Abstract

Design and optimization of a Monkeypox virus specific serological assay

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Monkeypox virus (MPXV), a member of the Orthopoxvirus (OPXV) genus, is a zoonotic virus, endemic to central and western Africa that can cause smallpox-like symptoms in humans with fatal outcomes in up to 15% of patients. Incidence of MPXV infections in the Democratic Republic of the Congo, where the majority of cases occur, has been estimated to have increased as much as 20-fold since the end of smallpox vaccination in 1980. Considering the risk global travel carries for future disease outbreaks as well as the potential for use as a bioweapon, accurate epidemiological surveillance of MPXV is warranted. Early in illness, MPXV-related rash can be difficult to clinically differentiate from other rash illnesses such as varicella or other human herpes virus infections. Identification of OPXV infections can be done both serologically and by PCR; however, PCR is the only method that is currently capable of differentiating between OPXVs and can only be performed during active infections. Due to the high level of conservation within OPXV proteins, serological differentiation between vaccination and recent infection with MPXV or other OPXVs is difficult to ascertain. Comparative analysis of immunogenic proteins across human OPXVs, focusing on those that demonstrated differential reactivity between vaccinated (ACAM2000 vaccinia virus) and Congo Basin MPXV infected prairie dogs, identified a large subset of peptides that could potentially be specifically recognized during monkeypox infection. Peptides were chosen based upon MPXV sequence specificity and predicted immunogenicity, with preference given to those peptide sequences that contained multiple sequence substitutions that were predicted to be biologically significant. Peptides individually and combined were screened in an ELISA against serum from well characterized MPXV outbreaks, vaccinated sera, and smallpox sera. One peptide combination was successful with ~86% sensitivity and ~90% specificity. The performance of the MPXV peptide-based ELISA was assessed against the OPXV IgG ELISA in the context of a serosurvey by retrospectively screening a set of serum samples from the region in Ghana known to have harbored the MPXV infected rodents involved in the 2003 United States outbreak.

Epidemiology Protection of the public in relation to communicable diseases including prevention or control Public health or related research