Title Molecular diagnostic tools for detecting arthropod contamination in stored products.

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## Abstract

Arthropod contamination in raw commodities and finished food products is significant for both commercial and human health reasons. Rapid and precise methods for detecting these arthropod pests in food and bulk commodities could benefit public health research and grain evaluation. DNA markers based on the polymerase chain reaction (PCR) were developed that can specifically detect the evidence of the mould mite, *Tyrophagus putrescentiae*, the rice weevil, *Sitophilus oryzae*, and the lesser grain borer, *Rhyzopertha dominica* at very low levels in food and grain. Dilution series analyses determined that a single mite or beetle could be detected at less than 10-9 equivalents for either fresh, frozen or dried samples. Single rice weevils were identified from fresh, frozen and dried samples, and the same primer set could detect contamination of the congeners maize weevil, *S. zeamais*, and granary weevil, *S. granarius*. Extreme sensitivity in some cases was attributed to the presumed high copy number of mitochondrial DNA in cells of these insects. Studies with contaminated food found that markers for rice weevils and lesser grain borers were easily detected at concentrations of 1 larva per 1000 g of wheat. Work on extraction methods for arthropod DNA at low densities from food, and for non-gel methods for detection and quantification of PCR products, is reported. These methods should lead to a practical detection tool for these and other pests in food.