

mothers. Hypothetically, specific host-virus interactions account for the differential GpSGHV pathobiologies observed in different tsetse species and colonies. To test this hypothesis, we used mass spectrometry to investigate GpSGHV-induced protein expression modulations in the salivary gland (SG) proteomes of F1 progenies of two *Glossina* model species, *G. pallidipes* (SGH-susceptible) and *G. morsitans* (SGH-refractory). We identified 540 host proteins, of which 23 and 9 proteins were significantly up and down-regulated, respectively, in *G. pallidipes* compared to *G. morsitans*. We also detected 58 and 5 GpSGHV proteins in *G. pallidipes* and *G. morsitans*, respectively. Whilst *G. pallidipes* had significantly high GpSGHV titres, viral titres in the *G. morsitans* were insignificant, confirming that *G. morsitans* is largely SGH-refractory as compared to *G. pallidipes*. Finally we will discuss how GpSGHV seizes cohorts of intracellular signaling pathways to induce overt SGH in *G. pallidipes*, how robust immune responses block SGH expression in *G. morsitans*, and potential applications of our findings in management of viral infection in insect mass rearing facilities.

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#### The salivary gland proteome of *Glossina m. morsitans*, parasitized with *Trypanosoma b. brucei*

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*Trypanosoma brucei* spp, causative agent of African trypanosomiasis, completes metacyclo- genesis (development of mammalian-infective trypomastigotes) in the salivary glands (SGs) of its tsetse vector. Since metacyclic trypomastigotes are largely uncultivable, information on the molecular processes that underpin SG metacylogenesis is scanty. To bridge this knowledge gap, we employed LC-MS/MS to investigate protein expression modulations in SGs of *T. b. brucei*- infected and uninfected *Glossina m. morsitans*. We identified 361 (host) and 158 (parasite) proteins. Compared to uninfected SGs, the repertoire of the parasitized SG proteome contained proteins that were up-regulated (n = 276), down-regulated (n = 81) or un-modulated (n = 4). Whilst 11.5% (n = 32) of the 276 host proteins were significantly up-regulated, only one of the 81 proteins was significantly down-regulated. Despite high abundance, proteins associated with blood feeding process were down-regulated in parasitized SGs, probably to reduce feeding performance and thus promote vector competence (via increase of biting frequency). Amongst the differentially modulated host proteins in parasitized SGs were also proteins associated with translational regulation (protein translation, stabilization and degradation), immunity, homeostasis and cytoskeletal traffic. Notable proteins specific to metacyclic trypomastigotes included GPI- anchored surface glycoproteins kinetoplastid calpain, peroxiredoxin AhpC-type, *Trypanosoma* RHS multigene, membrane transporters and molecular chaperone protein families. These data will be discussed in view of strategy development to combat African trypanosomiasis via enhancement of tsetse *Trypanosoma*-refractoriness.

Contributed paper. Wednesday, 14:30, 156

#### Characterization of Bustos virus, a new member of the Negevirus group isolated from a *Mansonia* mosquito in the Philippines

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Mosquitoes are known to be important vectors for arthropod-borne viruses (arboviruses), which cause public health issues. In surveillance of mosquito-borne arboviruses, we isolated two distinct viruses from mosquitoes collected in Bustos Bulacan province, Philippines in 2009, where dengue fever was prevalent. These viruses show rapid replication and strong cytopathic effects in mosquito C6/36 cells. Whole-genome analysis of these viruses demonstrated that both viruses belong to the Negevirus group. One of the viruses, from *Culex vishunui* mosquitoes, is a new strain of Negev virus. The other virus, from a *Mansonia* sp. mosquito, is a new Negevirus designated Bustos virus. Gene expression analysis of the Bustos virus revealed that infected cells contain viral subgenomic RNAs that probably encode proteins from open reading frame (ORF)2 or ORF3. In Bustos virus-infected C6/36 cells, the ORF2 and ORF3 products were distributed in cytoplasm, whereas the ORF1 products formed foci nearby perinuclear region. Purified Bustos virus particles contained at least three proteins, and the major component is encoded by ORF3 and the minor component is encoded by ORF2. Bustos virus did not show infectivity to mammalian BHK-21 cells, suggesting an insect-specific virus.

Contributed paper. Wednesday, 14:45, 157

#### RNA activation in mosquito cells and its suppression by the dengue virus NS5 protein

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RNA activation (RNAa) is one of the emerging research areas in molecular biology, which involves small RNAs inducing gene expression by targeting the promoter. Thus far, RNAa has only been found in mammals, including humans, and *Caenorhabditis elegans*, but not in insects. Furthermore, there is no report about the effect of pathogen infection on RNAa. In this study, we employed dsRNA targeting the OpIE2 promoter with the GFP gene as the reporter, and checked its effect on GFP expression. In addition to that, the effect of dsRNA to the promoter on GFP expression was evaluated upon dengue virus infection. Our results clearly showed that dsRNA targeting the TATA box of the promoter could induce GFP expression in mosquito cells. In addition, dengue virus, in particular its non-structural protein 5 (NS5) could inhibit the RNA activation. The outcome of this research opens new avenues for RNAa-related research into insect's biology and its potential role in host-pathogen interactions.