Dear Aspergillus fumigatus microarray users-

A number of the members of your community have contacted me to say that that the data being generated using the PFGRC A. fumigatus microarrays are sub-standard and that has raised questions about the quality of the slides. The PFGRC and the NIAID are committed to providing you with the best reagents possible and if problems with these reagents exist, we certainly want to know about it. While we are confident in our qc process, it is always possible that troubles with our printing can go unrecognized. However, in the 6 years I have been working with this technology, slide problems have been the cause of bad hybridizations on only a few occasions. In each case we have recognized this as the confounding issue and brought it to the attention of the slide manufacturer before they were aware of the problem. To help you evaluate the arrays I explain our qc process below and present qc data in the attached file.

Our oligos come from Illumina Inc, a long and trusted partner. Once received the oligos are mixed 1:1 (v/v) with commercial printing buffer (Corning). This material is spotted onto glass slides (amino-silane) from Corning (UltraGAPS) and subsequently cross-linked using a UV cross-linker (Stratagene). The slides are stored desiccated until shipped to you, the end user.

Our qc process involves evaluating a few slides from each print run (lot). We use a product called SpotQC which contains dye labeled 9-mers that hybridize non-specifically to the DNAs spotted on the array. We interpret the signals obtained in these hybridizations to be directly proportional to the amount of bound 70-mer on the slide surface.

The attached PowerPoint presentation shows an examination of 8 different organism-specific microarrays including A. fumigatus. We have published experiments using four of these organism-specific microarrays (B. anthracis, S. pneumoniae, Y. pestis and S. mutans) (Peterson et al., 2004; Identification of competence pheromone responsive genes in Streptococcus pneumoniae by use of DNA microarrays. Mol. Microbiol 51,1051-70). As I believe you will see the arrays performed very well experimentally and the A. fumigatus arrays appear to be at least as good in our qc evaluation.

Based on our current information the problems that some of you have reported do not appear to be caused by slide printing. In our preliminary experiments using genomic DNA we have found that probe sample treatment can make a huge difference in hybridization background. We are continuing these experiments and will post our findings on the FGSC site once they are verified. We are continuing to work with your community representatives to try to figure out where the problem might be and to develop controls that will help users better track experimental problems.

Best of luck,

Scott

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