Rapid identification of OXA-23-subfamily in carbapenem-resistant Acinetobacter spp. with a novel immunochromatographic lateral flow assay

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Introduction

The global spread of carbapenem-resistant Acinetobacter spp. has led to an emerging worldwide healthcare problem. The carbapenem-hydrolysing 

oxacillines (OXAs) are the most commonly reported carbapenem-resistance determinants in Acinetobacter spp., particularly in A. baumannii. There are six identified OXA-subgroups associated with carbapenem-resistance in A. baumannii: the intrinsic OXA-51-like and the acquired OXA-23-like, OXA-68-like, OXA-40-like, OXA-143-like and OXA-235-like. Of these, OXA-23 is the most prevalent carbapenem-resistance determinant among isolates in Germany, Europe and worldwide.

The lack of effective and reliable tests to detect OXA-mediated carbapenem-resistance is a serious challenge to modern medicine. There is an unmet medical need for reliable and rapid diagnostic tools to detect OXA-23-like producing strains to ensure a successful treatment of patients and prevent the spread of carbapenemase-producers.

The aim of this work is the development an antibody-based OXA-23-like

Selection of hybridoma clones

Mice were immunized with purified recombinant OXA-23. After a standardized immunization protocol, one mouse was sacrificed, spleen was processed, and generated splenocytes were fused with myeloma cells to generate hybridomas. Hybridoma clones were picked into 96-well plates.

Conclusions

- no expensive or specialized equipment is required to use this test strip
- antibody-based OXA-23 detection assay is able to detect OXA-23-mediated carbapenem-resistant Acinetobacter spp. with 100% specificity
- sensitivity has been determined to detect between 10^6 to 10^9 cfu per sample, making a point-of-care device feasible
- result in < 20 min which saves 12-48 hours in diagnostic time, avoiding treatment with inappropriate antibiotics and enables earlier intervention to control transmission of OXA-23 producing carbapenem-resistant Acinetobacter spp.

Specificity of OXA-23 prototype

A well characterized collection of carbapenem-resistant Acinetobacter spp. isolates (n=108) with defined carbapenem resistance mechanism were used to evaluate specificity of our OXA-23 prototype.

Results of ten representative test strains

- OXA-23-producer (carb+)
- Other carb-

Limit of detection is between 10^6 and 10^9 cfu