

Detection of Viable *Cryptosporidium parvum* Based on Nucleic Acid Sequence-Based Amplification (NASBA) and Polydiacetylene Vesicles

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A novel method is described enables the detection of viable *Cryptosporidium parvum* by amplifying the RNA from viable *C. parvum* using nucleic acid sequence-based amplification (NASBA) and subsequent detection with polydiacetylene vesicles. The analytic target is a 121-nt sequence from *C. parvum* heat shock protein hsp70mRNA. The mRNA acting as a template for NASBA is produced in response to a heat shock. Once a brief heat shock is applied to the *C. parvum* oocysts, the nucleic acid is extracted from the cells, followed by purification and amplification in an isothermal water bath with NASBA, an efficient method to amplify specifically RNA molecules. The RNA product can be detected by hybridizing specific DNA probes immobilized on the polydiacetylene vesicles which give a blue-purple-pink-red transition depending on the analytic concentration. An CR (colorimetric response) value is calculated as an index to indicate the corresponding analytic concentration.