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*Session: Diagnosis*
*Date: Saturday, April 5, 2014*
*Time: 12:45-14:15*
*Room: Ballroom*

### Toward a rapid and accurate point-of-care test for active pulmonary tuberculosis: Multiplexed proteomic assay (SOMAscan™) of human serum for microbial and host markers

M.A. De Groote, L. Green, D. Sterling, R. Ostroff, N. Janjic, U. Ochsner

*Somalogic, Boulder, USA*

**Background:** A rapid, accurate, and inexpensive tuberculosis (TB) diagnostic test would allow earlier treatment and reduce transmission.

**Methods & Materials:** We used slow off-rate modified aptamers (SOMAmers) in a highly multiplexed proteomic assay (SOMAscan) to measure 1129 human proteins and 16 Mtb proteins in serum samples provided by the Foundation for Innovative New Diagnostics (FIND).

**Results:** Among the top host serum biomarkers distinguishing TB from non-TB regardless of the HIV status were kallistatin, TSP4, gelsolin, and CDON, which were lower in TB compared to non-TB, and LBP, ITI heavy chain H4, NPS-PLA2, and IP-10, which were higher in TB.

A 9-marker model performed well in a training set of 173 TB *vs*. 160 non-TB samples (sens 89% / spec 88%, AUC = 0.94), which was confirmed in a blinded verification set of 132 TB *vs*. 118 non-TB (sens 80% / spec 84%, AUC = 0.88). Mtb pathogen-specific SOMAmers showed non-specific background in serum and are pending analytical optimization to improve performance. Additional work to improve sensitivity of the host markers is ongoing.

**Conclusion:** The discovery of robust, quantitative, non-culture based diagnostic biomarkers of active pulmonary TB has great potential to facilitate the rapid and accurate diagnosis of TB disease. Our goal is that a combined host and microbial point-of-care diagnostic test could ultimately be tested and applied in peripheral microscopy centers or in primary care clinics.

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### Identification and evaluation of *Schistosoma mansoni* proteins as diagnostic targets for schistosomiasis

P. Ogongo

*Institute of Primate Research, Nairobi, Kenya*

**Background:** Demonstration of schistosome eggs in stool/urine is the definitive clinical test for clinical examination of schistosomiasis but this test has low sensitivity. Antibody based assays cannot distinguish between past and active infections thus unsuitable for follow up after drug administration while molecular techniques are expensive and unavailable at the point of care centers. Bioinformatics and Proteomics can be used to characterize schistosome proteins from different life cycle stages that include worm gut, worm tegument, egg secretions or released products of dead eggs that are released into the bloodstream and/or urine forming good diagnostic targets.

**Methods & Materials:** We have identified five schistosome proteins; Cathepsin B (Sm31), Asaparaginyl endopeptidase (Sm32), schistosome alpha-2 macroglobulin, Sm LMWP and *Sm*200 tegument protein using these approaches. Peptide sequences from these proteins synthesized as multiple antigenic peptides (MAPs) or conjugated to carrier proteins were used to immunize rats and rabbits. Serum from immunized rats were used to test the suitability of these targets using Enzyme Assays, Blot Assays and Immunocytochemistry with worm sections.

**Results:** Results show that antibodies to MAPs have good specificity for the peptides and native schistosome antigens while coupling to carrier proteins improves the immunogenicity of the peptides.

**Conclusion:** This approach has shown that these peptides are possible diagnostic targets that can be developed further to assess their sensitivity aiming at developing an assay capable of detecting the lowest number of worms in the host.

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### Rapid detection of ESBL-producing *Enterobacteriaceae* from blood cultures: A prospective study

P. Nordmann[1], L. Poirel[1], L. Dortet[2]

[1] *University of Fribourg, Fribourg, Switzerland*
[2] *Hopital de Bicetre, Kremlin-Bicetre, France*

**Background:** Enterobacterial strains producing clavulanic-acid inhibited extended-spectrum beta-lactamases (ESBLs) are increasingly reported worldwide. The rapid detection of ESBL-producing *Enterobacteriaceae* responsible for bacteremia is of utmost importance since their successful treatment depends on prompt administration of the appropriate antimicrobial agents. The ESBL NDP test has been evaluated here prospectively to detect ESBL-producing *Enterobacteriaceae* directly from blood cultures.

**Methods & Materials:** From November 2012 to May 2013, the ESBL NDP test, a rapid chromogenic test based on detection of cefotaxime hydrolysis, was performed with 96 blood cultures positive for Gram negatives. Results of the ESBL NDP test, obtained in less than 30 min, were compared to those obtained with the double disk diffusion technique. All ESBLs were then characterized at the molecular level. Identification of the Gram-negative bacteria was also performed directly on positive blood cultures using MALDI-TOF technology, and confirmed by a biochemical identification.

**Results:** Eighteen blood cultures infected with ESBL-producing *Enterobacteriaceae* (15 CTX-M producing *E. coli* and 3 CTX-M producing *K. pneumoniae*) were correctly detected, whereas 78