



Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Elsevier hereby grants permission to make all its COVID-19-related research that is available on the COVID-19 resource centre - including this research content - immediately available in PubMed Central and other publicly funded repositories, such as the WHO COVID database with rights for unrestricted research re-use and analyses in any form or by any means with acknowledgement of the original source. These permissions are granted for free by Elsevier for as long as the COVID-19 resource centre remains active.



Contents lists available at ScienceDirect

Journal of Infection

journal homepage: www.elsevier.com/locate/jinf

Letter to the Editor

Implementation study of SARS-CoV-2 antigen lateral flow tests in men's professional (Premiership) rugby union sports squads in England during the COVID-19 pandemic


Dear Editor,

Since the 16th March 2021, following the initial suspension all Rugby Union and Premiership Rugby in response to the COVID-19 pandemic, UK elite sports have been led by guidance issued by the Department for Digital, Culture, Media and Sport, for a phased return to competition^{1,2}. The minimum operating standards set out by English men's Professional Rugby stipulated that each team or training group must ensure all relevant persons have a COVID-19 RT-qPCR test weekly and as close to a game as operationally possible (<72 h)^{2,3}. With reports of turnaround times of up to 72 h⁴ impeding the correct triage and isolation measures, it also became apparent, that the frequency of testing did not allow for the optimum risk management of both the training environment and the match environment. There was an urgent need to review the testing programme in Professional Rugby to agree the most cost-effective and accurate testing modality. We read with interest the real-world evaluation of antigen-based rapid diagnostic tests (Ag-RDTs) for COVID-19 and the high uptake for mass testing amongst healthcare workers⁵. To provide oversight, advice, and guidance on the scope of testing for SARS-CoV-2 by Ag-RDT, Premiership Rugby and Rugby Football Union approved this rapid collaborative service evaluation.

In this study we evaluated the NowCheck COVID-19 Ag Test, Bionote Inc and the Mologic COVID-19 Ag Test, Mologic Ltd, referred to as Bionote and Mologic, respectively. Over a 6-week study period three men's professional Premiership Rugby Union sports squads in England, performed side by side SARS-CoV-2 testing by RT-qPCR and Ag-RDT. Data from matched RT-qPCR-to-Ag-RDT collected swabs was available for 2097 samples.

With written consent, 265 player and support staff of three men's professional (premiership) rugby union sports squads were recruited through the "Evaluation of antigen RDT and PCR for diagnostic screening of SARS-CoV-2 Incidence in Sports Squads" study approved by the Liverpool School of Tropical Medicine Research Ethics Committee (Study reference: 20-099). Twice weekly combined throat and nose swab samples were collected by healthcare professionals and sent for SARS-CoV-2 RT-qPCR testing by Randox Laboratories, Ireland. Twice weekly antigen testing with Ag-RDT was performed using the Bionote, during the first four-weeks of the study, on nasopharyngeal swabs. Followed by the Mologic, for the final two-weeks, using combined throat and nasal swabs. Swabs for the Ag-RDTs were collected and performed immediately either by the HCP or self-sampled by participants according to manufacturer's instruction. Results were captured after 15 min by Chief Medical Officers for each club. Sampling for RT-qPCR tests

and Ag-RDT occurred on the same day; Mondays and 24–48 h before fixtures each week. On testing days where self-sampling was required, this was performed under the instruction of observing medical staff. Participants with positive SARS-CoV-2 Ag-RDT results were immediately excluded from squad activities and instructed to isolate by the Team Doctor at each club in line with PHE guidance and the outcome of the PCR result used as confirmatory diagnostic testing.

Eight positive cases were detected by SARS-CoV-2 RT-qPCR during the first 14 days of the Bionote evaluation, with three of the nine samples provided detected by Ag-RDT (Table 1). The six undetected samples had RT-qPCR cycle threshold (Ct) values in the range of 19.81–34.23. One HCP swab ($n = 2$), collected for Case 5, was negative for Bionote Ag-RDT in the first round (R1) of testing. This case was detected three days later by Ag-RDT with a self-swab in the second round (R2) of testing that week (Ct value of 29 for target 1). Five self-sampled swabs ($n = 7$) were undetected by Ag-RDT, with Ct values ranging from 19.81 to 34.23. No positive cases were detected by RT-qPCR or Ag-RDT during the Mologic evaluation as prevalence dropped due to national lockdown measures. The overall sensitivity of the Bionote Ag-RDT was found to be 33.33% [95% CI 7.49–70.07%] (Table 2). For HCP-sampled and self-sampled swabs, the sensitivity was 50.00% [95% CI 1.26–98.74%] and 28.57% [95% CI 3.67–70.96%], respectively (Table 2). The specificity for both the Bionote and Mologic Ag-RDTs was 100% (Table 2).

Responses from an "End of evaluation" questionnaire were received from a total of 52 participants. All participants responded that they would be happy to continue Ag-RDT testing, with 94% agreeing that they would be confident to run the test at home before attending training. Many participants agreed or were impartial to the comfort, ease of use and interpretation of the Bionote and Mologic Ag-RDTs. Whilst many medical staff administering the Ag-RDT testing program found the Ag-RDT swab easier or about the same as the RT-qPCR swab to administer, with 7.3% finding it harder to administer. The Ag-RDT was also found to be less time consuming for 73.5% of administrators compared to the RT-qPCR testing program.

Whilst overall events were too low to comment on the accuracy of Ag-RDTs, the sensitivity was much lower with a wider CI than expected. Additional clinical evaluation studies of symptomatic participants had a sensitivity and specificity for the Bionote Ag-RDT ranging from 84.5% to 89.2% and 94.4%, to 97.3%, respectively^{6,7}. Despite lower sensitivity to SARS-CoV-2 RT-qPCR, clinical evaluations have demonstrated that most individuals with a high-viral load (Ct values ≤ 25.0 or $> 10^6$ genomic virus copies/ml) have been accurately detected by Ag-RDTs^{8–10}. Results support this trend with five of the six SARS-CoV-2 confirmed cases undetected by Ag-RDT having Ct values > 25.0 . In this study, HCP collected Ag-RDT swabs ($n = 2$) detected one case, Case 8 (Ct values of 25.02

Table 1
SARS-CoV-2 RT-qPCR confirmed cases detected in three men's professional (premiership) rugby union sports squads during the Bionote Ag-RDT evaluation.

Squad	Case ^a	Date Sampled(Testing round) ^b	RT-qPCR Target 1 (Ct)	RT-qPCR Target 2(Ct)	Ag-RDT(Sampler)
Club 1	1	11/01/2021 (R1)	13.92	15.64	Positive (self-swab)
	2	11/01/2021 (R1)	19.81	21.18	Negative (self-swab)
	3	11/01/2021 (R1)	32.94	34.23ss	Negative (self-swab)
	4	18/01/2021 (R1)	25.83	26.54	Negative (self-swab)
Club 2	5 ^d	11/01/2021 (R1)	32	33	Negative (professional)
		21/01/2021 (R2)	29.33	-	Positive (self-swab)
	6	21/01/2021 (R2)	29.11	31.49	Negative (self-swab)
Club 3 ^c	7 ^e	11/01/2021 (R1)	32.07	27.73	Negative (self-swab)
	8	11/01/2021 (R1)	25.02	25.81	Positive (professional)

^a All cases were detected during the Bionote evaluation period.

^b R1: Round 1 of testing week, R2: Round 2 of testing week.

^c Self-sampling for all participants during first 2-weeks of evaluation.

^d Previous PCR result, excluded from PCR for 90 days, second Ag-RDT sampling (R2) upon return after 10 days of isolation.

^e Only self-swab sampling of this squad.

Table 2
Sensitivity and specificity of the Bionote and Mologic Ag-RDTs in total samples and sampler sub-groups three men's professional (premiership) rugby union sports squads.

Sample Group (%)	Bionote		Mologic		95% CI	95% CI	95% CI	95% CI
	RT-qPCR positive (n)	95% CI	Sensitivity	Specificity				
All Samples	9	33.33	7.49 – 70.07	100	0	N/A	100	
HCP-swab	2	50.00	1.26–98.74	100	-	N/A	100	
Self-swab	7	28.57	3.67 – 70.96	100	-			

CI, confidence interval; Ct, cycle threshold of RT-qPCR.

and 25.81 for target 1 and target 2, respectively) within this cut-off (Ct < 25) and missed one case, Case 5 (R1 Ct values of 32 and 33 for target 1 and target 2, respectively) outside of this described cut-off. Whilst five self-sampled swabs ($n = 6$) were undetected by Bionote Ag-RDT, all but one case had Ct values >25.0. The exception being Cases 2 (Ct values of 19.81 and 21.18 for target 1 and target 2, respectively).

Despite the high acceptability of Ag-RDTs in this the study the performance needs ongoing real-world evaluation to support rapid transition to self-swabbing. A portfolio of new testing regimes for Elite English Professional and European competitions has since been introduced after this evaluation, based on prevalence and cross-border travel requirements, and following consultation with organising bodies and Public Health England. This has included three or twice weekly Ag-RDT testing, daily Ag-RDT testing and a combined Ag-RDT and RT-qPCR weekly testing programme. The cost of testing for SARS-CoV-2 for professional squads is already challenging, c. £60-£80 per test + transportation costs. While low cost RDTs become more widely available, their ability to mitigate the transmission of SARS-CoV-2 is not independent, further evaluation is required to identify the most cost-effective option. The optimal testing strategies for professional (men's) rugby squads may not be feasible for all amateur sports. Clear best practice guidance and standards concerning social distancing, hand washing, masks and isolation remain paramount to curtail the spread of SARS-CoV-2. The uptake in vaccination of adults across the UK offers much needed optimism, however, with at the time of writing 41% of the UK fully vaccinated with boosters and the increasing circulation of variants of concern, testing remains an important pillar for the return to sport as we once knew it.

Although this service evaluation was able to determine the feasibility and acceptability of Ag-RDT in a professional contact team sports environment COVID-19 events were too few to measure the accuracy of either Ag-RDT. Having the ability to test frequently, easily, and rapidly during a training and match week should reduce risk of transmission and outbreaks within and between squads. Ag-RDT options will be able to reduce the high cost associated with current screening programmes and will likely make asymptomatic COVID-19 surveillance testing possible and sustainable for a number of sports. Continued larger implementation studies are needed to identify sensitive and specific Ag-RDTs and agree the optimal implementation in specific elite sport settings. This study is an example of service evaluation towards the phased return to sports in the UK.

Authors' contributions

The study was conceived by ERA and TF. The study design was developed by ERA, LF, MR, MC and SK and TF. Data extraction was conducted by CL KJ, LF, LJ, ML, TW. Data analysis and interpretation were conducted by LF. The initial manuscript was prepared by LF. All authors edited and approved the final manuscript.

Conflicts of Interest

ER is Director of Epidemic and Neglected Diseases at Mologic. All remaining authors have no conflicts of interest to declare.

Sources of funding

This work was supported by Philanthropic donations. The funders had no input into the design, interpretation or reporting of the study.

Acknowledgements

The authors wish to acknowledge the contributions of FIND and Mologic Ltd for the supply of Ag-RDT kits, Dawn Veal and Randox Laboratories healthcare professionals for supporting recruitment and sample processing.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at [doi:10.1016/j.jinf.2021.12.037](https://doi.org/10.1016/j.jinf.2021.12.037).

References

1. Elliott N, et al. Infographic. Graduated return to play guidance following COVID-19 infection. *Br J Sports Med* 2020;**54**(19):1174–5.
2. Kemp S, et al. Sports medicine leaders working with government and public health to plan a 'return-to-sport' during the COVID-19 pandemic: the UK's collaborative five-stage model for elite sport. *British J Sports Med* 2021;**55**(1):4–5.
3. Rankin A, et al. Infographic. COVID-19 RT-PCR testing for elite athletes. *Br J Sports Med* 2021:103751 p. bjsports-2020.
4. Kretzschmar ME, et al. Impact of delays on effectiveness of contact tracing strategies for COVID-19: a modelling study. *Lancet Public Health* 2020;**5**(8):e452–9.
5. Lamb G, et al. Real-world evaluation of COVID-19 lateral flow device (LFD) mass-testing in healthcare workers at a London hospital; a prospective cohort analysis. *J. Infect.* 2021;**83**(4):452–7.
6. FIND. *FIND Evaluation of Bionote, Inc. NowCheck COVID-19 Ag Test External Report Version 1.3*, 5. November 2020, 2020 16/04/2021; Available from: https://www.finddx.org/wp-content/uploads/2020/11/Bionote_Ag-INTERIM-Public-Report_202011105-v1-3.pdf.
7. Onsongo, S.N., et al., *Field performance of NowCheck rapid antigen test for SARS-CoV-2 in Kisumu County, western Kenya*. medRxiv, 2021: p. 2021.08.12.21261462.
8. Singanayagam A, et al.. *Duration of infectiousness and correlation with RT-PCR cycle threshold values in cases of COVID-19, England, January to May 2020*, 25. *Eurosurveillance*; 2020.
9. Pekosz A, et al. Antigen-based testing but not real-time PCR correlates with SARS-CoV-2 virus culture. *medRxiv* 2020 2020.10.02.20205708.
10. Cubas-Atienzar AI, et al. Limit of detection in different matrices of nineteen commercially available rapid antigen tests for the detection of SARS-CoV-2. *medRxiv* 2021 2021.03.19.21253950.

Lorna S Finch
Centre for Drugs and Diagnostics, Liverpool School of Tropical
Medicine, Liverpool, UK

Adrian Harris
Exeter Chiefs RUFC, Exeter, UK

Catherine Lester
Northampton Rugby Football Club Ltd, Northampton, UK
Centre for Sports and Exercise Medicine, Queen Mary University,
London, UK

Dawn Veal
Exeter Chiefs RUFC, Exeter, UK

Karen Jones
Gloucester Rugby Ltd, Gloucester, UK

Jamie Fulton, Lewis Jones
Exeter Chiefs RUFC, Exeter, UK

Matt Lee
Northampton Rugby Football Club Ltd, Northampton, UK

Tony Walker
Exeter Chiefs RUFC, Exeter, UK

Mike Rossiter
Premiership Rugby, Twickenham, Middlesex, UK
Hampshire Hospitals NHS Foundation Trust, Basingstoke, UK

Matt Cross
Premiership Rugby, Twickenham, Middlesex, UK

Simon Kemp
Rugby Football Union, Twickenham, UK
London School of Hygiene and Tropical Medicine, London, UK

Tom Fletcher
Centre for Drugs and Diagnostics, Liverpool School of Tropical
Medicine, Liverpool, UK
Liverpool University Hospitals NHS Foundation Trust, Liverpool, UK

Emily R Adams*
Centre for Drugs and Diagnostics, Liverpool School of Tropical
Medicine, Liverpool, UK
Mologic, Thurleigh, UK

*Corresponding author.

E-mail address: Emily.Adams@lstmed.ac.uk (E.R. Adams)