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PRELIMINARY MICROARRAY ANALYSIS FOR THE IDENTIFICATION OF DENGUE-REFRACTORINESS CANDIDATE GENES IN MOSQUITO VECTORS

… **Gomez-Machorro**, Brent Harker, Diane Lovin, Becky … Jeanne Romero-Severson, David W. Severson

… Notre Dame, South Bend, IN, United States

Aedes aegypti is the primary vector for the yellow fever and dengue fever viruses. It is also an excellent laboratory model for investigating other mosquito-borne diseases. In this report we present preliminary results of our efforts to identify genes that are differentially regulated in *Ae. aegypti* in response to exposure to Dengue-2 JAM1409 virus between two laboratory strains: DS3 (susceptible) and Moyo-In-Dry (refractory). The microarray platform consists of spotted PCR amplicons representing unique cDNAs identified as part of the *Ae. aegypti* whole-genome sequencing project. Two point times were examined for this initial study: three hours and three days post feeding on both dengue-infected and uninfected blood meals. Transcriptional differences were evaluated in midgut tissues. Using stringent criteria, we identified twenty-eight genes at three hours and twelve genes at three days that showed significant differential expression between the DS3 and Moyo-In-Dry strains. Pending release of the annotated *Ae. aegypti* genome sequence will facilitate identification of putative gene functions. In addition, qRT-PCR validation of our microarray results will be presented. Additional experiments are ongoing to examine a more detailed time course following an infected blood feeding, and the potential for dengue serotype-specific gene response by the mosquito. Our goal is to identify clusters of co-expressed genes up- or down-regulated at different time points in response to the virus and correlate this response with dengue vector competence.

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A GEOGRAPHICAL SAMPLING STRATEGY FOR FIELD SURVEYS IN AN URBAN AREA USING HIGH-RESOLUTION SATELLITE IMAGERY

Adriana Troyo[1], Douglas O. Fuller[2], Olger Calderon-Arguedas[3], John C. Beier[4]

[1]Global Public Health Program, Department of Epidemiology and Public Health, University of Miami, Coral Gables, FL, United States, [2]Department of Geography and Regional Studies, University of Miami, Miami, FL, United States, [3]Centro de Investigacion en Enfermedades Tropicales, Departamento de Parasitologia, Facultad de Microbiologia, Universidad de Costa Rica, San Jose, Costa Rica, [4]Global Public Health Program, Department of Epidemiology and Public Health, University of Miami, Miami, FL, United States

Field evaluations for studying the epidemiology of vector-borne diseases like dengue in urban areas are often restricted to selection of households and buildings for field surveys. Therefore, the resulting sampling frame may exclude specific locations within the urban environment that contain vector habitats and thus may bias the results. A sampling strategy was developed for field surveys in an urban area using high-resolution satellite imagery. The site selected was Puntarenas, a city affected by dengue on the pacific coast of Costa Rica, for which high-resolution satellite imagery was available from the Advanced Spaceborne Thermal Emission and Reflection Radiometer (ASTER, 15 m spatial resolution) and QuickBird (0.6 m and 2.4 m spatial resolution for panchromatic and multispectral bands, respectively). Grids obtained from the ASTER imagery and a cover map generated from the QuickBird multispectral bands were used to determine the optimal grid area of 100 km^2, which contain 13±6 houses. A final grid 42 by 42 pixels (100.8 x 100.8 m) was created using the multispectral Quickbird imagery, and cells that had an area less than 90% within one specific locality of Puntarenas were excluded. The remaining cells were grouped according to locality and a random sample (10%) was selected from each. These cells would be used for field sampling of mosquitoes and larval habitats by evaluating the entire area within the geographical limits of each cell. To assess the suitability of the selected grid cells, the proportion of tree area ("tree" class Kappa = 0.91) was extracted for the individual cells from the QuickBird cover map. The mean percentage of tree cover in each locality and total area was compared between the selected sample cells and the total cells of the Puntarenas image. Overall, the sample adequately represented the total area and most of the individual localities in terms of tree cover. In 8 of 10 localities the difference between the estimate (sample) and the real percentage of tree cover was less than 3%. These results show that high-resolution satellite imagery and geographical information systems are useful in evaluating urban areas and selecting sections for field sampling of larval and adult mosquitoes that are practical, representative, and will reduce bias.

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SPATIAL ANALYSIS OF SPILL-OVER EFFECTS OF INSECTICIDE-TREATED MATERIALS IN A CLUSTER-RANDOMIZED TRIAL AGAINST *AEDES AEGYPTI* MOSQUITOES IN TRUJILLO, VENEZUELA

Neal Alexander[1], Audrey Lenhart[2], Elci Villegas[3], Michael Levy[4], Rana Moyeed[5], Axel Kroeger[6], P. J. McCall[2]

[1]London School of Hygiene and Tropical Medicine, London, United Kingdom, [2]Liverpool School of Tropical Medicine, Liverpool, United Kingdom, [3]Universidad de los Andes, Trujillo, Venezuela, [4]Emory University, Atlanta, GA, United States, [5]University of Plymouth, Plymouth, United Kingdom, [6]World Health Organization/TDR, Geneva, Switzerland

A cluster-randomized trial of insecticide-treated curtains and water jar covers against *Aedes aegypti* mosquitoes was done in Trujillo, NW Venezuela. Entomological indices declined from baseline in both intervention and control clusters. Control houses positive for Aedes at baseline were more likely to be negative one month later if they were within 50m of an intervention house. The results were interpreted as the intervention's effect 'spilling over' from intervention to control clusters. Here we extend the analysis to a) use the number of positive containers per house as the outcome (Breteau Index, BI), b) include the effect of all intervention houses, rather than only the nearest one, c) directly estimate the scale of spill-over effect, and d) estimate the difference between intervention and control arms. The study area extended 1.4km north-south and 0.8km east-west. No buffer areas were used, and most of the 18 clusters were separated by less than 10m from their nearest neighbouring cluster. The median cluster extent was 119m north-south, and 167m east-west. In the baseline survey of 1091 houses, the between-house spatial correlation of BI was estimated to reduce by half over a distance of 110m: similar to the cluster dimensions. At the final follow-up, 9 months after baseline, 730 houses were surveyed. Preliminary results from the spatial model estimate an 18% reduction in BI in intervention versus control arm (95% CI 58% reduction to 14% increase). The intervention effect was estimated to reduce by half over a distance of 8.7m (95% CI 0.5-20m). This implies that the decrease in BI in an intervention house is the same as, for example, the decrease in BI in a control house within 8.7m of 2 intervention houses, or within 17.4 meters of 4 intervention houses. Mass effects of this magnitude would be important in areas of high housing density, especially in the likely circumstance that not all houses in an intervention area deploy the intervention correctly or at all.