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## ULTRASENSITIVE NANOGOLD-LABELLED IMMUNOASSAY FOR THE DETECTION OF SUGARCANE STREAK MOSAIC VIRUS

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Molecular assays, especially PCR based diagnostic assays are regularly employed in India to diagnose different viruses and phytoplasmas infecting sugarcane. Although these assays are sensitive, there is a need to develop field diagnostic kits to detect the pathogens. Lateral flow assay (LFA) using gold nanoparticles (GNPs) has been found to be a valuable assay for field diagnosis of viruses in different crop plants. In this study we have standardized an LFA for diagnosing viruses causing mosaic in sugarcane. In this assay, Sugarcane streak mosaic virus(SCSMV)specific antibodies were blotted and immobilized on nitrocellulose membrane in a spot, followed by incubation with suspected virus samples. After the incubation, the target viral antigen was allowed to hybridize with gold nanoparticles (GNPs) (~27nm) labeled SCSMV-specific antibody probes. Here, GNPs were synthesized by a standard citrate method and the gold labelled antibody probes were prepared by covalent conjugation using EDC/NHS linkers. Conjugation was confirmed by both UV-Vis spectral and gel retardation assays. Hybridization of the target with the nanogold labeled probes resulted in a change in spot colour. The detection limit was found up to one nanomolar concentration of SCSMV antigens in the samples. However, the detection limit can be further increased up to femtomolar concentration by the silver enhancement method. The new assay has been found to be fast and requires less consumables. Above all, it is portable and cost-effective. This new development in virus diagnostics would further strengthen production of healthy planting materials in sugarcane.