CORRESPONDENCE

Detection of adenovirus and rotavirus antigens by an immuno-gold lateral flow test and ultrasound-enhanced latex agglutination assay

Viral gastro-enteritis is a significant cause of infant morbidity and mortality in the developing world, with group A rotavirus causing an estimated 800 000 childhood deaths each year [1]. Viruses responsible for gastro-enteritis include rotavirus, adenovirus and members of the Caliciviridae. Viral antigen detection is achieved through enzyme-linked immunosorbant assay, latex agglutination tests (LATs) and more recently, immuno-gold lateral flow tests (LFTs). The sensitivity of LFTs for the detection of rotavirus has been reported to be comparable to enzyme immunoassay and superior to LATs [2].

The sensitivity of conventional test-card LATs is improved by concentration of the antibody-coated latex beads in a non-cavitating ultrasonic standing wave [3]. It has been shown that rotaviral antigen detection can be enhanced with standing wave ultrasound [4]. We compared the sensitivity of a commercial LFT (Adenov/rotavirus) and Strip Quick Test, Coris Bioconcepts, Leeds, W. Yorks), two LAT kits (Rotascreen and Adenoscreen, Microgen Bioproducts, Surrey) and ultrasound-enhanced latex agglutination (using the test-card latex reagents) for detection of rotavirus and adenovirus antigens.

Test procedures for commercial kits were performed according to the manufacturers’ instructions. The ultrasound equipment and ultrasound test procedure have been described previously [5, 6]. Essentially, equal volumes of a 1 in 8 dilution (in PBS) of latex suspension and test sample (doubling dilutions of kit positive control antigen in PBS) were mixed and drawn into a 1-mm square internal dimension glass micropipette (Vitrocom, NJ, USA) and sealed as described in the Immunoscreen instruction manual (EMS Ltd, Wantage). The micropipette was located on the axis of a tubular transducer (Morgan Matroc, Wrexham) and sonicated for 60 s [5]. After sonication the sample was expelled on to a solid non-absorbing surface and the droplets were stirred five times (to dissociate aggregates not bridged by antigen) before microscopic examination [7].

The antigen detection limit (defined as the highest dilution of inactivated adenovirus and rotavirus antigen solution at which a positive result was observed) was determined by the lateral flow tests, the standard and the ultrasound-enhanced latex agglutination tests (Table 1).

The sensitivity of the lateral flow test was comparable with that of latex agglutination for detection of adenovirus antigen, but was greater than that of test-card agglutination for detection of rotavirus antigen. In clinical studies, LATs have been shown to be less sensitive than ELISA and EIA for detection of human rotavirus in stools [8]. Although LATs are suitable for rapid diagnosis, their use is limited when sensitive and accurate diagnosis is required. It has been shown here that the sensitivity of detection of purified viral antigen with commercial diagnostic microparticles can be increased 8-fold and 16-fold above that of a commercial lateral flow test for rotavirus and adenovirus, respectively. Ultrasonic enhancement of particle-based immunoassays has been demonstrated in clinical samples without occurrence of non-specific reactions [3]. Therefore, the increased sensitivity offered by ultrasound, as shown in this preliminary study, may have applications for clinical faecal samples.

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References


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<th>Table 1. Comparison of antigen detection limits as determined with the LFT, LAT and ultrasound-enhanced LAT</th>
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<td>Antigen</td>
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* Repeat experiments were performed on duplicate dilution series.

* A 1 in 8 dilution of test latex gave the maximum limit of detection.