Clarifying the evidence on SARS-CoV-2 antigen rapid tests in public health responses to COVID-19

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The use of rapid lateral flow antigen testing (LFT) for SARS-CoV-2 has been questioned[1–3] with uncorroborated[4] reports of poor LFT sensitivity. The debate surrounding the use of the Innova Lateral Flow SARS-CoV-2 Antigen Test in the UK risks confusing policy makers internationally and potentially stalling deployment of LFTs in other countries.[5] As scientists and health professionals evaluating some of the world's largest pilots of LFT, we wish to challenge those interpretations and clarify the evidence on how such testing might be used to detect SARS-CoV-2 in minutes and improve COVID-19 control measures.

Testing for SARS-CoV-2 is central to COVID-19 management and has relied on quantitative reverse transcriptase polymerase chain reaction (PCR) technology. PCR seeks the genetic code of the virus from nose or throat swabs and amplifies it over 30–40 cycles, doubling each cycle, enabling even miniscule, potentially single, copies to be detected. PCR is thus a powerful clinical test, specifically when a patient is, or was recently, infected with SARS-CoV-2. Fragments of RNA can linger for weeks after infectious virus has been cleared,[6] often in people without symptoms or known exposures.[7]

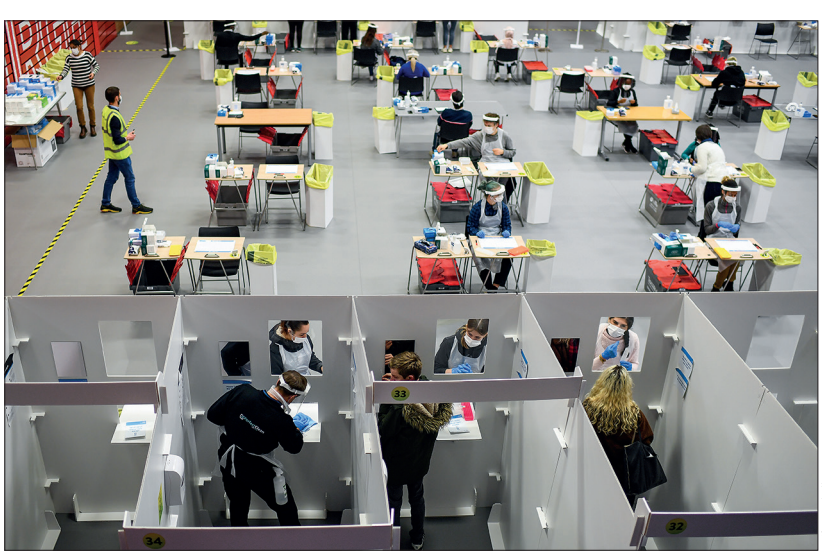
However, for public health measures, another approach is needed. Testing to help slow the spread of SARS-CoV-2 asks not whether someone has RNA in their nose from earlier infection, but whether they are infectious today. It is a net loss to the health, social, and economic well-being of communities if post-infectious individuals test positive and isolate for 10 days. In our view, current PCR testing is therefore not the appropriate gold standard for evaluating a SARS-CoV-2 public health test.

Most people infected with SARS-CoV-2 are contagious for 4–8 days.[7] Specimens are generally not found to contain culture-positive (potentially contagious) virus beyond day 9 after the onset of symptoms, with most transmission occurring before day 5.[7,8] This timing fits with the observed patterns of virus transmission (usually 2 days before to 5 days after symptom onset), which led public health agencies to recommend a 10-day isolation period.[9] The short window of transmissibility contrasts with a median 22–33 days of PCR positivity (longer with severe infections and somewhat shorter among asymptomatic individuals).[10] This suggests that 50–75% of the time an individual is PCR positive, they are likely to be post-infectious.[11,12]

Once SARS-CoV-2 replication has been controlled by the immune system, RNA levels detectable by PCR on respiratory secretions fall to very low levels when individuals are much less likely to infect others.[13–15] The remaining RNA copies can take weeks, or occasionally months,[16,17] to clear, during which time PCR remains positive.[7]

A public health test for detecting someone who might be contagious is, by logical deduction, expected to have a sensitivity of around 30–40% versus PCR when testing a random sample of asymptomatic people in a steady-state outbreak.[18] Furthermore, the asymmetry of RNA reflecting more infectiousness nearer to the time of exposure, means that the sensitivity of the ideal test of infectiousness when measured against PCR may vary across the epidemic curve, from as high as 50–60% when an outbreak is surging to 20–30% or less as infections decline.[19]

LFT and the UK testing programme have been criticised[1–3,5] for poor sensitivity in people without symptoms. In our view, these criticisms misinterpreted data from the interim report on the pilot of community testing in Liverpool, UK.[20,21] When paired LFT and PCR testing was done in Liverpool, the epidemic curve was declining.[20] At this point, a priori one should expect a



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public health test that is highly sensitive for detecting infectious virus to show low overall sensitivity relative to PCR in people without symptoms or known exposures.

Further confusion reigns over PCR cycle threshold (Ct) values, viral loads, and infectiousness. In the Liverpool pilot, Innova LFT picked up 19 of 24 (79%) samples with Ct below 20 and ten of 11 (91%) samples with Ct below 18.[20] The 66% sensitivity in the Liverpool interim report[20] was based cautiously on Ct below or equal to 25 indicating viable virus. For the laboratory processing of the Liverpool samples, Ct values of 21–18 most likely reflect the 100 000 to 1 million RNA copies per mL, quantities below which virus cultures usually become negative and transmission risks are low.[22-24] Other laboratories place this threshold at a Ct of 30.[24] There is no international standardisation between laboratories and assays, leaving Ct calibration with viral load poorly reported and easy to misunderstand.

Early findings, widely reported,[3] from a study by Ferguson and colleagues,[25] suggested that LFT had only 3% sensitivity for detecting SARS-CoV-2 among PCR-positive students at Birmingham University. Test underperformance was implied to explain finding only two positive results among 7189 individuals tested with Innova LFT.[25] In that study,[25] in a random sample of 710 (10%) LFT-negative individuals there were six PCR-positive results. That finding was extrapolated to 60 cases in the whole cohort, giving an extrapolated sensitivity of two of 62 (3·2%). The Ct values from the six PCR-positive samples were projected to Ct values for the 60 cases (54 unobserved plus six observed); in all six observed cases, viral loads were very low (Ct ≥29 reflecting around <1000 RNA copies per mL in the laboratory used)—when LFT should be negative. By comparison, the Liverpool pilot saw virus levels 1000 to 1 million times higher.[20] In our view, the Birmingham study showed that PCR-positive asymptomatic students at a time of falling COVID-19 incidence had low viral loads compared with symptomatic members of the public attending testing centres and that LFT was working as expected.[26]

We wholeheartedly support healthy scientific debate to inform policies promptly. The COVID-19 road ahead looks challenging; therefore, we need big, bold actions across science and society, such as the Liverpool community testing pilot. The prompt evidence from such pilots can inform policies and help maintain public confidence in the public health responses needed to navigate this pandemic's onward path.

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