

On-site sequencing speeds up and re-directs field-based microbiology

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As a field-going molecular microbial ecologist, I often experience an extreme high feeling or bout of excitement during immersion in my study environment. However, when I return to my laboratory, generally in the cold and dark winter months, I ‘crash’ or reach a low point, due to my realization of how many samples we obtained in the field and the extent of processing, sequencing and analysis time that we still need to devote to the samples before we can draw any conclusions and share our findings.

In an effort to lessen my now regular post-field crashes, I have started following the development of mobile sequencing technology. Oxford Nanopore Technologies has recently made large strides in this area, producing the MinION a hand-held flow-cell costing \$1,000 that can connect to a laptop computer. The company is developing the even more mobile SmidgION, a lower power device that will run on a smartphone. Depending on the type of MinION sequencing conducted (amplicon, metagenomic, transcriptomic, etc.), preparation after extraction of nucleic acids can take as little as 10-minutes to a few hours, and sequencing reads are immediately available for viewing during the sequencing run. While the platform has been primarily used for DNA molecules, the MinION can directly sequence RNA (without conversion to cDNA), opening up new avenues for exploring microbial transcripts as well as RNA viruses while on-site. Within environmental microbiology, this technology has recently been applied at field sites, including soils and remote glacier environments (Cummings *et al.*, 2017; Edwards *et al.*, 2018) and has also been used to track wastewater contamination from urban storm water (Hu *et al.*, 2018).

The ability to produce sequencing data while in the field could transform the timeline of many environmental microbiology-based studies. Personally, I fantasize about the ability to produce and analyse data and even write

drafts of papers while I am at sea or in the field (Fig. 1). Being on-site and away from other home and work distractions offers a unique opportunity to focus on the project, and immediately correspond with fellow collaborators about the science findings. This innovation would likely lead to more rapidly produced and shared datasets. Speeding up the sharing of our environmental microbiology data and findings carries many advantages, and especially to time-sensitive questions, such as diagnosing diseases, and following the impacts of anthropogenic and climate impacts to our environment. This ability to produce sequence data while in the field could additionally curb the nail-biting stress of transporting microbiological samples internationally, although collection and research permits would still be necessary, and there are always added benefits to archiving samples.

Near real-time sequencing information could transform and re-direct field-sampling strategies. Generally, microbiologists begin fieldwork with a well-organized plan of study sites and samples to obtain. In some cases, field teams can be flexible, such as following oceanographic features using sensors or satellite-based data. But, what if we could use sequencing data to follow microbial patterns? By understanding the distribution of cells, specific functional genes or transcripts in near-real time, we could re-design our sampling strategies to account for these otherwise unseen patterns or features in the environment. On-site sequencing could also facilitate complete re-directions in sampling plans. For example, precious time in the field could be applied to sample in a diagnostic-type fashion, to understand the microorganisms associated with massive animal die-offs or disease-type conditions, and local experts could be consulted in near real-time to alter sampling plans, rather than waiting several weeks or even months after samples are taken (Fig. 1). For scientists focused on the microbiomes of host organisms, this rapid sequencing innovation may be helpful to confirm the taxonomy of host species, and to ensure that the appropriate samples are obtained in the field.

On-site sequencing will also impact the expertise needed for field going research teams. A larger team, equipped with sequencing and bioinformatics technical skills could be indispensable on-site. This requirement opens a door for these personnel to better experience

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Fig. 1. A comic depiction of how rapid, on-site sequencing can be used in a diagnostic-type fashion to produce new findings within environmental microbiology. Timeframe may be slightly exaggerated. Illustration by WHOI Creative Natalie Renier.

their study environment. Additionally, these added personnel would benefit from living together with other scientists in the field, which generally involves strengthened comradery amongst the team and mentoring and exchange of career advice in an informal setting. And as field-based work frequently serves as a recruitment aid for future scientists (especially scenarios offering SCUBA diving on tropical coral reefs), the need for field-going sequencing and bioinformatics technical expertise may help support the recruitment of more technically minded students into environmental microbiology. Even if the

bioinformatics support personnel do not travel to the field, with the proper communications they could more readily participate in the field campaign by analysing data and sharing findings nearly immediately after production.

While sequencers have previously been taken into the field (i.e., Lim *et al.*, 2014), vessels were not optimized for hosting this application. At least two oceanic research vessels have already committed space and data processing support for sequencing efforts at sea, including the Schmidt Ocean Institute's R/V *Falkor* and OceanX's *Alucia*² vessel, which is under construction. These

sponsoring organizations focus on technological developments and discovery-based science and exploration, and can support a fairly large scientific team, making them ideal programs to trial sequencing-driven sampling strategies.

The lessened cost and accessibility of sequencing technology has already had a large impact on the field of environmental microbiology. In my crystal ball, on-site, rapid sequencing has the potential to innovate field-based microbiological research and hopefully lessen my post-cruise crash.

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