The specificity of the mariAST® RUO test was 91% (569/624).

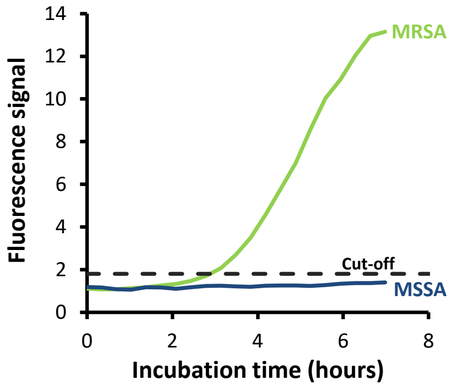

Figure 2. mariAST® detection of MRSA and MSSA

Discussion

This preclinical study evaluated the performance of the mariAST® MRSA RUO test and its suitability for current MRSA screening routines. The mariAST® test could provide results within the same day of sampling and 24–48 hours faster than the current routines.

The sensitivity and the result times improved when optimal growth temperature was used. The sensitivity can be further improved, for example, by analyzing bigger sample volume which would increase the likelihood to have live bacteria in a test well. The specificity of the test can be improved by introducing higher specificity antibodies and/or selective growth agents into the culture media.

The cefoxitin breakpoint concentration of 4 mg/l set by the EUCAST seems to be appropriate for rapid screening of MRSAs and resistance testing directly from clinical samples. In addition, mariAST® could also suit for MIC determination and susceptibility testing with different antibiotics.

mariAST® platform differentiates from other rapid AST concepts as it is suited truly for direct testing of clinical samples, not just for positive blood cultures. The analyzer is suitable for 24/7 decentralized use needed in the future to cope with common infections caused by multiresistant bacteria.

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