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A single-tube approach for in vitro diagnostics using diatomaceous earth and optical sensor

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ABSTRACT

Versatile, simple and efficient sample preparation is desirable for point-of-care testing of emerging diseases such as zoonoses, but current sample preparation assays are insensitive, labour-intensive and time-consuming and require multiple instruments. We developed a single-tube sample preparation approach involving direct pathogen enrichment and extraction from human specimens using diatomaceous earth (DE). Amine-modified DE was used to directly enrich a zoonotic pathogen, *Brucella*, in a large sample volume. Next, a complex of amine-modified DE and dimethyl suberimidate was used for nucleic acid extraction from the enriched pathogen. Using our single-tube approach, the pathogen can be enriched and extracted within 60 min at a level of 1 colony formation unit (CFU) from a 1 ml sample volume in the same tube. The performance of this approach is 10–100 times better than that of a commercial kit (10^2 to 10^3 CFU/ml) but does not require a large centrifuge. Finally, we combined the single-tube approach with a bio-optical sensor for rapid and accurate zoonotic pathogen detection in human urine samples. Using the combination system, *Brucella* in human urine can be efficiently enriched (~8-fold) and the detection limit is enhanced by up to 100 times (1 CFU/ml bacteria in urine) compared with the commercial kit. This combined system is fast and highly sensitive and thus represents a promising approach for disease diagnosis in the clinical setting.

Keywords: Bio-sensing, In vitro diagnostics, Sample enrichment and extraction, Single tube approach, Zoonosis

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