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(54) **Title:** MULTIPLEX IMMUNO SCREENING ASSAY

(57) **Abstract:** The present invention provides kits and assay methods for the early detection of pathogens, precise identification of the etiologic agent, and improved disease surveillance. More specifically, the present invention discloses an immunoassay leading to the rapid and simultaneous detection of antibodies to a wide range of infectious pathogens in biological fluids of infected patients. This immunoassay involves the covalent and oriented coupling of fusion proteins comprising an AGT enzyme and a viral antigen on an identifiable solid support (e.g. fluorescent microspheres), said support being previously coated with an AGT substrate. This coupling is mediated by the irreversible reaction of the AGT enzyme on its substrate. The thus obtained antigen-coupled microspheres show enhanced capture of specific antibodies as compared to antigen-coupled microspheres produced by standard amine coupling procedures. The methods of the invention possess the ability to multiplex, minimize the amount of biological sample, and have enhanced sensitivity and specificity toward target antibodies as compared with classical ELISA or Radio-Immunoprecipitation assays.



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