To: Elizabeth Holmes[eholmes@theranos.com]
From: Kingshuk Das[/O=THERANOS ORGANIZATION/OU=EXCHANGE ADMINISTRATIVE GROUP (FYDIBOHF23SPDLT)/CN=RECIPIENTS/CN=KINGSHUK DASE2B]
Sent: Thur 3/3/2016 6:42:31 PM (UTC)
Subject: 16S rRNA sequencing

Just wanted to f/u on this item you brought up at the scientific meeting last Friday--didn't want to mention in front of the group, but have been working a bit on 16S rRNA sequencing here at UCLA, with a resident of mine. Using the Oxford Nanopore MinION instrument--very cheap, easy, and gets the job done. Anything in this realm will work--i.e., massively parallel sequencing allows agnostic 16S rRNA sequencing, so you're not limited to panels of bugs (or viruses, which are also excellent targets of this tech), and read depth stats w/o amplification allow at least relative quantitation (and can do absolute quant with spike-in controls at extraction, if you like, analogous to most mass spec protocols).

Now that I've seen "under the hood" of your instrument, something along these lines would be perfect as far as space, plus only needs a laptop processor for analysis.

The other (molecular) technique that seemed to be a good fit, especially since you already have flow cytometer capabilities built-in = ddPCR (droplet digital PCR): allows much lower limit of detection, better accuracy, no need for real-time PCR, etc., and can have very small footprint. You already have the thermocycler, so just a matter of matching flow cell/detector. Would be great for any quantitative molecular assay at the small panel or smaller level.

An even cooler option to ddPCR (for similarly limited # of targets), though still in development, but would fit your footprint nicely = electrodetection molecular assays. There are several flavors, but the one that fits best, in my opinion, is one that assays sample directly (no extraction, no amplification) and works from very small volumes (10uL, based on redox read-out of hybridization, nucleic acid is released by method similar to electroporation). Some data suggests it's actually even more sensitive, or at least comparable to ddPCR, but skips almost every step. If interested, can connect you, since the founder is a friend of mine (I have no financial or other interest in his company, don't worry).

Anyway, it was great to see the inner workings of the instrument--gave me a much better sense of molecular possibilities.

If you're not already working on these, and have time, maybe worth discussing a bit once I'm up there full-time (soon!)?

-k