Irish Coronavirus Sequencing Consortium:

What we're doing and Why?

Zoom Webinar, 7th October 2020

Panellists: Prof. Paul Cotter, Dr. Fiona Crispie, Dr. John Kenny (Teagasc), Dr. Patrick Stapleton (HSE), Dr. Fidelma Fitzpatrick (RCSI) and Dr. Kate Reddington (NUI Galway).

View the full webinar at: https://youtu.be/y6DZ8vQk2xY

The following questions were not answered live due to time constraints and were instead answered by panellists following the webinar.

While the initial study of few thousand samples was very informative, are there plans to expand and continue WGS sequencing particularly as rate of infection continues to increase? And alignment from government to support?

The original funding and ethics approval for the consortium was predicated on sequencing samples from prior to June 25th. However, some of the newer samples are being sequenced through the National Virus Reference Laboratory. The NVRL is part of the consortium, but the ethics for accessing the new samples are different to that being used by the overall consortium described during the presentation. The NVRL are using the network, protocols and pipelines set up within the consortium for their workflows.

Regarding government support, we hope to demonstrate soon that sequencing SARS-CoV-2 at scale is possible in Ireland using a distributed consortium model and appropriate pipelines. Prospective real-time sequencing could be particularly useful once the current surge of infection is controlled and cases are at low numbers, as it would aid public health authorities to uncover hidden transmission networks, second or third generation transmission events following importation from abroad, and link suspected "community transmission" cases with known outbreaks. This could help prevent a third wave of infection.

In the recent escalation of cases - are there variable strains or is there a predominance of one/few strains i.e. is there increased virulence enabling increased transmission?

The simple answer is that we don't have enough data to fully know this. However, the level of sequencing to date would indicate that versions of the virus carrying the G614 mutation predominate, with other various mutational profiles alongside it. Studies on versions of the virus carrying this mutation would indicate it replicates more, and is therefore potentially more infective, but does not lead to an increased severity of infection when compared to D614 variants (https://www.cell.com/cell/pdf/S0092-8674(20)30820-5.pdf).

What timeline are samples being collecting over? Are the coronavirus cases that are in circulation at present going to be sequenced??? It would be interesting to compare genomes from the current 'spike' with those from the original spike?

The original consortium samples were predominantly collected prior to June 25th due to the basis of the consent and ethical approval from the Human Research Consent Declaration Committee and the National Research Ethics Committee, respectively. As mentioned in answers above, work to investigate the current circulating lineages is on-going at a limited scale in Ireland, and globally is being pursued at a national scale (Australia plans to sequence all SARS-CoV-2 genomes).

The tree that was shown in the presentation was built based on the entire genome, the core genome or specific genes previously chosen? How close in terms of entire genomes are the strains of SARS-Cov2 involved in the study so far?

The trees are built on the basis of the entire genomes. There is a variation in the quality of assemblies uploaded internationally, making a final estimate of variation somewhat challenging, and analysis which groups the variants allows for the variation in sequence quality. The nature of viral genomes and evolution means that many, many variants will arise, but only a few of these are likely to be selected for and become commonly occurring. This selection for a particular variant would be expected to happen if it made the virus more able to spread (e.g. the as we appear to be seeing for the G614 variant).

Is it important that these sequencing efforts continue now over the winter months? To monitor change?

As mentioned above, this work is continuing but it is important to note that when there is widespread prevalence of the infection (in terms of number of people infected) a more practical approach in terms of public health benefit is a well-resourced tracing system to identify the contacts of Covid positive patients. Sequencing in the winter months would allow for better understanding of the background strain diversity in Ireland. This could be particularly important in spring with lower case numbers, as public health follow up of small outbreaks can be assisted by sequencing data, and such sequencing data is more powerful if it can be interpreted in the context of well sampled national and international strain diversity.

Any evidence from the sequencing data that the spike proteins are mutated in a way that might affect vaccine development?

The good news is that analysis indicates that a SARS-CoV-2 vaccine candidate would likely match all currently circulating variants. A publication on this can be found here: https://www.pnas.org/content/117/38/23652

Do all strains bind easily to ACE2 receptors or have differences been seen with regard to this?

Yes, in the case of the G614 variant we can observe differences in cell binding efficiencies.

Any data on how similar SARS-2 is to other known coronaviruses, in particular, those that are endemic in the humans?

Yes, studies have looked at the similarity to existing coronaviruses, including in the likely predecessors in bats. SARS-CoV-2 itself is not a recombinant of any sarbecoviruses detected to date, and its receptor-binding motif, important for specificity to human ACE2 receptors, appears to be an ancestral trait shared with bat viruses and not one acquired recently via recombination (https://www.nature.com/articles/s41564-020-0771-4).

When testing we noticed that if the samples were vortexed it destroyed the RNA. How is the virus so infective if it can be destroyed so easily?

That depends on the type of sample you're referring to. In molecular biology a number of enzymes cannot be exposed to vortexing as the violence dissociates the enzymes, inactivating them. The level of shearing stress encountered during vortexing is not the kind of thing a virus would be exposed to in nature.

Will the sequence data be linked to clinical data for those cases that are in hospital?

No, in the case of our project we do not have the approval to link this information. This type of linkage is being done in other studies internationally.

Is there any plan to look at the variations you are picking up to see whether they are affecting the behaviour/virulence of the virus?

Not in this work. The viruses have not been extracted from these patient samples, only the RNA of the genomes. Therefore we cannot take the samples further for studies of the viruses. However, work has been performed to look at the effect of majorly abundant mutations on infection outcomes (e.g. the G164 lineage described here: https://www.cell.com/cell/pdf/S0092-8674(20)30820-5.pdf).

What about gender? Hormones are known to influence the immune response….

Gender may be a factor in patient outcomes, but to date no obvious trait is observed in our data linking virus lineage to patient gender.

Are you collecting clinical data so you can link genome variation with clinical outcome?

No, our remit for this work is circumscribed by the terms of the research protocol agreed with the National Research Ethics Board for Covid-19 and we will not be combining this work with clinical outcomes.

For more information on the Irish Coronavirus Sequencing Consortium, visit: https://teagasc.ie/food/research-and-innovation/research-areas/food-bioscience/irish-coronavirus-sequencing-consortium/