to eliminate or neutralize interfering "natural inhibitors" that could be present in biological samples. In order to improve virus diagnostics we wanted to exploit the "non-self" recognition and binding properties of human apolipoprotein H (ApoH. ApoH binds and captures pathogens enabling their concentration from different kinds of biological samples. We have used magnetic beads coated with ApoH recombinant protein as a pre-treatment step for orthopox viruses to improve the detection threshold and to increase the sensitivity for diagnosis. With this approach virus was concentrated, DNA was extracted and subsequently detected and quantified by real-time PCR. After ApoH-treatment, Vaccinia Virus was detected from highly diluted samples where diagnosis had been negative with a standard DNA preparation protocol. At present the concentration and improved detection of other viruses with an ApoH-enhanced protocol is under investigation.

(23) ApOH-capture technology enhances Andes Hantavirus Detection allowing virus concentration from plasma and urine Samples of patients with acute Hantavirus Cardiopulmonary Syndrome.

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Background: Hantavirus cardiopulmonary syndrome (HCPS) is an emerging disease caused by new world hantaviruses. Hantaviruses (HV) are segmented RNA viruses belonging to the genus *Hantavirus* in the family *Bunyaviridae*. Hantavirus (HV) infections are mainly transmitted to humans by inhalation of virus-contaminated aerosols of rodent excreta and secretions, however, sporadic person-to-person transmissions of the Andes hantavirus (ANDV) have been reported. Based on the knowledge that human apolipoprotein H (ApoH), a constituent of human plasma, interacts with viral proteins, we wished to asses a possible interaction between ApoH and ANDV, the major etiological agent HCPS in South America.

Materials and Methods. Blood and urine samples from acute-HCPS patients were selected on the basis of their availability. Samples collected as part of the research initiative NIH/NIAID #AI 45452 were kindly supplied for this study. Donor patients met the clinical criteria for HCPS and harbored IgM antibodies reactive with hantavirus antigens. HV genomic RNA was confirmed in plasma by an in-house developed RT-PCR/hemi-nested PCR, using primers designed to partially encompass the S segment ORF of the Andes virus, strain CHI-7913. Samples used as negative control were collected among the laboratory staff. ApoH-coated magnetic beads and ApoH-coated ELISA plates used in this study were supplied by ApoH Technologies S.A. and used following their instructions.

Results: We report that ANDV interacts with ApoH, and that ApoH-coated magnetic beads or ApoH-coated ELISA plates can be used to capture and concentrate virus from serum and urine samples, allowing virus detection by both immunological and molecular approaches. We then developed an

ANDV-high throughput screen assay and assessed ANDV in urine samples, from 50 patients with acute ANDV-HCPS, collected during 5 days following hospitalization. 45 patients showed detectable amounts of ANDV in urine in at least one tested sample.

Conclusions: ApoH capture assay increases the sensitivity of virus detection by both molecular and immunological methods. This apparent enhancement in sensitivity most probably stems from the fact that virus is being concentrated from a larger sample volume. Additionally, we demonstrate that ANDV can be shed in the urine of infected individuals. Although, our data do not necessarily predict the presence of infectious virus in urine, the fact that ANDV is readily detected in urine samples of acute-HCPS patients not only lends support to the possibility that urine is a route for person-to-person transmission of HCPS but also raises the intriguing prospect that virus might be present in other biological secretions.

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