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Rapid, Sensitive, And Quantitative Detection Of Ricin Toxin Using A Suspension Microsphere Array Immunoassay.

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The toxin Ricin, which is extracted from beans of the plant *Ricinus communis*, has been identified as a category B Select Agent and has been previously used as a biological weapon. While PCR-based assays can be used to detect the *R. communis* nucleic acid which remains associated with crude toxin preparations, direct toxin detection is critical for forensic testing and clinical applications. To this end, a sensitive and specific sandwich-type immunoassay was developed using fluorescent microspheres as solid support for capture antibodies. Monoclonal anti-Ricin antibody was covalently coupled to carboxylated polystyrene beads available from Bio-Rad. Polyclonal anti-Ricin antibodies, biotinylated and coupled with streptavidin phycoerythrin, were used as the detector moiety. The fluorescent microspheres were analyzed on a Bio-Plex analyzer (Bio-Rad Laboratories) following a protocol of less than four hours. Analysis of serial dilutions indicates the assay is quantitative through a three log range, with a limit of detection below 25 pg/ml. Evaluation of complex matrices, such as powder and environmental samples, will be presented. The ability to combine the Ricin-specific microspheres with other colored microspheres allows multiplexing and will facilitate generation of a toxin screening assay for other Select Agents such as botulinum toxin, staphylococcal enterotoxins, and *Clostridium perfringens* epsilon toxin. In combination with real-time PCR technology, microsphere array immunoassays will become critical in a public health response to biotreats.