Screening antibodies raised against the spike glycoprotein of SARS-CoV-2 to support the development of rapid antigen assays

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Abstract

Severe acute respiratory coronavirus-2 (SARS-CoV-2) is a novel viral pathogen and therefore a challenge accurately diagnose infection. Asymptomatic cases are common and so it is difficult to accurately identify infected cases to support surveillance and case detection. Diagnostic test developers are working to meet the global demand for accurate and rapid diagnostic tests to support disease management. However, the focus of many of these has been on molecular diagnostic tests, and more recently serologic tests, for use in primarily high-income countries. Low- and middle-income countries typically have very limited access to molecular diagnostic testing due to fewer resources. Serologic testing is an inappropriate surrogate as the early stages of infection are not detected and misdiagnosis will promote continued transmission. Detection of infection via direct antigen testing may allow for earlier diagnosis provided such a method is sensitive. Leading SARS-CoV-2 biomarkers include spike protein, nucleocapsid protein, envelope protein, and membrane protein. This research focuses on antibodies to SARS-CoV-2 spike protein due to the number of monoclonal antibodies that have been developed for therapeutic research but also have potential diagnostic value. In this study we assessed the performance of antibodies to the spike glycoprotein, acquired from both commercial and private groups in multiplexed liquid immunoassays, with concurrent testing via a half strip lateral flow assays to indicate antibodies with potential in LFA development. These processes allow for selection of pairs of high affinity anti-spike antibodies are suitable for liquid immunoassays and LFA assays, some of which with sensitivity into the low picogram range with the liquid immunoassay formats with no cross reactivity to other coronavirus S antigens. Discrepancies in optimal ranking was observed with the top pairs used in the liquid and LFA formats. These findings can support the development of SARS-CoV-2 LFAs and diagnostic tools.

Introduction

The appearance of a novel coronavirus disease 2019 (COVID-19) was first reported in the city of Wuhan, Hubei Province, China in 2019[1]. Since then COVID-19 has progressed to pandemic levels with over 23 million reported cases including at least 800,000 associated deaths reported globally[2]. The pathogen responsible is the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), a novel betacoronavirus. The coronaviruses are enveloped positive-stranded RNA viruses that are 70 - 90 nm in size and characterized by a crown like morphology associated with the display of spike (S) glycoproteins on the host membrane-derived and lipid bilayered viral envelope[3;4]. The structure of the S glycoprotein of SARS-CoV-2 has been resolved and is known to be essential for the viral infection of the host cell via its binding to the cellular receptor angiotensin-converting enzyme 2 to promote fusion and entry into the cell[5]. The S glycoprotein is poorly conserved across coronaviruses, with 85.3% of the antibody epitopes found in SARS CoV-2 S protein considered unique[6;7]. Conversely, high conservation is noted across SARS CoV-2 isolates from Europe, Asia, and the US, resulting in an antigen that offers greater specificity over more conserved targets like the N antigen.

The rapid spread of COVID-19 has resulted in an urgent need for effective diagnostic tests to support disease management, monitoring, surveillance and pandemic control against SARS-CoV-2[8]. In high income countries molecular testing, typically using real time reverse transcription PCR (RT PCR), has been the primary test method implemented to diagnose SARS-CoV-2 in both symptomatic and asymptomatic cases but the accurate detection of early infection remains challenging giving false negative results[9;10]. As of August 24th, 141 commercial or clinical laboratory derived molecular tests have been granted emergency use authorization (EUA) in the USA by the Food and Drug Administration (FDA)[11]. The vast majority of the tests are predominantly unsuitable for use at the point of care as many are in open assay format with which significant engagement are needed from skilled operators to prepare the samples for testing, prepare test reagents, operate complex equipment and finally to process the data and interpret the test results. Automated high throughput molecular platforms are available and are capable of processing large numbers of samples with significantly reduced operator input[12-15]. However, acquiring and operating such equipment comes with high capital costs and a need for appropriate

infrastructure, not only for housing the equipment and reagents but also requiring effective specimen collection and transport and the reporting of test data to patients, clinicians, and health care programs after processing. In the current pandemic, global demand for has affected all countries and so sufficient access to reagents, consumables and more other materials such as personnel protective equipment, swabs and transport media is necessary ensure consistent testing[16].

Lack of access to key reagents and consumables has highlighted that there is a market for SARS-CoV-2 diagnostic immunoassay-based lateral flow assays (LFAs) in high income countries. Low- and middle-income countries (LMICs) already faced serious constraints in diagnostic capacity and accessibility before the COVID-19 pandemic stuck. SARS-CoV-2 will have an amplified effect in these countries that have limited access to care with and already greater burden of infectious diseases[17]. LMICs lack time and finances for the swift uptake of new diagnostic technologies. Furthermore, a lack of resources and skilled laboratorians limits the number of test facilities, the ability to scale testing, while access to critical reagents is limited as high-income countries dominate procurement, culminating in inability to perform molecular tests at the scale required[18]. Without access to expanded molecular test capacity and capability, other diagnostic tools must be developed to support COVID-19 infection control. Therefore, LFAs serve as a best alternative in regions lacking sufficient access to widespread molecular testing for SARS-coV-2.

For detection and control of COVID-19 in LMICs, an antigen LFA format makes a more viable option to the serologic LFAs that currently dominate the market due to their ability to detect SARS-CoV-2 directly and earlier in the infection process. Serology-based assays are insensitive in early infection requiring individuals to be diseased for at least a week before the antibody response can first be detected (IgA, IgM, and/or IgG)[19], which is enough time for infected individual to unknowingly spread the disease[20]. In terms of operation and cost, LFAs can be manufactured at a very large scale and at a relatively low cost per unit in comparison to molecular tests. While LFAs typically require some limited training of users, they are easy to use, give a test result in minutes, most do not require associated equipment and their use is broadly disseminated from hospitals to clinics to community-and home-based testing (e.g. malaria, HIV-1, and pregnancy testing).

The performance of antigen detection LFAs is variable depending on the performance of the antibodies used in the test and while visually read LFA reach the level of sensitivity that molecular assays offer, the use of readers can further increase test senstivity. The recent FDA EUA to Lumira Diagnostics (Stirling, UK) for their SARS-CoV-2 Ag assay has claims of a sensitivity of 97.6% as compared to RT PCR testing. Therefore, rapid antigen assays using high performance antibodies can offer may offer sufficient clinical sensitivity to detect infectious patients in decentralized settings where molecular testing is not readily available today. Furthermore, the LFA format can be manufactured at extremely high volumes and very low costs, and can offer increased testing capacity in LMICs where molecular testing is not readily or sufficiently. Other markets where LFAs can play a key role is in disseminated testing models such as employed in community- and home-based testing, and self-testing[21-23].

The WHO's recently released target product profile for a point of care test for suspected COVID-19 cases (e.g. a rapid antigen assay) has listed the acceptable characteristics for sensitivity and specificity at ≥70% and ≥97%, respectively[24]. A current challenge to antigen test development is understanding the performance of the SARS-CoV-2 antibodies that are on or entering the market, with the screening of large numbers of unqualified antibodies a resource sink for developers aiming to develop direct antigen tests. Abundant targets include the four major structural proteins: the spike (S), membrane (M), envelope (E) and the nucleocapsid (N) proteins. The S glycoprotein represented an attractive candidate due to the unique structural changes relative to SARS-COV1 and other seasonal coronaviruses, offering the potential of high specificity for SARS-COV2[6].

We have assessed the performance of anti-N protein antibodies via half paper LFAs in recent studies but with the spike, while a lower prevalence target, the structural role of S may present better epitopes to antibodies and so could be an attractive target for a rapid LFA[25;26]. In this study we accessed multiple antibodies targeting the S

glycoprotein by leveraging the antibody therapeutics industry and also commercially available sources. We assessed their performance for sensitivity using recombinant S antigens and inactivated cultured SARS-CoV-2 virus and their sensitivity to other S glycoproteins from other human coronavirus species. Antibody pairs were assessed in a highly sensitive liquid immunoassay format to indicate sensitivity and specificity to the SARS-CoV-2 S glycoprotein in addition to a high throughput half-strip LFA screen to identify candidates with the greatest performance as observed on nitrocellulose[25;27]. These screens enabled us to down select and identify the optimal pairs that offer the greatest sensitivity and specificity for further development and incorporation into liquid and LFA immunoassay formats for direct antigen detection of SARS-CoV-2 virus via the S glycoprotein.

Materials and Methods

Antibodies and Antigens

Antibodies to the S glycoprotein were procured from Leinco Technologies (Fenton, MO, USA), Sino Biological (Wayne, PA, USA), Cedar Lane (Burlington, NC, USA), and Creative Diagnostics (Shirley, NY, USA). AbCellera Biologics Inc. (Vancouver, BC, Canada) provided a private collection of 41 recombinant antibodies engineered from B cells harvested from a convalescent patient after SARS-CoV-2 infection. A list of all anti-spike antibodies screened in this work are provided in Table 1si (suppl. info).

The full-length trimeric SARS-CoV-2 Spike antigens expressed in mammalian cells were purchased from Acro Biosystems (Newark, DE, USA), and Leinco Technologies or made in-house (Global Health Labs only). S antigens expressed in Baculovirus-insect cells were obtained from Sino Biological and Biodefense and Emerging Infections Research Resources Repository (BEI Resources, Manassas, VA, USA). Heat-inactivated and gamma-irradiated cell culture lysates of SARS-CoV-2, and irradiated cell culture lysates of the middle eastern respiratory syndrome virus (MERS) and SARS-CoV-1 were also acquired from BEI Resources. Titered HEK293 cell culture supernatants of human coronaviruses OC43 and 229E were generously gifted from the laboratory of Dr Scott Meschke, University of Washington (Seattle, WA, USA). SARS-CoV-2 positive and negative nasopharyngeal specimens were acquired from the Washington COVID-19 Biorepository[28]. These samples were discarded clinical specimen from a laboratory that used the Applied Biosystems TaqPath COVID-19 assay (ThermoFisher Scientific, Waltham, MA, USA), a SARS-CoV-2 RT PCR assay with FDA EUA.

Viral load determination via qRT-PCR

Clinical specimens were prepared in one of two ways. 1) RNA was extracted from 50 µL of specimen using the QIAamp Viral RNA Mini Kit (Qiagen, Valencia, USA) according to the manufacturer's instructions 2) 40 µL of specimen were heated to 95°C for 10 mins to lyse virions. Next, 5 µL of extracted RNA or 2.5 µL of heat-treated specimens were added to qRT-PCR reactions containing TaqPath™ 1-Step RT-qPCR Master Mix (ThermoFisher Scientific) and the Centers for Disease Control and Prevention N1 primer set (IDT, Coralville, USA). Reactions were carried out per the CDC protocol with an ABI7300 Fast Real-Time PCR System (Applied Biosystems). A standard curve was generated using quantified genomic RNA from SARS-Related Coronavirus 2, Isolate USA-WA1/2020, NR-52285 (BEI Resources) and used to determine the viral load of each sample.

Liquid Immunoassay Screening for Optimal Antibody Pairs

Labelling of antibodies for use on the Meso Scale Discovery immunoassay platform

Per the protocol, two aliquots of each antibody (100 µg/mL) were labelled with biotin (EZ-Link Sulfo-NHS-LC-Biotinylation Kit, ThermoFisher Scientific) for capture and SULFO-TAG (GOLD SULFO-TAG NHS-Ester) for detection using the Meso Scale Discovery (MSD, Rockville, MD, USA) electrochemiluminescent immunoassay platform. Unbound biotin or SULFO-TAG was removed using Zeba™ spin desalting columns (ThermoFisher

Scientific), and the incorporation ratio for each label was measured. Briefly, the concentration of biotinylated antibodies after desalting was measured at 280 nm via spectrophotometer (Nanodrop ND-1000, ThermoFisher Scientific); biotin incorporation was measured using a Biotin quantification kit (Pierce™, ThermoFisher Scientific). For measuring the incorporation of the SULFO-TAG, the protein concentration was estimated using the bicinchoninic acid (BCA) protein assay (ThermoFisher Scientific), and the SULFO-TAG label spectrophotometrically measured at 455 nm.

Preparation of U-plex plates

The biotinylated capture antibodies were coupled via biotin-streptavidin binding to U-PLEX linkers. To prepare the capture antibody arrays, up to 10 antibody-linker conjugates were pooled together in U-PLEX stop buffer at a concentration of 0.29 µg/mL per antibody, and 50 µL of this mixture was added to individual wells of the U-PLEX plates. The plates were incubated for 1 hour with shaking (500 rpm) to allow the antibody array to self-assemble to the complimentary antibody linker binding sites and unbound material then removed by washing 3 times with 150 µL/well of phosphate buffered saline + 0.05% Tween 20 (PBS-T, pH 7.5) using a BioTek ELX405R microplate washer (BioTek Instruments Inc., Winooski, VT, USA).

Processing U-plex plates

Appropriate serial dilutions of the trimeric S glycoprotein in Diluent 100 (MSD) were prepared. Clinical specimens and cell lysates were prepared by adding 25 µL into 25 µL of Diluent 100. The 50 µL of each prepared sample was added to each antibody array well in the U-PLEX plate, and incubated with shaking for 1 hour at room temperature. Plates were washed 3 times in 1X PBST and then 25 µL of 2 µg/mL SULFO-TAG-labeled detection antibody in Diluent 3 (MSD) was added to each well with incubation for an hour with shaking. Plates were then washed 3 times to remove excess detection reagent and the wells filled with 150 µL of 2X read buffer T (MSD). The plates were inserted into the MESO QuickPlex SQ 120 plate reader (MSD) and the electrochemiluminescence (ECL) from each individual array spot was subsequently measured. In the absence of a control, the array spot that gave the highest signal to noise in each plate was expressed as 100% and each of the array spots in each plate expressed as percentile of this value. When serial dilutions of the S glycoprotein were used to generate a calibration curve, the relationship of ECL signal to S glycoprotein concentration was then fitted to a four-parameter logistic (4-PL) function in the Discovery Workbench v4 program. S glycoprotein concentrations for gamma-irradiated SARS-CoV-2 were calculated by back-fitting ECL signals to the 4-PL fit.

Antibody Evaluation

The identification of the optimal antibody pairs for capture and detection of the S glycoprotein was determined via a three-stage process using the MSD immunoassay platform. MSD U-plex plates with a 10-plex array/well format were prepared for capture antibody binding as above. Antibodies were screened in a matrix format, acting both as capture and detector antibody.

Round 1. All 41 AbCellera antibodies were screened together in a matrix format using 10 ng/mL of trimeric S glycoprotein antigen (Acro Biosystems) in triplicate. The capture and detection antibody pairs that recorded 25% or greater ECL per plate were further evaluated over a greater range of S antigen concentration (1000, 100 and 10 ng/mL) to verify the initial results. The highest ECL readings across each concentration ranges were then used to rank antibodies for round 2 screening.

Round 2. Six antibody candidates from round 1 were evaluated further in a matrixed format alongside 3 antibodies from Sino Biological using 7-point dilutions of the S glycoprotein antigen in diluent 100 (ranging from 1250 to 0.016 pg/mL) in duplicate. Antibody pairs were ranked in terms of the limit of detection (LOD). Specificity was evaluated by challenging the pairs with irradiated viral cultures of SARS-CoV-2 and other human coronavirus species at concentrations equivalent to 10^4 $TCID_{50}$/mL or PFU/mL in Diluent 100.

Round 3: An additional 4 antibodies from Leinco were evaluated in a matrix format with the 4 best performing antibodies from round 2 and 2 from round 1. Their analytical sensitivity was evaluated by challenging the antibody pairs with a 7-point calibration curve of the S antigen, and a dilution series of the irradiated SARS-CoV-2. Specificity was evaluated by challenging the pairs with irradiated viral cultures and supernates of other human coronavirus species (OC43,229E, MERS and SARS) at concentrations equivalent to 10^4 $TCID_{50}$/mL or PFU/mL in Diluent 100. Antibody pairs were ranked in terms of LOD and ECL signal with the best performing pair further evaluated for clinical sensitivity and specificity with 53 clinical specimens.

Lateral flow assay Screening for Optimal Antibody Pairs

Antibody/antigen evaluation by SDS-PAGE

Antigens were evaluated for purity and size using SDS-PAGE. Concentration was measured for all proteins using BCA assay (ThermoFisher Scientific). Samples were premixed NuPAGE™ LDS 4x Sample Buffer (ThermoFisher Scientific) and heated at 70^{o}C for 10 minutes. Gels with a 4-12% Bis-Tris gradient were used to achieve separation. Novex Sharp Pre-stained protein standard (ThermoFisher Scientific) was used as a molecular weight marker Coomassie Imperial™ Protein Stain (ThermoFisher Scientific) was used to stain each gel and visualize protein bands.

Latex bead conjugation

For both test and control line detection conjugates, 400 nm carboxylic blue latex beads (Magsphere, Pasadena CA, USA) were washed three times with 0.1 M MES buffer (pH 6). Then, latex beads were activated using 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride / *N*-hydroxysuccinimide (ThermoFisher Scientific) coupling reagents at 0.15 and 10 mg/mL respectively for 30 minutes. Afterwards, the blue latex particles were conjugated in 1× PBS (pH 7.2) to various anti-spike antibodies at a w/w ratio of 20:1 and 10:1 (bead: antibody) for test and control line antibodies, respectively, for three hours. Finally, latex conjugates were quenched using 0.1 M ethanolamine before being washed and blocked with 6% (w/v) casein in H_2O (preparation method is proprietary), final concentration 1.2%, overnight. The latex conjugates were stored in buffer containing 50 mM borate (pH 8.5) and 1% casein. The latex conjugates were quantified using the spectrophotometer by measuring absorbance at 660 nm and comparing to absorbance of unconjugated beads.

LFA reagent deposition and strip assembly

Unlabeled capture antibodies were diluted to 1 mg/mL in 1× PBS (pH 7.4) with 2.5% (w/v) sucrose, and were striped at 1 µL/cm (ZX1010, BioDot, Irvine, CA, USA) on nitrocellulose CN95 (20 mm wide, CN95, Sartorius Lab Instruments GmbH & Co. KG, Otto-Brenner-Straße 20, Göttingen, Germany) and dried at 25°C for 30 min. The control line was 0.75 mg/mL Donkey anti-Chicken IgY (Jackson ImmunoResearch, West Grove, PA, USA), striped at 1 µL/cm. The test and control lines were located at 8 mm and 13 mm from the upstream edge of the nitrocellulose membrane. For antibody screening, the nitrocellulose was left unblocked.

The conjugate pad was dip-coated with two blocking solutions. First, 6613 conjugate pads (Ahlstrom-Munksjö Helsinki, Finland) were soaked in a 0.05% (w/v) Tween-20 in diH_2O solution for 15–20 seconds and dried at 40°C for 60 min. Pads were again soaked in 50mM borate (pH 8.5); 0.25% (w/v) Triton X-100; 1% (w/v) Surfactant-10G; 1% (w/v) sucrose; and 6% (w/v) casein for another 15–20 seconds. The conjugate pad was dried for 60 min at 40°C before assembly.

Card assembly was performed on a clamshell laminator (Matrix 2210, Kinematic Automation, Sonora, CA, USA). Pads were placed on the backing card in the following order: nitrocellulose, cover tape, conjugate pad, sample pad, wicking pad. Individual strips (3.3 mm wide) were cut with a Matrix 2360 sheet cutter (Kinematic Automation, Mono Vista, CA, USA) and assembled in cassettes (proprietary design) using an assembly roller (YK725, Kinbio Tech Co., Shanghai, China).

Hamilton screening procedure for LFA screening of antibodies

Antibody pairs were screened on an integrated robotic system we have previously used to test antibody performance directly on nitrocellulose[25;27]. In this system, the Hamilton STAR automated liquid handling robot (Hamilton Company, Reno, NV, USA), camera (IDS UI-1460SE-C-H detector with a Tamron M118FM16 lens) custom LFA holders, and custom control software developed in-house were combined to allow rapid screening of antibody pairs directly in LFA format. The robot used 8-channel pipetting for parallel application to LFAs and the camera for imaging. The custom LFA framework held a maximum of 96 LFA cassettes per robot run. The custom control software applied 1 µL of latex bead conjugate mix (0.15% anti-spike -latex bead, 0.1% or 0.05% Chicken IgY latex bead in 50mM borate [pH 8.5]) to the conjugate pad in the LFA. After a 10-minute delay to let the conjugate mix dry, 75 µL of sample diluted in 2.5% BSA in PBST, spike glycoprotein or buffer (2.5% BSA in PBST or 2.5% BSA and 1% IGEPAL in 1× PBS) was added to the sample pad. Images were acquired 20 minutes after sample addition. Four technical replicates were run for each antibody pair per sample type.

Screening recombinant antigens on LFAs

LFAs were screened across two rounds using a recombinant spike glycoprotein as the as the antigen target. The first, with the best-available at the time spike antigen (from Sino Biological), at 80 ng/mL. The second round used a different recombinant antigen produced in house was subsequently determined preferable, was also used at a concentration of 80 ng/mL,. A complete list of all pairs screened from all rounds is in Table 1si (suppl. info).

Results

Liquid immunoassay screening

All of the data generated from screening antibodies using the liquid platform in the following section is publicly accessible.[29] A total of 48 human monoclonal antibodies (AbCellera, 41; Sino Biological, 3; Leinco, 4) were assessed for their performance as capture and detection antibodies for the SARS CoV-2 S glycoprotein using the MSD U-PLEX immunoassay format across 3 rounds of testing. Each well in a 96 well U-PLEX plate can host 10 different capture antibodies in a geometric planar array by assessing ten capture antibodies per well (960 per plate) enabled rapid screening of multiple combinations to identify the most promising candidate pairs that would enable sensitive and specific capture and detection of SARS CoV-2.

In the preliminary evaluations, a recombinant S glycoprotein antigen expressed from insect cells (BEI) was used to screen the AbCellera antibodies however, this particular antigen resulted in the generation of very low ECL signals, at the concentration used. We postulate that as the post-translational modifications that can arise during antigen production will differ between insect cells and mammalian cells, the antigen initially used may have had or lack modifications that made it unsuitable for our study[30]. To identify an antigen most suitable for this work we evaluated 3 recombinant S glycoproteins across a range of dilutions (1000 to 0.24 pg/mL) using AbC525 and AbC397 as capture and detector respectively; this pair had generated the strongest ECL in the preliminary screen. The signal intensities and LOD varied with respect to each of the three antigens used. The

mammalian cell-derived recombinant S glycoprotein from Acro Biosystems produced the strongest and more consistent signal as compared to the baculovirus expressed antigens, and the lowest LOD (Figure 1). Thus, this antigen was selected for use as the standard in all antibody screens.

In round 1, 41 antibodies from AbCellera were assessed in both capture and detector format (1681 unique antibody pairings in total) using a low S glycoprotein antigen concentration of 10 ng/mL to allow for more stringent down-selection. Table 1 summarizes the Round 1 screening results in a matrixed array for each antibody combination. In the absence of a positive control assay, the ECL values from each array spot in each well were normalized based on the percentile of signal-noise (S-N) in each plate versus the spot with the maximum S-N produced in each plate. A total of 117 (7.0%) antibody pairs produced at least 25% of the maximum signal (marked in blue). These pairs, that consisted of 20 capture and 23 detection antibodies, were then further screened in a total of 460 combinations with S antigen in a range of 10, 100 and-1000 ng/mL to confirm the initial results (Figure 2). The ten antibody pairs that generated highest ECL signals were selected for evaluation in round 2, and included two capture antibodies (AbC447 and AbC525) and five detector antibodies (AbC513, AbC518, AbC459, AbC447 and AbC511). No self-pairing antibodies were identified presumably due to the presence of only single epitope on the recombinant antigen that would limit binding to only one form of the respective labeled antibody.

Figure 1. | Curves demonstrating assay performance of 3 commercially available trimeric S glycoproteins across a range of dilutions when screened via AbC525-AbC397 pair. ($LOD_{Acros\ Biosystems}$ = 286 pg/mL, $LOD_{Sino\ Biological}$ = 768 pg/mL, LOD_{BEI} = 19665 pg/mL).

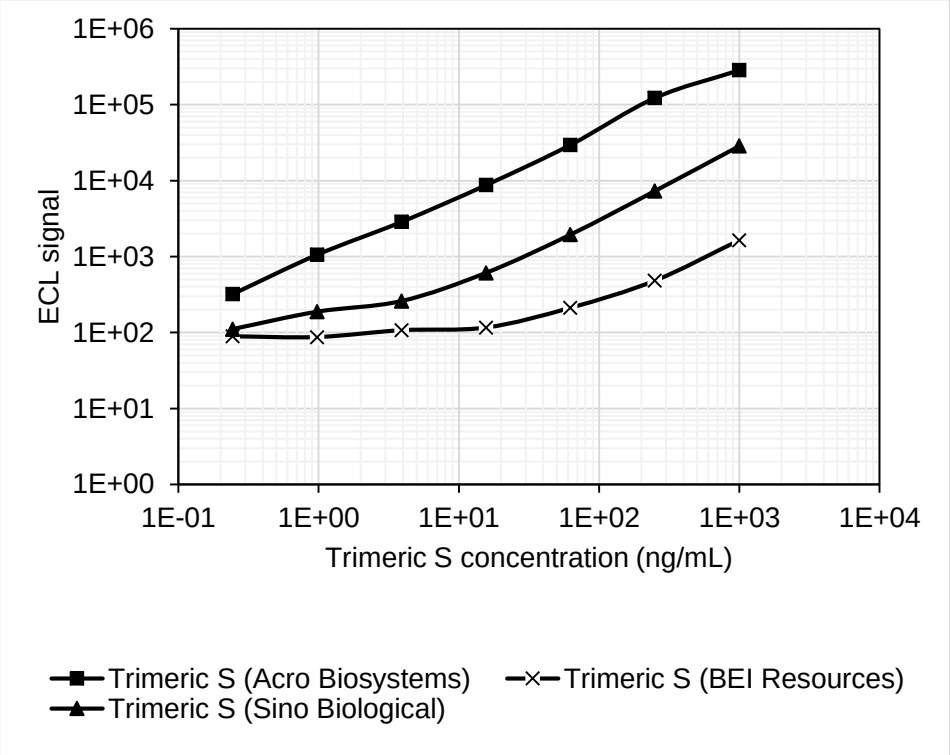
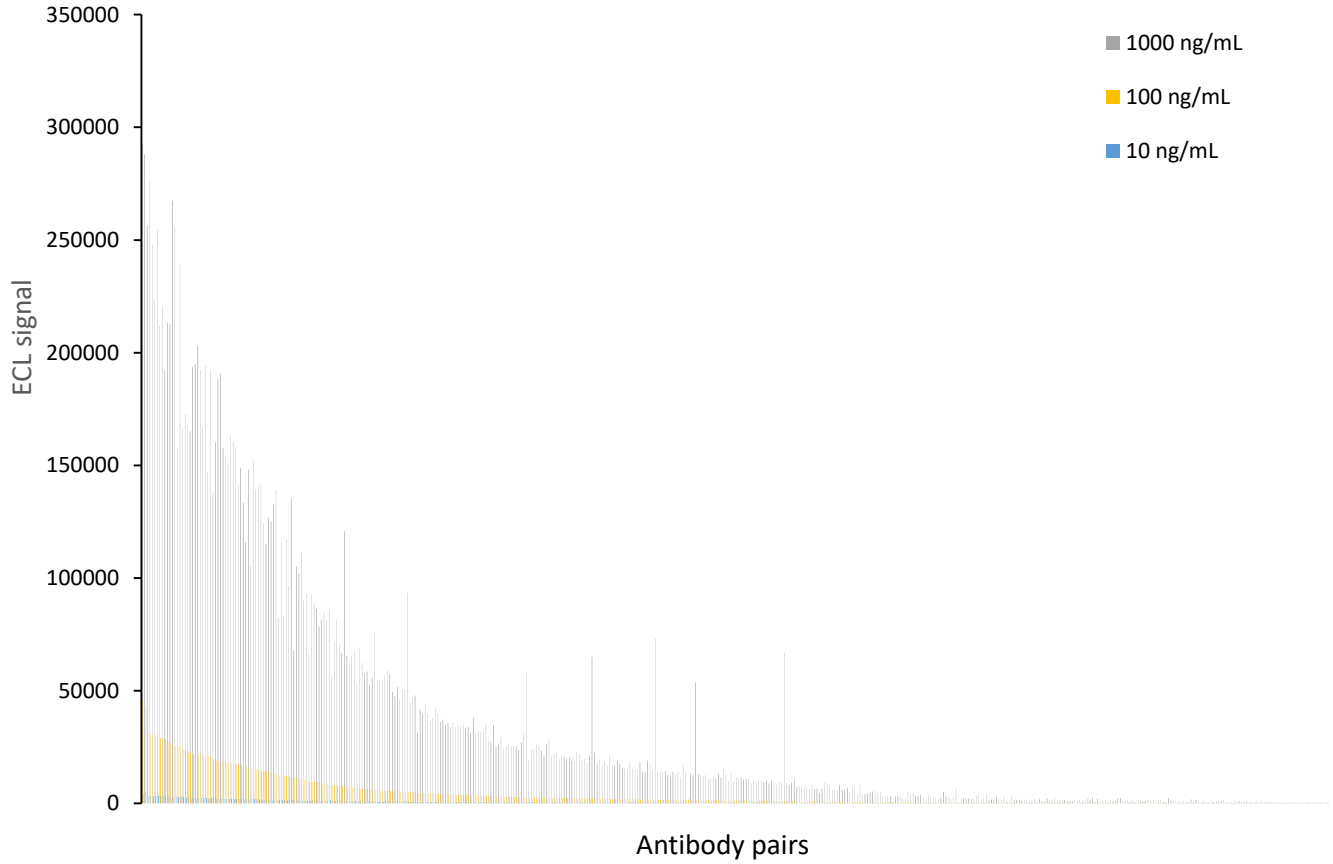


Table 1. | Performance of the 41 AbCellera antibodies in capture and detector formats in sandwich assays. The color gradient represents pair performance, measured as signal –noise. Darker indicates a pair performed better. Numbers inside the grid are normalized 0-1.0 according to the pairs with lowest and highest S-N. Legend ■0.75 – 1.00, ■ 0.50 – 0.74, ■ 0.25 – 0.49, □ 0.00 – 0.24.

AbC antibodies as captures (0.29 µg/mL) \ AbC antibodies as detectors (at 2 µg/mL)	275	277	283	285	291	308	310	313	353	357	359	369	397	415	429	453	459	463	478	489	491	500	530	554	258	298	393	400	447	455	469	486	511	513	518	525	557	558	561	574	585
275	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	-0.1	0.0	-0.1	0.1	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.1	0.1	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
277	0.1	0.0	0.3	0.0	0.1	0.2	0.0	0.0	0.1	0.0	0.0	0.1	0.5	0.0	0.1	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-0.1	0.1	0.0	0.0	0.5	0.4	0.2	0.2	0.3	-0.1	0.1	0.1	0.0	0.0	0.0	-0.1	0.0
283	0.0	0.0	0.0	0.0	0.1	0.1	0.0	0.1	0.0	0.0	-0.1	0.1	0.2	0.1	0.0	0.1	0.2	0.1	0.0	0.1	0.0	0.0	0.0	0.1	0.0	0.2	0.1	0.0	0.1	0.3	0.2	0.1	0.3	0.1	0.2	0.0	0.1	0.1	-0.1	0.0	0.0
285	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-0.1	0.1	0.0	-0.1	0.1	0.2	0.0	0.1	0.0	0.0	0.0	0.0	0.1	0.1	0.0	0.0	-0.1
291	0.0	0.0	1.0	0.0	0.0	0.4	0.0	0.0	0.0	0.0	0.0	0.0	0.9	0.0	0.4	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	-0.1	-0.1	-0.1	0.7	0.7	0.2	0.2	0.4	0.0	0.0	0.3	0.0	0.0	0.0	0.0	0.0
308	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	-0.1	0.0	0.2	0.1	0.1	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
310	0.0	0.1	0.1	0.0	0.1	0.1	0.0	0.1	0.0	0.1	-0.4	0.4	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.1	0.0	0.0	0.1	0.0	0.0	0.0	0.0	-0.2	0.0	0.2	0.0	0.0	0.3	-0.1	0.1	0.0	0.0	0.1	0.0	0.0	0.0
313	0.0	0.0	0.5	-0.1	0.0	0.3	0.0	0.0	0.0	0.0	0.0	0.0	0.9	0.1	0.1	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-0.1	-0.2	0.0	0.6	0.2	0.1	-0.1	0.0	-0.1	0.1	0.2	0.0	0.0	0.0	0.0	0.0
353	0.0	0.0	0.3	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.2	0.3	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	-0.2	0.4	0.4	0.1	0.2	0.2	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.1
357	0.0	0.0	-0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-0.1	0.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-0.2	0.1	0.1	-0.1	0.2	0.2	0.1	0.1	0.1	-0.1	0.1	0.0	0.0	0.0	0.0	0.0	0.0
359	0.0	0.0	0.2	-0.2	0.0	0.0	-0.1	0.0	0.0	0.0	0.0	0.0	0.2	-0.1	0.1	0.1	0.0	0.0	0.0	-0.1	0.0	0.0	0.0	0.0	0.1	-0.1	-0.1	-0.2	0.3	0.1	-0.1	-0.2	-0.1	0.0	-0.4	0.1	-0.1	0.0	0.0	0.0	-0.1
369	0.0	0.0	0.7	0.0	0.0	0.3	0.0	0.0	0.0	0.0	0.0	0.0	0.7	0.0	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	-0.1	-0.2	0.0	0.4	0.2	-0.1	0.0	-0.1	-0.1	-0.2	0.1	0.0	0.0	0.0	0.0	0.0
397	0.0	0.0	0.2	0.0	0.1	0.0	-0.1	0.1	0.1	0.0	0.0	0.0	0.0	0.3	0.0	0.1	0.3	0.2	0.0	0.2	0.0	0.1	0.0	0.2	0.2	0.0	0.0	0.0	0.2	0.2	-0.1	-0.1	-0.1	0.2	0.0	0.1	0.0	0.0	0.0	0.0	0.0
415	0.0	0.0	0.8	-0.1	0.0	0.3	0.0	0.0	0.0	0.0	0.1	0.0	1.0	0.1	0.2	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-0.2	-0.2	-0.1	0.5	0.3	-0.1	-0.3	-0.1	0.0	-0.2	0.2	0.0	0.0	0.0	0.1	0.1
429	0.0	0.2	0.3	-0.2	0.3	-0.1	-0.1	0.2	0.1	0.0	0.1	0.1	0.2	0.2	0.0	0.0	0.3	0.1	0.0	0.2	0.0	0.0	0.0	0.2	-0.1	-0.2	-0.1	-0.1	0.4	0.0	-0.1	0.2	-0.1	0.5	0.1	0.1	0.0	0.1	0.0	0.0	0.0
453	0.0	0.0	0.1	0.0	0.1	0.0	-0.1	0.1	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.1	-0.1	-0.1	0.0	0.0	0.3	0.0	0.1	-0.1	0.0	0.0	-0.1	0.1	0.0	0.0	0.0	0.0	-0.1
459	0.0	0.0	0.9	0.0	0.1	0.5	0.0	0.0	0.1	0.0	0.0	0.0	0.8	0.1	0.2	0.3	0.1	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-0.2	-0.1	-0.2	1.0	0.6	0.0	-0.1	0.2	0.2	-0.1	0.3	0.0	0.0	0.0	0.0	0.0
463	0.0	0.0	0.5	-0.2	0.0	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.8	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-0.3	-0.6	-0.3	-0.4	0.3	0.4	0.0	-0.1	-0.2	-0.1	-0.3	0.1	0.0	0.0	0.0	0.0	0.0
478	0.0	0.1	0.2	-0.1	0.1	-0.1	-0.2	0.0	0.0	0.0	0.0	0.0	0.2	0.1	0.0	0.0	0.2	0.1	0.0	0.0	0.0	0.0	0.0	0.1	-0.1	-0.1	0.0	-0.2	0.1	-0.1	-0.1	0.0	-0.3	0.2	-0.1	0.1	0.0	0.0	0.0	0.0	-0.1
489	0.0	0.0	0.4	-0.1	0.0	0.3	-0.1	0.0	0.0	-0.1	-0.1	-0.1	0.7	0.0	0.1	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.1	-0.1	0.0	0.1	0.1	0.1	-0.1	-0.2	-0.1	0.1	-0.1	0.1	0.0	-0.1	0.0	0.1	0.0
491	0.0	0.0	0.2	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.1	0.0	0.0	0.1	0.0	0.0	0.2	0.0	0.0	0.0	0.0	0.1
500	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1
530	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	-0.1	0.0
554	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
258	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-0.1	0.0	0.0	0.0
585	0.0	0.2	0.0	0.0	0.3	0.3	0.0	0.3	0.2	0.0	0.0	0.1	0.5	0.1	0.1	0.2	0.3	0.1	0.0	0.2	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.2	0.1	0.0	0.1	0.1	0.1	0.2	0.0	0.2	0.2	0.0	0.0	0.0
393	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-0.1	0.0
400	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	-0.1	0.1	0.0	0.1
447	0.2	0.8	1.0	0.0	0.8	0.4	0.0	0.7	0.3	0.0	0.1	0.3	0.6	0.7	0.0	0.4	0.8	0.4	0.0	0.6	0.1	0.1	0.0	0.6	0.0	0.0	0.0	0.0	0.1	0.4	0.0	0.3	0.3	1.0	0.6	1.0	0.2	0.9	0.0	0.0	0.5
455	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.1	0.1	0.0	0.0	0.0	0.0	0.0
469	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-0.1
486	0.0	0.0	0.3	0.0	0.0	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.4	0.0	0.2	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.1	0.0	0.0	0.0	0.0	0.0	0.2	0.0	-0.1	-0.1	-0.1	0.1
511	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.1	0.0	0.0	0.1	0.0	0.0	0.1	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.0	0.1	0.1	0.0	0.1
513	0.0	0.0	0.1	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.1
518	0.0	0.0	0.3	0.0	0.0	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.5	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.3	0.2	0.0	0.0	0.1	0.0	0.0	0.5	0.0	0.0	-0.1	-0.1	0.2
525	0.2	0.7	0.0	0.0	0.7	1.0	0.0	0.6	0.4	0.0	0.0	0.4	1.0	0.5	0.1	0.4	0.8	0.5	0.0	0.6	0.1	0.1	0.0	0.4	0.0	0.0	0.0	0.0	0.4	0.2	0.0	0.2	0.2	0.5	0.4	0.0	0.3	0.6	0.0	0.0	-0.1
557	0.0	0.0	0.1	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.0	0.0	0.0	-0.1	0.0
558	0.0	0.0	0.4	0.0	0.0	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.4	0.0	0.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.1	0.0	0.0	0.0	0.0	0.0	0.3	0.0	0.0	0.0	0.0	0.1
561	0.0	0.0	0.4	0.0	0.0	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.4	0.0	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.3	0.0	0.0	0.0	0.0	0.2
574	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.1	0.0	0.0	-0.1	0.0

Figure 2. | The 460 AbCellera antibody pairs ranked based on signal intensity of a range of concentrations of the trimeric S glycoprotein antigen. ECL, electrochemiluminescence.



In round 2, the 10 AbCellera optimal antibody pairs were assessed further in a matrix format alongside three antibodies from Sino Biological (MM443, MM57 and D003). Screening with an 8-point standard curve indicated that the Sino Biological antibodies resulted in higher ECL signals and lower LODs than the best AbCellera pair (AbC447/AbC513) (Table 2). Notably the Sino 447/MM43 and 447/D003 pairs exhibited similarly low LODs at 43 and 45 pg/mL respectively, in addition to the highest ECL signals at when challenged with ≥ 625 ng/mL of trimeric S antigen. Antibody pairs AbC447/MM43 and AbC447/D003 were then further challenged with a range of concentrations of SARS-CoV-2 virions, both

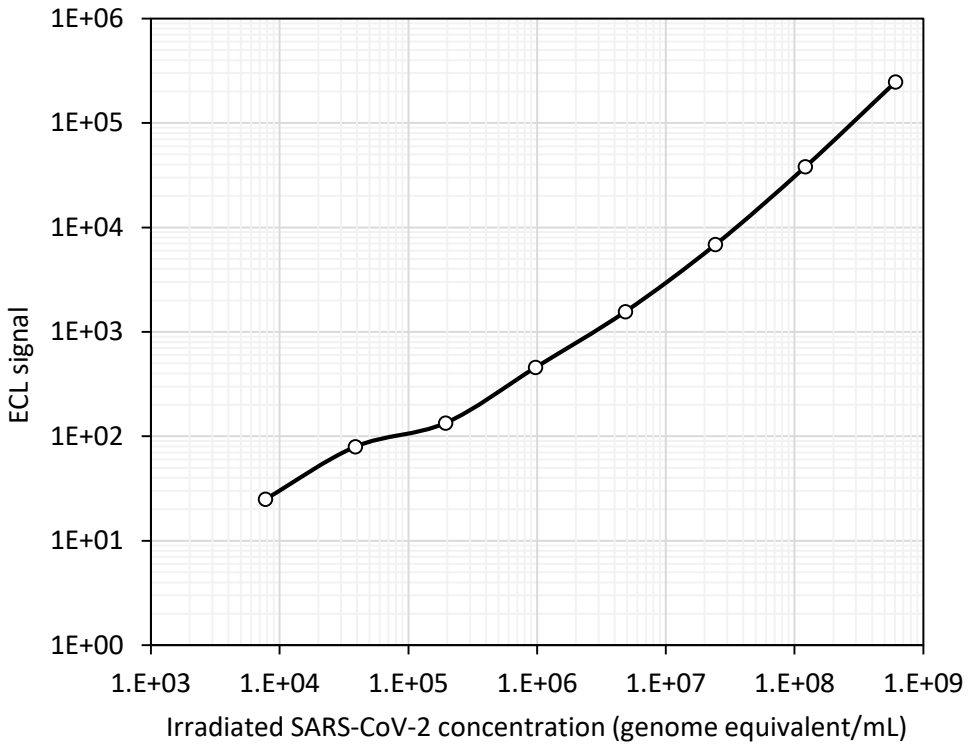
detecting down to 80 $TCID_{50}$/mL or approximately 4.86 x 10^5 genome equivalents/mL, with the AbC447-MM43 pair was determined to have the best performance characteristics at this stage.

Table 2. | Top antibody pairs from rounds 2 and 3 when challenged with S glycoprotein ranked based on their LOD. The electrochemiluminescent (ECL) signal intensity generated with 625 ng/mL S antigen is also included.

Capture Ig	Detector Ig	ECL signal (625 ng/mL antigen)	LOD (pg/mL)
Round 2			
AbC447	MM43	801003	43
AbC447	D003	871186	45
AbC447	AbC513	215672	71
D003	MM43	168029	94
MM43	AbC447	199763	174
Round 3			
L2381	MM43	1097669	3
L2355	L2215	2200950	4
L2838	L2215	2580454	6
L2381	L2215	1760625	7
L2355	MM43	1568224	8
AbC447	MM43	1145169	37

In round 3, four antibodies procured from Leinco (L2215, L2355, L2381 and L2838) screened with the 6 top antibody candidates identified from round 2 (D003, MM42, AbC447, and AbC513) and round 1 (AbC353 and AbC525). When used either as a capture or detector, the Leinco antibodies typically generated higher ECL signal and lower LODs than previously observed (Table 2), many with the LOD generally 5-10 times lower than for the best performing antibody in round 2. The L2381/MM43 and L2355/L2215 combinations had near identical LODs at 3 and 4 pg/mL respectively, with L2355/L2215 were selected for further study due to greater affinity to the target as indicated by significantly higher ECL signal when challenged with S antigen at 625 ng/mL (Table 2). The antibody pair L2355/L2215 was challenged with a titered SARS-CoV-2 (BEI), resulting in the generation of a dose-dependent curve (Figure 3) with an estimated LOD of 2 $TCID_{50}$/mL virions or 7.4 x10^3 genome equivalents/mL.

Figure 3. |The detection of serial dilutions of inactivated SARS-CoV-2 using the L2355 (capture)/L2215 (detector) antibody pair.



To demonstrate assay performance of the L2355/L2215 antibody pair with clinical samples, a panel of fifty-three de-identified clinical samples, comprising of 20 COVID19-negatives and 33 COVID19-positives were used to challenge the assay. Of these, 44 of the 53 samples were correctly identified as either positive or negative (Table 5). The viral load of the specimen was important as nine positive samples, each with a cycle threshold of > 29.5, were incorrectly scored as negative. This was likely in part due to dilution of the sample as each nasopharyngeal swab was collected in 3 mLs of viral transport medium. Overall the assay had a sensitivity and specificity of 73% and 100% respectively (Table 5) when compared to the RT-PCR results.

Table 5. | Performance characteristics of the L2355 (capture)/L2215 (detector) pairing relative to qRT-PCR when challenged with 53 clinical specimens.

	MSD Positive	MSD Negative	Total	Sensitivity (%)	Specificity (%)
RT PCR Positive	24	9	33	73	100
RT PCR Negative	0	20	20		
Total	24	29	53		

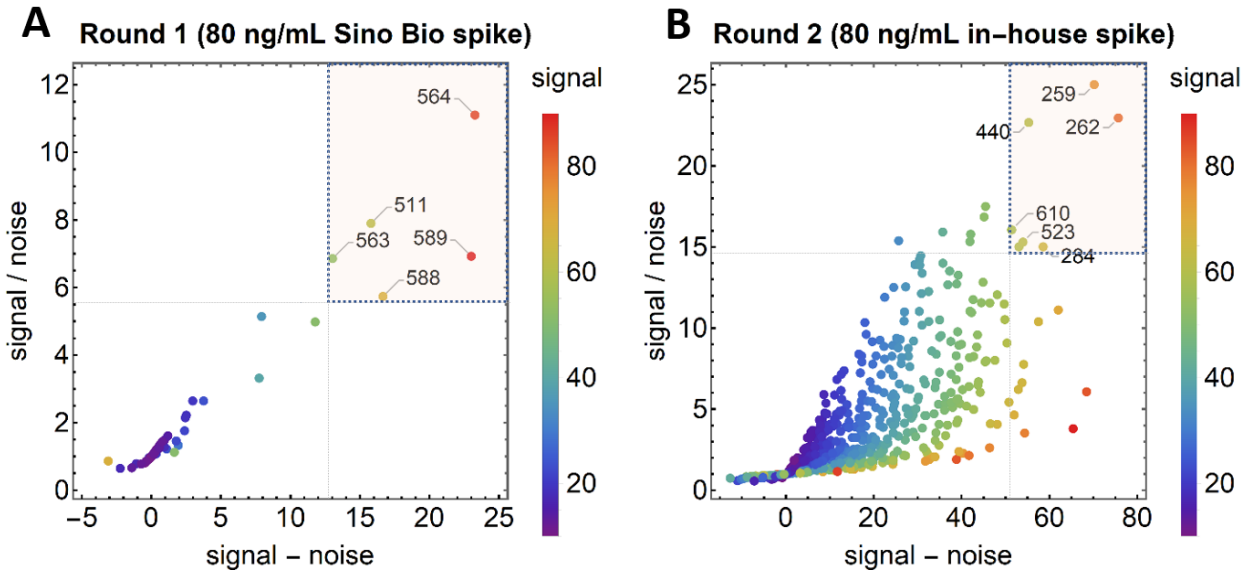
Specificity screening of top candidate pairs with other species of coronavirus in liquid immunoassays

The cross-reactivity of the antibody pairs from rounds 2 and 3 (Table 2) were also evaluated by challenging them with alpha- and beta-coronavirus isolates including inactivated MERS and SARS virions and human coronaviruses OC43 and 229E cell culture lysates at concentrations equivalent to 10^4 $TCID_{50}$/mL or 10^4 PFU/mL. None of the ten antibody pairs showed any cross-reactivity with other human CoV indicating a high specificity towards SARS-CoV-2.

Candidate screening via lateral flow assays.

The candidate antibodies were also evaluated in the LFA format in two rounds of screens, to assess if the performance of the candidate antibodies varied between the liquid and LFA test formats. A total of 8 antibodies (AbC131 from AbCellera, D003 from Sino Biological and 6 other antibodies from Sino Biological and Creative Diagnostics) were evaluated in Round 1 on LFAs in an 8 × 8 matrix (64 unique pairs, see Table 1si). For each pair, one antibody was striped on nitrocellulose as a test line (the “capture” antibody) and the other was coupled to latex nanoparticles using EDC/NHS chemistry (the “detector” antibody). The results of the first round are given in Figure 5(A). The positive control used round 1 was 80 ng/mL S glycoprotein from Sino Biological, selected due the presence of both the S1 and S2 domains of the native spike trimer. The negative control was 2.5% BSA in PBST.

Figure 5. | Performance of 621 individual antibody pairs in 2 rounds of screening on the LFA format as a function of signal / noise and signal - noise. Line intensities are shown as scatter plots for round 1 (**A**) and round 2 (**B**). Antibody pairs performing in the top 5 [for average rank by S/N and S-N] are overlaid with a semi-transparent box and numbered by their index (full list in Table 2si). "Sino Bio" antigen was sourced from Sino Biological and "in-house spike" recombinant antigen was produced in and purified at Global Health Labs.



After the first round, the best five pairs were D003/D002, D004/D002, D001/D004, D004/D001 and D003/D001 (index 564, 589, 511, 568, and 563, Table 3). Each of the top pairs from round 1 consisted exclusively of antibodies from Sino Biological, which was unsurprising considering recombinant antigen choice and the fact that most antibodies screened were from Sino Biological. As with the liquid immunoassay screen, self-pairs did not perform well, as expected, a consequence of the monomeric recombinant antigen likely containing a single copy of the target sequence. However, we would expect self-pairs to do better against the native antigen in clinical samples because it is trimeric. After round 1, 57 anti-S pairs were eliminated and the top seven pairs carried to round 2, along with 22 new antibodies. These new antibodies included the 12-top performing AbCellera antibodies from round 1 liquid immunoassay screen, MM43 from Sino Biological, and 9 antibodies from Leinco Technologies, including the 4 antibodies already screened with liquid immunoassay (Figure 1si).

Table 3. | Antibody pairs in the top five for both the signal to noise ratio [S/N] and signal minus noise [S-N] are ranked according to the round in which they were tested. Table 2si (supp. info) contains a complete list of all pairs screened.

Index	Capture	Detector	Average Rank	
			RD. 1	RD. 2
Round 1 Top 5 Performers				
564	D003	D002	1	302
589	D004	D002	2.5	-
511	D001	D004	3	-
588	D004	D001	4	-
563	D003	D001	4.5	156.5
Round 2 Top 5 Performers				
259	AbC459	L2355	-	1.5
262	AbC459	D001	-	1.5
440	L2355	AbC459	-	5.5
284	AbC525	L2355	-	9
523	D002	AbC459	-	11

The grid for round 2 was larger at 26 × 26 (616 pairs), however limited access to material meant 60 pairs were ultimately excluded (Figure 1si). Results from round 2 are shown in a scatterplot in Figure 5(B). The positive control used here was a trimeric spike glycoprotein produced in-house, considered superior to the recombinant form due to its ability to better mimic the protein folding seen in native structures. The negative control used was 2.5% BSA in PBST. Based on S/N and S-N metrics, the five best performing antibody indices from round 2 were 259, 262, 440, 284, 523, and 610 (Table 3).

Discussion

In this paper, we present the screening of a panel of antibodies targeting the S glycoprotein of SARS Cov-2 to identify candidate capture and detector pairs that may be suitable for development of LFA antigen detection assays. We gained access to a large private collection but with limited access to sufficient materials resulting in some antibodies being screened in one assay and not the other. Commercially available antibodies were typically screened on both formats. A key to this work is the availability of a good native antigen proxy, and as antigen sources can vary considerably, it is important to assess them prior to commencing work. Using the highly sensitive MSD immunoassay platform we are able to achieve an analytical sensitivity in the range of to 7.4×10^3 genomic copies/mL and a specificity of 100% when using a limited specimen panel. The TPP for a test for diagnosis or confirmation of acute or subacute SARS-CoV-2 infection, suitable for low or high-volume needs notes a

sensitivity of under 1000 copies which this test does not currently meet. However, the intent of this project was to screen antibodies that have optimal potential for implementation in LFAs, and not to develop a diagnostic assay. If necessary, the platform can use a further enhance signal format not used here, the S-PLEX which MSD claim can further improve sensitivity by 10 - 100X or into the lower femtogram range. While the S glycoprotein is less abundant that the N protein, there may be utility for combining S as a target to create highly sensitive multiplex immunoassays, with its additional distinct epitopes enabling improved accuracy, especially at lower limits of detection[31;32].

The liquid assays identified pairs that gave an analytical sensitivity to the S antigen into the low picogram range, a tenfold improvement over previous N immunoassays reported for SARS but the ECL detection feature of the MSD device does also offer greater sensitivity over traditional colorimetric detection employed by most enzyme immunoassay methods[33;34]. Interestingly, the assay format had a distinct effect on the optimal candidate pairs identified. The L2135 clone was the best antibody in either format and as both capture and detector. In contrast, no AbCellera antibodies showed good performance in the liquid assay, though in the LFA format AbC459 was present as capture or detector in 4/5 top pairs. The use of a different source of recombinant antigen may have played a role in this as we did observe some difference in binding using mammalian recombinant sources of antigen. This finding serves as an insight to LFA developers wherein screening of all antibodies should be performed on nitrocellulose rather than using traditional liquid immunoassays. The best antibodies candidates screened in the liquid format appeared to be highly specific to SARS-CoV-2 as they were not reactive with SARS, MERS and OC43 HCoVs that are in the same genus as SARS-CoV-2[35;36]. While we did not have access to HKU1, another beta-CoV species associated with respiratory illness, we do expect it is unlikely to be reactive as the other more closely related beta-CoVs screened were non-reactive.

On the LFA platform, the best pairs, as measured by S/N and S-N, were from a combination of vendors (e.g. AbCellera, Leinco, and Sino Biological), likely because these high-affinity antibodies were raised via unique processes and therefore recognize different epitopes on the antigen.

Interestingly, the liquid and LFA formats did identify very different optimal pairs for the detection of the S antigen. Restricted resources meant that entire antibody sets could not be fully evaluated on both platforms but it was evident that some pairs were better suited to one format over the other. In the liquid format, none of the AbCellera antibodies were in the top candidates as either capture or detector by round 3 but with the LFA, AbC459 and AbC525 were represented in several optimal pairings (Table 2). With the Sino Biological antibodies a similar trend was noted wherein no candidates shone with the liquid immunoassay format while the LFA had two, D001 and D002 (Tables 2 and 3). Antibodies from Leinco were highly represented in the optimal liquid assay design with each of the top five pairs having at least one Leinco antibody in the pairing. By contrast, with the top five candidates in the LFA format, three pairs used a single Leinco antibody, L2355, either as capture or detector, though in combination with differing antibodies to the liquid format.

Our goal is to qualify reagents and methods that are publicly available to any developer who sees value in their use, removing the need for them to invest time and resources on antibodies with little or no potential. Further work is ongoing with our groups to develop a POC LFA with the potential for manufacturing at scale. An advantage of using recombinant antibodies like those from AbCellera and Leinco is that the variable antibody region of single antigen-specific memory B cells derived from convalescent patients is cloned into an expression vector enabling cost

efficient scaled production of antibodies. In addition, this work uses recombinant IgG antibodies which are monomeric; with the possibility of manipulating the same variable region sequences to create recombinant IgM type antibodies, decameric forms of which may improve capture and/or detector efficiency leading to more effective rapid antigen assays for COVID-19 diagnosis.

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Competing interests

BCB, LK, KH, VP and YH are employees of AbCellera biologics.

Reference List

1. Huang, C.; Wang, Y.; Li, X.; Ren, L.; Zhao, J.; Hu, Y.; Zhang, L.; Fan, G.; Xu, J.; Gu, X.; Cheng, Z.; Yu, T.; Xia, J.; Wei, Y.; Wu, W.; Xie, X.; Yin, W.; Li, H.; Liu, M.; Xiao, Y.; Gao, H.; Guo, L.; Xie, J.; Wang, G.; Jiang, R.; Gao, Z.; Jin, Q.; Wang, J.; Cao, B. *Lancet* **2020,** *395*, 497-506.

2. WHO. Coronavirus disease (COVID-19) Situation Reports. 2020.
   Ref Type: Report

3. Cui, J.; Li, F.; Shi, Z. L. *Nat.Rev.Microbiol.* **2019,** *17*, 181-92.

4. Park, W. B.; Kwon, N. J.; Choi, S. J.; Kang, C. K.; Choe, P. G.; Kim, J. Y.; Yun, J.; Lee, G. W.; Seong, M. W.; Kim, N. J.; Seo, J. S.; Oh, M. D. *J.Korean Med.Sci.* **2020,** *35*, e84.

5. Yan, R.; Zhang, Y.; Li, Y.; Xia, L.; Guo, Y.; Zhou, Q. *Science* **2020,** *367*, 1444-48.

6. Zheng, M.; Song, L. *Cell Mol.Immunol.* **2020,** *17*, 536-38.

7. Premkumar, L.; Segovia-Chumbez, B.; Jadi, R.; Martinez, D. R.; Raut, R.; Markmann, A.; Cornaby, C.; Bartelt, L.; Weiss, S.; Park, Y.; Edwards, C. E.; Weimer, E.; Scherer, E. M.; Rouphael, N.; Edupuganti, S.; Weiskopf, D.; Tse, L. V.; Hou, Y. J.; Margolis, D.; Sette, A.; Collins, M. H.; Schmitz, J.; Baric, R. S.; de Silva, A. M. *Sci.Immunol.* **2020,** *5*.

8. Patel, R.; Babady, E.; Theel, E. S.; Storch, G. A.; Pinsky, B. A.; St, G. K.; Smith, T. C.; Bertuzzi, S. *mBio.* **2020,** *11*.

9. Dramé, M.; Tabue, M. T.; Proye, E.; Hequet, F.; Hentzien, M.; Kanagaratnam, L.; Godaert, L. *J.Med.Virol.* **2020**.

10. Kucirka, L. M.; Lauer, S. A.; Laeyendecker, O.; Boon, D.; Lessler, J. *Ann.Intern.Med.* **2020,** *173*, 262-67.

11. US food and Drug Administration. In Vitro Diagnostics EUAs. 2020. 7-14-**2020**.

12. Gorzalski, A. J.; Tian, H.; Laverdure, C.; Morzunov, S.; Verma, S. C.; VanHooser, S.; Pandori, M. W. *J.Clin.Virol.* **2020,** *129*, 104501.

13. Lieberman, J. A.; Pepper, G.; Naccache, S. N.; Huang, M. L.; Jerome, K. R.; Greninger, A. L. *J.Clin.Microbiol.* **2020**.

14. Pujadas, E.; Ibeh, N.; Hernandez, M. M.; Waluszko, A.; Sidorenko, T.; Flores, V.; Shiffrin, B.; Chiu, N.; Young-Francois, A.; Nowak, M. D.; Paniz-Mondolfi, A. E.; Sordillo, E. M.; Cordon-Cardo, C.; Houldsworth, J.; Gitman, M. R. *J.Med.Virol.* **2020**.

15. Kohmer, N.; Westhaus, S.; Rühl, C.; Ciesek, S.; Rabenau, H. F. *J.Clin.Virol.* **2020,** *129*, 104480.

16. ASM Advocacy. ASM Expresses Concern about Coronavirus Test Reagent Shortages. 4-20-**2020**. American Society for Microbiology.

17. Bong, C. L.; Brasher, C.; Chikumba, E.; McDougall, R.; Mellin-Olsen, J.; Enright, A. *Anesth.Analg.* **2020,** *131*, 86-92.

18. Niemz, A.; Ferguson, T. M.; Boyle, D. S. *Trends Biotechnol.* **2011,** *29*, 240-50.

19. Guo, L.; Ren, L.; Yang, S.; Xiao, M.; Chang, D.; Yang, F.; la Cruz, C. S.; Wang, Y.; Wu, C.; Xiao, Y.; Zhang, L.; Han, L.; Dang, S.; Xu, Y.; Yang, Q. W.; Xu, S. Y.; Zhu, H. D.; Xu, Y. C.; Jin, Q.; Sharma, L.; Wang, L.; Wang, J. *Clin.Infect.Dis.* **2020,** *71*, 778-85.

20. Li, R.; Pei, S.; Chen, B.; Song, Y.; Zhang, T.; Yang, W.; Shaman, J. *Science* **2020,** *368*, 489-93.

21. Figueroa, C.; Johnson, C.; Ford, N.; Sands, A.; Dalal, S.; Meurant, R.; Prat, I.; Hatzold, K.; Urassa, W.; Baggaley, R. *Lancet HIV.* **2018,** *5*, e277-e290.

22. Jang, I. K.; Das, S.; Barney, R. S.; Peck, R. B.; Rashid, A.; Proux, S.; Arinaitwe, E.; Rek, J.; Murphy, M.; Bowers, K.; Boadi, S.; Watson, J.; Nosten, F.; Greenhouse, B.; Chiodini, P. L.; Domingo, G. J. *Malar.J.* **2018,** *17*, 403.

23. Molesworth, A. M.; Ndhlovu, R.; Banda, E.; Saul, J.; Ngwira, B.; Glynn, J. R.; Crampin, A. C.; French, N. *J.Acquir.Immune.Defic.Syndr.* **2010,** *55*, 625-30.

24. WHO. COVID-19 Target product profiles for priority diagnostics to support response to the COVID-19 pandemic v.0.1. 1-41. 7-31-**2020**. Geneva, World Health Organization.

25. Grant, B. D.; Anderson, C. E.; Williford, J. R.; Alonzo, L. F.; Glukhova, V. A.; Boyle, D. S.; Weigl, B. H.; Nichols, K. P. *Anal.Chem.* **2020**.

26. Cate, D.; Hsieh, H.; Glukhova, V.; Bishop, J. D.; Hermansky, G. H.; Barrios-Lopez, B.; Grant, B. D.; Anderson, C. E.; Spencer, E.; Kuhn, S.; Gallagher, R.; Rivera, R.; Bennet, C.; Byrnes, S. A.; Connelly, J. T.; Dewan, P. K.; Boyle, D. S.; Weigl, B. H.; Nicols, K. P. *CHEMRxiv* **2020**.

27. Huynh, T., Cate, D. M., Nichols, K. P., Weigl, B. H., Anderson, C. E., Gasperino, D. J., Harston, S. P., Hsieh, H. V., Marzan, R., Williford, J. R., Oncina, C. I., and Glukhova, V. A. Robotic System for the Development Lateral Flow Assays. IEEE Global Humanitarian Technology Conference. **2019**. Institute of Electrical and Electronics Engineers Inc.

28. Washington COVID-19 Biorepository. 8-27-**2020**.

29. Cantera, J. L. Screening data for "Screening antibodies raised against the spike glycoprotein of SARS-CoV-2 to support the development of rapid antigen assays". **2020**. Harvard Dataverse.

30. McKenzie, E. A.; Abbott, W. M. *Methods* **2018,** *147*, 40-49.

31. Che, X. Y.; Qiu, L. W.; Pan, Y. X.; Wen, K.; Hao, W.; Zhang, L. Y.; Wang, Y. D.; Liao, Z. Y.; Hua, X.; Cheng, V. C.; Yuen, K. Y. *J.Clin.Microbiol.* **2004,** *42*, 2629-35.

32. Leung, D. T.; Tam, F. C.; Ma, C. H.; Chan, P. K.; Cheung, J. L.; Niu, H.; Tam, J. S.; Lim, P. L. *J.Infect.Dis.* **2004,** *190*, 379-86.

33. Che, X. Y.; Qiu, L. W.; Pan, Y. X.; Wen, K.; Hao, W.; Zhang, L. Y.; Wang, Y. D.; Liao, Z. Y.; Hua, X.; Cheng, V. C.; Yuen, K. Y. *J.Clin.Microbiol.* **2004,** *42*, 2629-35.

34. He, Q.; Du, Q.; Lau, S.; Manopo, I.; Lu, L.; Chan, S. W.; Fenner, B. J.; Kwang, J. *J.Virol.Methods* **2005,** *127*, 46-53.

35. Lau, S. K.; Li, K. S.; Tsang, A. K.; Lam, C. S.; Ahmed, S.; Chen, H.; Chan, K. H.; Woo, P. C.; Yuen, K. Y. *J.Virol.* **2013,** *87*, 8638-50.

36. Mesel-Lemoine, M.; Millet, J.; Vidalain, P. O.; Law, H.; Vabret, A.; Lorin, V.; Escriou, N.; Albert, M. L.; Nal, B.; Tangy, F. *J.Virol.* **2012,** *86*, 7577-87.

Supplementary Information

Figure 1si | Heat map of spike protein antibody pair performance in LFAs for round 2. Gray squares indicate pairs that were not tested. The color gradient represents pair performance, measured as signal – noise. Darker indicates a pair performed better. Numbers inside the grid are normalized 0-100 according to the pairs with lowest and highest S-N.

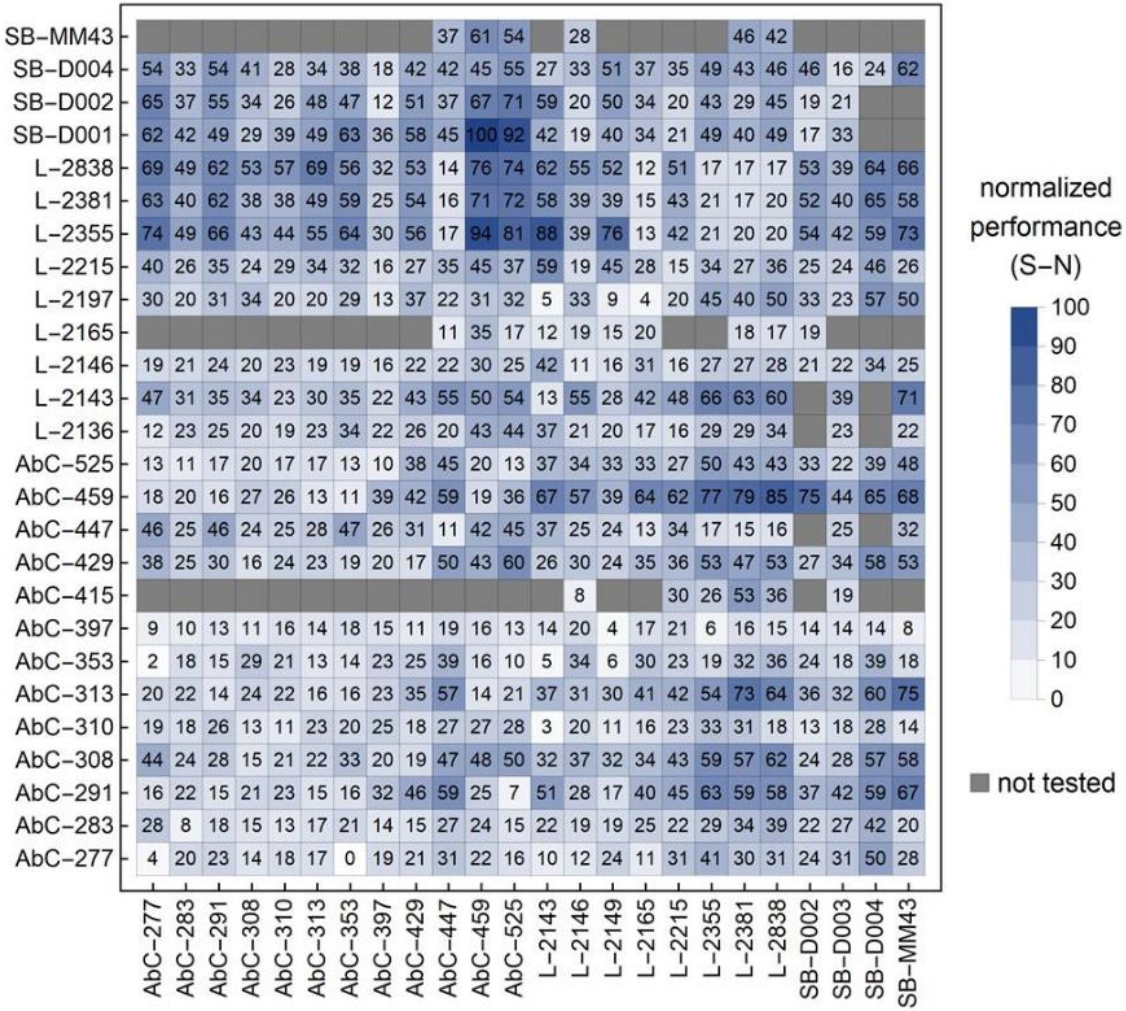

Table 1si | Vendor and other data for the anti-S glycoproteins antibodies screened via liquid and/or lateral flow immunoassay formats.

Vendor	Antibody name	Clone	Target region	Catalog / reference no.	Host	Isotype	Immunogen	Liquid immunoassay tested?	Lateral flow assay tested?
AbCellera	AbC275	275	S2	63974.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC277	277	S2	63997.1.a	Humanized	IgG1	trimeric spike	Y	Y
AbCellera	AbC283	283	S2	63980.1.a	Humanized	IgG1	trimeric spike	Y	Y
AbCellera	AbC285	285	S1	63983.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC291	291	S2	63986.1.a	Humanized	IgG1	trimeric spike	Y	Y
AbCellera	AbC308	308	S2	63992.1.a	Humanized	IgG1	trimeric spike	Y	Y
AbCellera	AbC310	310	undetermined	63997.1.a	Humanized	IgG1	trimeric spike	Y	Y
AbCellera	AbC313	313	S2	64000.1.a	Humanized	IgG1	trimeric spike	Y	Y
AbCellera	AbC353	353	S2	64003.1.a	Humanized	IgG1	trimeric spike	Y	Y
AbCellera	AbC357	357	undetermined	64006.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC359	359	undetermined	64009.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC369	369	S2	64012.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC397	397	S2	64020.1.a	Humanized	IgG1	trimeric spike	Y	Y
AbCellera	AbC415	415	S2	64026.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC429	429	S2	64031.1.a	Humanized	IgG1	trimeric spike	Y	Y
AbCellera	AbC453	453	S2	64036.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC459	459	S2	64042.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC463	463	S2	64045.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC478	478	undetermined	64051.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC489	489	S2	61061.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC491	491	undetermined	64064.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC500	500	S2	64067.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC530	530	undetermined	64082.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC554	554	S2	64087.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC258	258	S2	63971.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC298	298	S1	63989.1.a	Humanized	IgG1	trimeric spike	Y	N

AbCellera	AbC393	393	RBD	64017.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC400	400	RBD	64023.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC447	447	RBD	63900.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC455	455	S2	64039.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC469	469	undetermined	64048.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC486	486	S2	64058.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC511	511	S2	64070.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC513	513	S2	64073.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC518	518	S2	64076.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC525	525	S2	64079.1.a	Humanized	IgG1	trimeric spike	Y	Y
AbCellera	AbC557	557	S2	64090.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC558	558	S2	64093.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC561	561	S2	64096.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC574	574	undetermined	64101.1.a	Humanized	IgG1	trimeric spike	Y	N
AbCellera	AbC585	585	S2	64104.1.a	Humanized	IgG1	trimeric spike	Y	N
Creative Diagnostics	BIB112	BIB112	S1	CABT-CS031	Humanized	IgG	S1	N	Y
Creative Diagnostics	BIB112	BIB114	S1	CABT-CS033	Humanized	IgG	S1	N	Y
Sino Biological	D001	D001	undetermined	40150-D001	Mouse	IgG1	RBD	N	Y
Sino Biological	D002	D002	undetermined	40150-D002	Mouse	IgG1	RBD	N	Y
Sino Biological	D004	D004	undetermined	40150-D004	Mouse	IgG1	RBD	N	Y
Sino Biological	007	R007	undetermined	40150-R007	Rabbit	IgG	trimeric spike	N	Y
Sino Biological	D003	D003	RBD	40150-D003	Mouse/ human	IgG1	RBD	Y	N
Sino Biological	MM43	43	RBD	40591-MM43	Mouse	IgG1	S1	Y	Y
Sino Biological	MM57	57	RBD	40592-MM57	Mouse	IgG2b	RBD	Y	Y
Leinco	L2381	2381	RBD	LT4000	Human	IgG1	trimeric spike	Y	Y
Leinco	L2838	2838	RBD	LT3000	Human	IgG1	trimeric spike	Y	Y
Leinco	L2355	2355	RBD	LT5000	Human	IgG1	trimeric spike	Y	Y
Leinco	L2215	2215	S1 NTD	LT6000	Human	IgG1	trimeric spike	Y	Y
Leinco	L2136	2136	undetermined		Human	IgG1	trimeric spike	N	Y

Leinco	L2143	2143	undetermined		Human	IgG1	trimeric spike	N	Y
Leinco	L2146	2146	S1 NTD	LT2000	Human	IgG1	trimeric spike	N	Y
Leinco	L2197	2197	S1 NTD		Human	IgG1	trimeric spike	N	Y

Table 2si | Anti-spike protein antibody pairs screened on LFA.

Index	Capture antibody	Detector antibody	Average rank round 1	Average rank round 2
1	AbCellera clone 131	AbCellera clone 131	39	-
2	AbCellera clone 131	Creative Diagnostics CABT-CS031[BIB112]	45	-
3	AbCellera clone 131	Creative Diagnostics CABT-CS033[BIB114]	54.5	-
4	AbCellera clone 131	Sino Biological 40150-D001	25	-
5	AbCellera clone 131	Sino Biological 40150-D002	36.5	-
6	AbCellera clone 131	Sino Biological 40150-D003	50	-
7	AbCellera clone 131	Sino Biological 40150-D004	46	-
8	AbCellera clone 131	Sino Biological 40150-R007	35.5	-
9	AbCellera clone 277	AbCellera clone 277	-	551
10	AbCellera clone 277	AbCellera clone 283	-	349
11	AbCellera clone 277	AbCellera clone 291	-	473
12	AbCellera clone 277	AbCellera clone 308	-	113
13	AbCellera clone 277	AbCellera clone 310	-	440.5
14	AbCellera clone 277	AbCellera clone 313	-	368
15	AbCellera clone 277	AbCellera clone 353	-	563
16	AbCellera clone 277	AbCellera clone 397	-	549
17	AbCellera clone 277	AbCellera clone 429	-	222
18	AbCellera clone 277	AbCellera clone 447	-	139
19	AbCellera clone 277	AbCellera clone 459	-	392
20	AbCellera clone 277	AbCellera clone 525	-	523
21	AbCellera clone 277	Leinco clone 2136	-	526.5
22	AbCellera clone 277	Leinco clone 2143	-	149
23	AbCellera clone 277	Leinco clone 2146	-	449
24	AbCellera clone 277	Leinco clone 2197	-	308.5
25	AbCellera clone 277	Leinco clone 2215	-	127.5
26	AbCellera clone 277	Leinco clone 2355	-	13
27	AbCellera clone 277	Leinco clone 2381	-	57
28	AbCellera clone 277	Leinco clone 2838	-	106
29	AbCellera clone 277	Sino Biological 40150-D001	-	25
30	AbCellera clone 277	Sino Biological 40150-D002	-	17.5
31	AbCellera clone 277	Sino Biological 40150-D004	-	113.5
32	AbCellera clone 283	AbCellera clone 277	-	433
33	AbCellera clone 283	AbCellera clone 283	-	551.5
34	AbCellera clone 283	AbCellera clone 291	-	390
35	AbCellera clone 283	AbCellera clone 308	-	309.5
36	AbCellera clone 283	AbCellera clone 310	-	463
37	AbCellera clone 283	AbCellera clone 313	-	321.5
38	AbCellera clone 283	AbCellera clone 353	-	458
39	AbCellera clone 283	AbCellera clone 397	-	548.5
40	AbCellera clone 283	AbCellera clone 429	-	302.5
41	AbCellera clone 283	AbCellera clone 447	-	300
42	AbCellera clone 283	AbCellera clone 459	-	349.5
43	AbCellera clone 283	AbCellera clone 525	-	548
44	AbCellera clone 283	Leinco clone 2136	-	292.5
45	AbCellera clone 283	Leinco clone 2143	-	247
46	AbCellera clone 283	Leinco clone 2146	-	326.5
47	AbCellera clone 283	Leinco clone 2197	-	382.5
48	AbCellera clone 283	Leinco clone 2215	-	289
49	AbCellera clone 283	Leinco clone 2355	-	76
50	AbCellera clone 283	Leinco clone 2381	-	148.5
51	AbCellera clone 283	Leinco clone 2838	-	120.5
52	AbCellera clone 283	Sino Biological 40150-D001	-	114.5

Index	Capture antibody	Detector antibody	Average rank round 1	Average rank round 2
53	AbCellera clone 283	Sino Biological 40150-D002	-	156
54	AbCellera clone 283	Sino Biological 40150-D004	-	223
55	AbCellera clone 291	AbCellera clone 277	-	381
56	AbCellera clone 291	AbCellera clone 283	-	451.5
57	AbCellera clone 291	AbCellera clone 291	-	482.5
58	AbCellera clone 291	AbCellera clone 308	-	244.5
59	AbCellera clone 291	AbCellera clone 310	-	332
60	AbCellera clone 291	AbCellera clone 313	-	523
61	AbCellera clone 291	AbCellera clone 353	-	498
62	AbCellera clone 291	AbCellera clone 397	-	525.5
63	AbCellera clone 291	AbCellera clone 429	-	214
64	AbCellera clone 291	AbCellera clone 447	-	101.5
65	AbCellera clone 291	AbCellera clone 459	-	443
66	AbCellera clone 291	AbCellera clone 525	-	433
67	AbCellera clone 291	Leinco clone 2136	-	273.5
68	AbCellera clone 291	Leinco clone 2143	-	210.5
69	AbCellera clone 291	Leinco clone 2146	-	247
70	AbCellera clone 291	Leinco clone 2197	-	224
71	AbCellera clone 291	Leinco clone 2215	-	134.5
72	AbCellera clone 291	Leinco clone 2355	-	16
73	AbCellera clone 291	Leinco clone 2381	-	39.5
74	AbCellera clone 291	Leinco clone 2838	-	40
75	AbCellera clone 291	Sino Biological 40150-D001	-	64.5
76	AbCellera clone 291	Sino Biological 40150-D002	-	43.5
77	AbCellera clone 291	Sino Biological 40150-D004	-	65.5
78	AbCellera clone 308	AbCellera clone 277	-	510.5
79	AbCellera clone 308	AbCellera clone 283	-	497.5
80	AbCellera clone 308	AbCellera clone 291	-	386
81	AbCellera clone 308	AbCellera clone 308	-	480.5
82	AbCellera clone 308	AbCellera clone 310	-	518
83	AbCellera clone 308	AbCellera clone 313	-	319.5
84	AbCellera clone 308	AbCellera clone 353	-	284.5
85	AbCellera clone 308	AbCellera clone 397	-	550
86	AbCellera clone 308	AbCellera clone 429	-	451.5
87	AbCellera clone 308	AbCellera clone 447	-	262.5
88	AbCellera clone 308	AbCellera clone 459	-	199
89	AbCellera clone 308	AbCellera clone 525	-	354.5
90	AbCellera clone 308	Leinco clone 2136	-	360.5
91	AbCellera clone 308	Leinco clone 2143	-	183
92	AbCellera clone 308	Leinco clone 2146	-	387
93	AbCellera clone 308	Leinco clone 2197	-	226
94	AbCellera clone 308	Leinco clone 2215	-	230.5
95	AbCellera clone 308	Leinco clone 2355	-	99.5
96	AbCellera clone 308	Leinco clone 2381	-	153
97	AbCellera clone 308	Leinco clone 2838	-	78.5
98	AbCellera clone 308	Sino Biological 40150-D001	-	206.5
99	AbCellera clone 308	Sino Biological 40150-D002	-	157
100	AbCellera clone 308	Sino Biological 40150-D004	-	129.5
101	AbCellera clone 310	AbCellera clone 277	-	430
102	AbCellera clone 310	AbCellera clone 283	-	534
103	AbCellera clone 310	AbCellera clone 291	-	302
104	AbCellera clone 310	AbCellera clone 308	-	336
105	AbCellera clone 310	AbCellera clone 310	-	546.5
106	AbCellera clone 310	AbCellera clone 313	-	317.5
107	AbCellera clone 310	AbCellera clone 353	-	338
108	AbCellera clone 310	AbCellera clone 397	-	464.5
109	AbCellera clone 310	AbCellera clone 429	-	267

Index	Capture antibody	Detector antibody	Average rank round 1	Average rank round 2
110	AbCellera clone 310	AbCellera clone 447	-	246
111	AbCellera clone 310	AbCellera clone 459	-	256.5
112	AbCellera clone 310	AbCellera clone 525	-	408.5
113	AbCellera clone 310	Leinco clone 2136	-	357.5
114	AbCellera clone 310	Leinco clone 2143	-	290
115	AbCellera clone 310	Leinco clone 2146	-	270.5
116	AbCellera clone 310	Leinco clone 2197	-	336
117	AbCellera clone 310	Leinco clone 2215	-	185.5
118	AbCellera clone 310	Leinco clone 2355	-	99
119	AbCellera clone 310	Leinco clone 2381	-	133
120	AbCellera clone 310	Leinco clone 2838	-	45.5
121	AbCellera clone 310	Sino Biological 40150-D001	-	121.5
122	AbCellera clone 310	Sino Biological 40150-D002	-	379.5
123	AbCellera clone 310	Sino Biological 40150-D004	-	231.5
124	AbCellera clone 313	AbCellera clone 277	-	470
125	AbCellera clone 313	AbCellera clone 283	-	446
126	AbCellera clone 313	AbCellera clone 291	-	473
127	AbCellera clone 313	AbCellera clone 308	-	293
128	AbCellera clone 313	AbCellera clone 310	-	339.5
129	AbCellera clone 313	AbCellera clone 313	-	452.5
130	AbCellera clone 313	AbCellera clone 353	-	536
131	AbCellera clone 313	AbCellera clone 397	-	503.5
132	AbCellera clone 313	AbCellera clone 429	-	303
133	AbCellera clone 313	AbCellera clone 447	-	227.5
134	AbCellera clone 313	AbCellera clone 459	-	533.5
135	AbCellera clone 313	AbCellera clone 525	-	391.5
136	AbCellera clone 313	Leinco clone 2136	-	276
137	AbCellera clone 313	Leinco clone 2143	-	239.5
138	AbCellera clone 313	Leinco clone 2146	-	348.5
139	AbCellera clone 313	Leinco clone 2197	-	344
140	AbCellera clone 313	Leinco clone 2215	-	152.5
141	AbCellera clone 313	Leinco clone 2355	-	47.5
142	AbCellera clone 313	Leinco clone 2381	-	78
143	AbCellera clone 313	Leinco clone 2838	-	25
144	AbCellera clone 313	Sino Biological 40150-D001	-	63
145	AbCellera clone 313	Sino Biological 40150-D002	-	68.5
146	AbCellera clone 313	Sino Biological 40150-D004	-	165
147	AbCellera clone 353	AbCellera clone 277	-	559
148	AbCellera clone 353	AbCellera clone 283	-	415.5
149	AbCellera clone 353	AbCellera clone 291	-	485
150	AbCellera clone 353	AbCellera clone 308	-	268
151	AbCellera clone 353	AbCellera clone 310	-	421
152	AbCellera clone 353	AbCellera clone 313	-	451.5
153	AbCellera clone 353	AbCellera clone 353	-	507
154	AbCellera clone 353	AbCellera clone 397	-	464
155	AbCellera clone 353	AbCellera clone 429	-	429.5
156	AbCellera clone 353	AbCellera clone 447	-	143
157	AbCellera clone 353	AbCellera clone 459	-	551.5
158	AbCellera clone 353	AbCellera clone 525	-	520.5
159	AbCellera clone 353	Leinco clone 2136	-	223
160	AbCellera clone 353	Leinco clone 2143	-	247
161	AbCellera clone 353	Leinco clone 2146	-	397.5
162	AbCellera clone 353	Leinco clone 2197	-	329
163	AbCellera clone 353	Leinco clone 2215	-	226
164	AbCellera clone 353	Leinco clone 2355	-	66.5
165	AbCellera clone 353	Leinco clone 2381	-	92.5
166	AbCellera clone 353	Leinco clone 2838	-	133

Index	Capture antibody	Detector antibody	Average rank round 1	Average rank round 2
167	AbCellera clone 353	Sino Biological 40150-D001	-	50.5
168	AbCellera clone 353	Sino Biological 40150-D002	-	88.5
169	AbCellera clone 353	Sino Biological 40150-D004	-	170.5
170	AbCellera clone 397	AbCellera clone 277	-	458.5
171	AbCellera clone 397	AbCellera clone 283	-	513
172	AbCellera clone 397	AbCellera clone 291	-	342.5
173	AbCellera clone 397	AbCellera clone 308	-	432
174	AbCellera clone 397	AbCellera clone 310	-	376.5
175	AbCellera clone 397	AbCellera clone 313	-	390.5
176	AbCellera clone 397	AbCellera clone 353	-	400.5
177	AbCellera clone 397	AbCellera clone 397	-	502.5
178	AbCellera clone 397	AbCellera clone 429	-	439.5
179	AbCellera clone 397	AbCellera clone 447	-	351.5
180	AbCellera clone 397	AbCellera clone 459	-	249.5
181	AbCellera clone 397	AbCellera clone 525	-	541
182	AbCellera clone 397	Leinco clone 2136	-	357.5
183	AbCellera clone 397	Leinco clone 2143	-	402.5
184	AbCellera clone 397	Leinco clone 2146	-	479
185	AbCellera clone 397	Leinco clone 2197	-	525.5
186	AbCellera clone 397	Leinco clone 2215	-	486
187	AbCellera clone 397	Leinco clone 2355	-	270.5
188	AbCellera clone 397	Leinco clone 2381	-	350.5
189	AbCellera clone 397	Leinco clone 2838	-	297.5
190	AbCellera clone 397	Sino Biological 40150-D001	-	288.5
191	AbCellera clone 397	Sino Biological 40150-D002	-	530.5
192	AbCellera clone 397	Sino Biological 40150-D004	-	455.5
193	AbCellera clone 429	AbCellera clone 277	-	415.5
194	AbCellera clone 429	AbCellera clone 283	-	493
195	AbCellera clone 429	AbCellera clone 291	-	174
196	AbCellera clone 429	AbCellera clone 308	-	413.5
197	AbCellera clone 429	AbCellera clone 310	-	444
198	AbCellera clone 429	AbCellera clone 313	-	193
199	AbCellera clone 429	AbCellera clone 353	-	335
200	AbCellera clone 429	AbCellera clone 397	-	545.5
201	AbCellera clone 429	AbCellera clone 429	-	422.5
202	AbCellera clone 429	AbCellera clone 447	-	236
203	AbCellera clone 429	AbCellera clone 459	-	128
204	AbCellera clone 429	AbCellera clone 525	-	226
205	AbCellera clone 429	Leinco clone 2136	-	281
206	AbCellera clone 429	Leinco clone 2143	-	163.5
207	AbCellera clone 429	Leinco clone 2146	-	303
208	AbCellera clone 429	Leinco clone 2197	-	252.5
209	AbCellera clone 429	Leinco clone 2215	-	215.5
210	AbCellera clone 429	Leinco clone 2355	-	48.5
211	AbCellera clone 429	Leinco clone 2381	-	87
212	AbCellera clone 429	Leinco clone 2838	-	99.5
213	AbCellera clone 429	Sino Biological 40150-D001	-	43
214	AbCellera clone 429	Sino Biological 40150-D002	-	63
215	AbCellera clone 429	Sino Biological 40150-D004	-	149.5
216	AbCellera clone 447	AbCellera clone 277	-	331.5
217	AbCellera clone 447	AbCellera clone 283	-	334.5
218	AbCellera clone 447	AbCellera clone 291	-	95.5
219	AbCellera clone 447	AbCellera clone 308	-	147
220	AbCellera clone 447	AbCellera clone 310	-	329.5
221	AbCellera clone 447	AbCellera clone 313	-	80.5
222	AbCellera clone 447	AbCellera clone 353	-	192
223	AbCellera clone 447	AbCellera clone 397	-	428.5

Index	Capture antibody	Detector antibody	Average rank round 1	Average rank round 2
224	AbCellera clone 447	AbCellera clone 429	-	102
225	AbCellera clone 447	AbCellera clone 447	-	552
226	AbCellera clone 447	AbCellera clone 459	-	53.5
227	AbCellera clone 447	AbCellera clone 525	-	188.5
228	AbCellera clone 447	Leinco clone 2136	-	350.5
229	AbCellera clone 447	Leinco clone 2143	-	107
230	AbCellera clone 447	Leinco clone 2146	-	288.5
231	AbCellera clone 447	Leinco clone 2165	-	542
232	AbCellera clone 447	Leinco clone 2197	-	397
233	AbCellera clone 447	Leinco clone 2215	-	134
234	AbCellera clone 447	Leinco clone 2355	-	396.5
235	AbCellera clone 447	Leinco clone 2381	-	471.5
236	AbCellera clone 447	Leinco clone 2838	-	511.5
237	AbCellera clone 447	Sino Biological 40150-D001	-	88
238	AbCellera clone 447	Sino Biological 40150-D002	-	143.5
239	AbCellera clone 447	Sino Biological 40150-D004	-	139.5
240	AbCellera clone 447	Sino Biological 40591-MM43[43]	-	223
241	AbCellera clone 459	AbCellera clone 277	-	403
242	AbCellera clone 459	AbCellera clone 283	-	365.5
243	AbCellera clone 459	AbCellera clone 291	-	369
244	AbCellera clone 459	AbCellera clone 308	-	123.5
245	AbCellera clone 459	AbCellera clone 310	-	327.5
246	AbCellera clone 459	AbCellera clone 313	-	505.5
247	AbCellera clone 459	AbCellera clone 353	-	480
248	AbCellera clone 459	AbCellera clone 397	-	468.5
249	AbCellera clone 459	AbCellera clone 429	-	171
250	AbCellera clone 459	AbCellera clone 447	-	156.5
251	AbCellera clone 459	AbCellera clone 459	-	391.5
252	AbCellera clone 459	AbCellera clone 525	-	405
253	AbCellera clone 459	Leinco clone 2136	-	159
254	AbCellera clone 459	Leinco clone 2143	-	136.5
255	AbCellera clone 459	Leinco clone 2146	-	207.5
256	AbCellera clone 459	Leinco clone 2165	-	249.5
257	AbCellera clone 459	Leinco clone 2197	-	332
258	AbCellera clone 459	Leinco clone 2215	-	80
259	AbCellera clone 459	Leinco clone 2355	-	1.5
260	AbCellera clone 459	Leinco clone 2381	-	38.5
261	AbCellera clone 459	Leinco clone 2838	-	47.5
262	AbCellera clone 459	Sino Biological 40150-D001	-	1.5
263	AbCellera clone 459	Sino Biological 40150-D002	-	29
264	AbCellera clone 459	Sino Biological 40150-D004	-	121
265	AbCellera clone 459	Sino Biological 40591-MM43[43]	-	95.5
266	AbCellera clone 525	AbCellera clone 277	-	487
267	AbCellera clone 525	AbCellera clone 283	-	499.5
268	AbCellera clone 525	AbCellera clone 291	-	545
269	AbCellera clone 525	AbCellera clone 308	-	156
270	AbCellera clone 525	AbCellera clone 310	-	312
271	AbCellera clone 525	AbCellera clone 313	-	383
272	AbCellera clone 525	AbCellera clone 353	-	549
273	AbCellera clone 525	AbCellera clone 397	-	526.5
274	AbCellera clone 525	AbCellera clone 429	-	174.5
275	AbCellera clone 525	AbCellera clone 447	-	227
276	AbCellera clone 525	AbCellera clone 459	-	229.5
277	AbCellera clone 525	AbCellera clone 525	-	515.5
278	AbCellera clone 525	Leinco clone 2136	-	124
279	AbCellera clone 525	Leinco clone 2143	-	137
280	AbCellera clone 525	Leinco clone 2146	-	235.5

Index	Capture antibody	Detector antibody	Average rank round 1	Average rank round 2
281	AbCellera clone 525	Leinco clone 2165	-	474.5
282	AbCellera clone 525	Leinco clone 2197	-	292.5
283	AbCellera clone 525	Leinco clone 2215	-	132
284	AbCellera clone 525	Leinco clone 2355	-	9
285	AbCellera clone 525	Leinco clone 2381	-	75.5
286	AbCellera clone 525	Leinco clone 2838	-	62.5
287	AbCellera clone 525	Sino Biological 40150-D001	-	59
288	AbCellera clone 525	Sino Biological 40150-D002	-	29.5
289	AbCellera clone 525	Sino Biological 40150-D004	-	100
290	AbCellera clone 525	Sino Biological 40591-MM43[43]	-	170.5
291	Creative Diagnostics CABT-CS031[BIB112]	AbCellera clone 131	32.5	-
292	Creative Diagnostics CABT-CS031[BIB112]	Creative Diagnostics CABT-CS031[BIB112]	58	-
293	Creative Diagnostics CABT-CS031[BIB112]	Creative Diagnostics CABT-CS033[BIB114]	25	-
294	Creative Diagnostics CABT-CS031[BIB112]	Sino Biological 40150-D001	25	-
295	Creative Diagnostics	Sino Biological 40150-D002	31	-
296	CABT-CS031[BIB112]	Sino Biological 40150-D003	29.5	-
297	Creative Diagnostics CABT-CS031[BIB112]	Sino Biological 40150-D004	11	-
298	Creative Diagnostics CABT-CS031[BIB112]	Sino Biological 40150-R007	23	-
299	Creative Diagnostics CABT-CS033[BIB114]	AbCellera clone 131	39.5	-
300	Creative Diagnostics CABT-CS033[BIB114]	Creative Diagnostics CABT-CS031[BIB112]	20.5	-
301	Creative Diagnostics CABT-CS033[BIB114]	Creative Diagnostics CABT-CS033[BIB114]	56	-
302	Creative Diagnostics CABT-CS033[BIB114]	Sino Biological 40150-D001	48	-
303	Creative Diagnostics CABT-CS033[BIB114]	Sino Biological 40150-D002	49	-
304	Creative Diagnostics CABT-CS033[BIB114]	Sino Biological 40150-D003	17	-
305	Creative Diagnostics CABT-CS033[BIB114]	Sino Biological 40150-D004	12	-
306	Creative Diagnostics CABT-CS033[BIB114]	Sino Biological 40150-R007	13	-
307	Leinco clone 2143	AbCellera clone 277	-	536
308	Leinco clone 2143	AbCellera clone 283	-	409
309	Leinco clone 2143	AbCellera clone 291	-	201.5
310	Leinco clone 2143	AbCellera clone 308	-	267
311	Leinco clone 2143	AbCellera clone 310	-	562
312	Leinco clone 2143	AbCellera clone 313	-	216.5
313	Leinco clone 2143	AbCellera clone 353	-	552
314	Leinco clone 2143	AbCellera clone 397	-	512
315	Leinco clone 2143	AbCellera clone 429	-	317.5
316	Leinco clone 2143	AbCellera clone 447	-	281.5
317	Leinco clone 2143	AbCellera clone 459	-	147
318	Leinco clone 2143	AbCellera clone 525	-	303
319	Leinco clone 2143	Leinco clone 2136	-	264
320	Leinco clone 2143	Leinco clone 2143	-	520.5
321	Leinco clone 2143	Leinco clone 2146	-	234

Index	Capture antibody	Detector antibody	Average rank round 1	Average rank round 2
322	Leinco clone 2143	Leinco clone 2165	-	527
323	Leinco clone 2143	Leinco clone 2197	-	547.5
324	Leinco clone 2143	Leinco clone 2215	-	179.5
325	Leinco clone 2143	Leinco clone 2355	-	104.5
326	Leinco clone 2143	Leinco clone 2381	-	200.5
327	Leinco clone 2143	Leinco clone 2838	-	181
328	Leinco clone 2143	Sino Biological 40150-D001	-	273
329	Leinco clone 2143	Sino Biological 40150-D002	-	171
330	Leinco clone 2143	Sino Biological 40150-D004	-	379
331	Leinco clone 2146	AbCellera clone 277	-	521.5
332	Leinco clone 2146	AbCellera clone 283	-	456
333	Leinco clone 2146	AbCellera clone 291	-	349
334	Leinco clone 2146	AbCellera clone 308	-	262
335	Leinco clone 2146	AbCellera clone 310	-	425.5
336	Leinco clone 2146	AbCellera clone 313	-	331.5
337	Leinco clone 2146	AbCellera clone 353	-	323.5
338	Leinco clone 2146	AbCellera clone 397	-	437
339	Leinco clone 2146	AbCellera clone 415	-	542.5
340	Leinco clone 2146	AbCellera clone 429	-	306
341	Leinco clone 2146	AbCellera clone 447	-	331
342	Leinco clone 2146	AbCellera clone 459	-	105.5
343	Leinco clone 2146	AbCellera clone 525	-	277.5
344	Leinco clone 2146	Leinco clone 2136	-	385
345	Leinco clone 2146	Leinco clone 2143	-	149
346	Leinco clone 2146	Leinco clone 2146	-	547
347	Leinco clone 2146	Leinco clone 2165	-	437.5
348	Leinco clone 2146	Leinco clone 2197	-	300
349	Leinco clone 2146	Leinco clone 2215	-	397
350	Leinco clone 2146	Leinco clone 2355	-	140.5
351	Leinco clone 2146	Leinco clone 2381	-	170.5
352	Leinco clone 2146	Leinco clone 2838	-	114.5
353	Leinco clone 2146	Sino Biological 40150-D001	-	384.5
354	Leinco clone 2146	Sino Biological 40150-D002	-	336
355	Leinco clone 2146	Sino Biological 40150-D004	-	194
356	Leinco clone 2146	Sino Biological 40591-MM43[43]	-	321
357	Leinco clone 2149	AbCellera clone 277	-	407.5
358	Leinco clone 2149	AbCellera clone 283	-	454.5
359	Leinco clone 2149	AbCellera clone 291	-	475.5
360	Leinco clone 2149	AbCellera clone 308	-	286
361	Leinco clone 2149	AbCellera clone 310	-	537
362	Leinco clone 2149	AbCellera clone 313	-	281.5
363	Leinco clone 2149	AbCellera clone 353	-	547.5
364	Leinco clone 2149	AbCellera clone 397	-	559.5
365	Leinco clone 2149	AbCellera clone 429	-	373
366	Leinco clone 2149	AbCellera clone 447	-	380
367	Leinco clone 2149	AbCellera clone 459	-	230.5
368	Leinco clone 2149	AbCellera clone 525	-	308.5
369	Leinco clone 2149	Leinco clone 2136	-	419.5
370	Leinco clone 2149	Leinco clone 2143	-	345.5
371	Leinco clone 2149	Leinco clone 2146	-	476.5
372	Leinco clone 2149	Leinco clone 2165	-	497
373	Leinco clone 2149	Leinco clone 2197	-	539
374	Leinco clone 2149	Leinco clone 2215	-	226.5
375	Leinco clone 2149	Leinco clone 2355	-	114
376	Leinco clone 2149	Leinco clone 2381	-	275.5
377	Leinco clone 2149	Leinco clone 2838	-	208.5
378	Leinco clone 2149	Sino Biological 40150-D001	-	258

Index	Capture antibody	Detector antibody	Average rank round 1	round 2
379	Leinco clone 2149	Sino Biological 40150-D002	-	192.5
380	Leinco clone 2149	Sino Biological 40150-D004	-	217.5
381	Leinco clone 2165	AbCellera clone 277	-	530
382	Leinco clone 2165	AbCellera clone 283	-	380
383	Leinco clone 2165	AbCellera clone 291	-	214
384	Leinco clone 2165	AbCellera clone 308	-	222.5
385	Leinco clone 2165	AbCellera clone 310	-	483
386	Leinco clone 2165	AbCellera clone 313	-	176
387	Leinco clone 2165	AbCellera clone 353	-	327.5
388	Leinco clone 2165	AbCellera clone 397	-	477
389	Leinco clone 2165	AbCellera clone 429	-	226.5
390	Leinco clone 2165	AbCellera clone 447	-	524.5
391	Leinco clone 2165	AbCellera clone 459	-	74.5
392	Leinco clone 2165	AbCellera clone 525	-	279.5
393	Leinco clone 2165	Leinco clone 2136	-	462.5
394	Leinco clone 2165	Leinco clone 2143	-	255.5
395	Leinco clone 2165	Leinco clone 2146	-	280.5
396	Leinco clone 2165	Leinco clone 2165	-	404.5
397	Leinco clone 2165	Leinco clone 2197	-	551.5
398	Leinco clone 2165	Leinco clone 2215	-	281
399	Leinco clone 2165	Leinco clone 2355	-	529
400	Leinco clone 2165	Leinco clone 2381	-	500.5
401	Leinco clone 2165	Leinco clone 2838	-	537.5
402	Leinco clone 2165	Sino Biological 40150-D001	-	274
403	Leinco clone 2165	Sino Biological 40150-D002	-	230
404	Leinco clone 2165	Sino Biological 40150-D004	-	235.5
405	Leinco clone 2215	AbCellera clone 277	-	285.5
406	Leinco clone 2215	AbCellera clone 283	-	369.5
407	Leinco clone 2215	AbCellera clone 291	-	140
408	Leinco clone 2215	AbCellera clone 308	-	139
409	Leinco clone 2215	AbCellera clone 310	-	361.5
410	Leinco clone 2215	AbCellera clone 313	-	147.5
411	Leinco clone 2215	AbCellera clone 353	-	388
412	Leinco clone 2215	AbCellera clone 397	-	379
413	Leinco clone 2215	AbCellera clone 415	-	322
414	Leinco clone 2215	AbCellera clone 429	-	256
415	Leinco clone 2215	AbCellera clone 447	-	235
416	Leinco clone 2215	AbCellera clone 459	-	27
417	Leinco clone 2215	AbCellera clone 525	-	300.5
418	Leinco clone 2215	Leinco clone 2136	-	433
419	Leinco clone 2215	Leinco clone 2143	-	133
420	Leinco clone 2215	Leinco clone 2146	-	462
421	Leinco clone 2215	Leinco clone 2197	-	428
422	Leinco clone 2215	Leinco clone 2215	-	484
423	Leinco clone 2215	Leinco clone 2355	-	92.5
424	Leinco clone 2215	Leinco clone 2381	-	107
425	Leinco clone 2215	Leinco clone 2838	-	70.5
426	Leinco clone 2215	Sino Biological 40150-D001	-	296
427	Leinco clone 2215	Sino Biological 40150-D002	-	321.5
428	Leinco clone 2215	Sino Biological 40150-D004	-	174.5
429	Leinco clone 2355	AbCellera clone 277	-	210.5
430	Leinco clone 2355	AbCellera clone 283	-	266
431	Leinco clone 2355	AbCellera clone 291	-	35.5
432	Leinco clone 2355	AbCellera clone 308	-	58.5
433	Leinco clone 2355	AbCellera clone 310	-	210
434	Leinco clone 2355	AbCellera clone 313	-	61.5
435	Leinco clone 2355	AbCellera clone 353	-	454.5

Index	Capture antibody	Detector antibody	Average rank round 1	Average rank round 2
436	Leinco clone 2355	AbCellera clone 397	-	559.5
437	Leinco clone 2355	AbCellera clone 415	-	364
438	Leinco clone 2355	AbCellera clone 429	-	95
439	Leinco clone 2355	AbCellera clone 447	-	437
440	Leinco clone 2355	AbCellera clone 459	-	5.5
441	Leinco clone 2355	AbCellera clone 525	-	104
442	Leinco clone 2355	Leinco clone 2136	-	201
443	Leinco clone 2355	Leinco clone 2143	-	55
444	Leinco clone 2355	Leinco clone 2146	-	233.5
445	Leinco clone 2355	Leinco clone 2197	-	162
446	Leinco clone 2355	Leinco clone 2215	-	175
447	Leinco clone 2355	Leinco clone 2355	-	304.5
448	Leinco clone 2355	Leinco clone 2381	-	315
449	Leinco clone 2355	Leinco clone 2838	-	429
450	Leinco clone 2355	Sino Biological 40150-D001	-	65.5
451	Leinco clone 2355	Sino Biological 40150-D002	-	78
452	Leinco clone 2355	Sino Biological 40150-D004	-	64.5
453	Leinco clone 2381	AbCellera clone 277	-	347.5
454	Leinco clone 2381	AbCellera clone 283	-	295
455	Leinco clone 2381	AbCellera clone 291	-	119.5
456	Leinco clone 2381	AbCellera clone 308	-	101.5
457	Leinco clone 2381	AbCellera clone 310	-	311
458	Leinco clone 2381	AbCellera clone 313	-	90.5
459	Leinco clone 2381	AbCellera clone 353	-	329.5
460	Leinco clone 2381	AbCellera clone 397	-	481.5
461	Leinco clone 2381	AbCellera clone 415	-	208
462	Leinco clone 2381	AbCellera clone 429	-	168
463	Leinco clone 2381	AbCellera clone 447	-	502
464	Leinco clone 2381	AbCellera clone 459	-	25.5
465	Leinco clone 2381	AbCellera clone 525	-	217.5
466	Leinco clone 2381	Leinco clone 2136	-	219
467	Leinco clone 2381	Leinco clone 2143	-	94
468	Leinco clone 2381	Leinco clone 2146	-	255.5
469	Leinco clone 2381	Leinco clone 2165	-	429.5
470	Leinco clone 2381	Leinco clone 2197	-	205.5
471	Leinco clone 2381	Leinco clone 2215	-	274
472	Leinco clone 2381	Leinco clone 2355	-	339.5
473	Leinco clone 2381	Leinco clone 2381	-	428
474	Leinco clone 2381	Leinco clone 2838	-	445
475	Leinco clone 2381	Sino Biological 40150-D001	-	103
476	Leinco clone 2381	Sino Biological 40150-D002	-	190
477	Leinco clone 2381	Sino Biological 40150-D004	-	101.5
478	Leinco clone 2381	Sino Biological 40591-MM43[43]	-	118.5
479	Leinco clone 2838	AbCellera clone 277	-	345
480	Leinco clone 2838	AbCellera clone 283	-	257
481	Leinco clone 2838	AbCellera clone 291	-	123
482	Leinco clone 2838	AbCellera clone 308	-	61
483	Leinco clone 2838	AbCellera clone 310	-	449.5
484	Leinco clone 2838	AbCellera clone 313	-	70
485	Leinco clone 2838	AbCellera clone 353	-	300.5
486	Leinco clone 2838	AbCellera clone 397	-	497.5
487	Leinco clone 2838	AbCellera clone 415	-	284
488	Leinco clone 2838	AbCellera clone 429	-	166.5
489	Leinco clone 2838	AbCellera clone 447	-	480
490	Leinco clone 2838	AbCellera clone 459	-	20
491	Leinco clone 2838	AbCellera clone 525	-	186.5
492	Leinco clone 2838	Leinco clone 2136	-	199.5

Index	Capture antibody	Detector antibody	Average rank round 1	Average rank round 2
493	Leinco clone 2838	Leinco clone 2143	-	81.5
494	Leinco clone 2838	Leinco clone 2146	-	224
495	Leinco clone 2838	Leinco clone 2165	-	446
496	Leinco clone 2838	Leinco clone 2197	-	137
497	Leinco clone 2838	Leinco clone 2215	-	171.5
498	Leinco clone 2838	Leinco clone 2355	-	351.5
499	Leinco clone 2838	Leinco clone 2381	-	356.5
500	Leinco clone 2838	Leinco clone 2838	-	440
501	Leinco clone 2838	Sino Biological 40150-D001	-	69.5
502	Leinco clone 2838	Sino Biological 40150-D002	-	91
503	Leinco clone 2838	Sino Biological 40150-D004	-	98
504	Leinco clone 2838	Sino Biological 40591-MM43[43]	-	179.5
505	Sino Biological 40150-D001	AbCellera clone 131	38	-
506	Sino Biological 40150-D001	Creative Diagnostics CABT-CS031[BIB112]	43.5	-
507	Sino Biological 40150-D001	Creative Diagnostics CABT-CS033[BIB114]	43	-
508	Sino Biological 40150-D001	Sino Biological 40150-D001	22.5	-
509	Sino Biological 40150-D001	Sino Biological 40150-D002	28	-
510	Sino Biological 40150-D001	Sino Biological 40150-D003	9	-
511	Sino Biological 40150-D001	Sino Biological 40150-D004	3	-
512	Sino Biological 40150-D001	Sino Biological 40150-R007	16.5	-
513	Sino Biological 40150-D002	AbCellera clone 131	61	-
514	Sino Biological 40150-D002	AbCellera clone 277	-	379.5
515	Sino Biological 40150-D002	AbCellera clone 283	-	402
516	Sino Biological 40150-D002	AbCellera clone 291	-	218
517	Sino Biological 40150-D002	AbCellera clone 308	-	282.5
518	Sino Biological 40150-D002	AbCellera clone 310	-	522
519	Sino Biological 40150-D002	AbCellera clone 313	-	156
520	Sino Biological 40150-D002	AbCellera clone 353	-	350
521	Sino Biological 40150-D002	AbCellera clone 397	-	512.5
522	Sino Biological 40150-D002	AbCellera clone 429	-	243.5
523	Sino Biological 40150-D002	AbCellera clone 459	-	11
524	Sino Biological 40150-D002	AbCellera clone 525	-	260.5
525	Sino Biological 40150-D002	Creative Diagnostics CABT-CS031[BIB112]	53.5	-
526	Sino Biological 40150-D002	Creative Diagnostics CABT-CS033[BIB114]	62.5	-
527	Sino Biological 40150-D002	Leinco clone 2146	-	358
528	Sino Biological 40150-D002	Leinco clone 2165	-	386
529	Sino Biological 40150-D002	Leinco clone 2197	-	276
530	Sino Biological 40150-D002	Leinco clone 2215	-	265
531	Sino Biological 40150-D002	Leinco clone 2355	-	67
532	Sino Biological 40150-D002	Leinco clone 2381	-	81.5
533	Sino Biological 40150-D002	Leinco clone 2838	-	126.5
534	Sino Biological 40150-D002	Sino Biological 40150-D001	32.5	420.5
535	Sino Biological 40150-D002	Sino Biological 40150-D002	21	394
536	Sino Biological 40150-D002	Sino Biological 40150-D003	10	-
537	Sino Biological 40150-D002	Sino Biological 40150-D004	6.5	147
538	Sino Biological 40150-D002	Sino Biological 40150-R007	35.5	-
539	Sino Biological 40150-D003	AbCellera clone 131	52.5	-
540	Sino Biological 40150-D003	AbCellera clone 277	-	316.5
541	Sino Biological 40150-D003	AbCellera clone 283	-	345
542	Sino Biological 40150-D003	AbCellera clone 291	-	146
543	Sino Biological 40150-D003	AbCellera clone 308	-	249.5
544	Sino Biological 40150-D003	AbCellera clone 310	-	448.5
545	Sino Biological 40150-D003	AbCellera clone 313	-	241.5

Index	Capture antibody	Detector antibody	Average rank round 1	Average rank round 2
546	Sino Biological 40150-D003	AbCellera clone 353	-	456.5
547	Sino Biological 40150-D003	AbCellera clone 397	-	505.5
548	Sino Biological 40150-D003	AbCellera clone 415	-	447
549	Sino Biological 40150-D003	AbCellera clone 429	-	253
550	Sino Biological 40150-D003	AbCellera clone 447	-	293.5
551	Sino Biological 40150-D003	AbCellera clone 459	-	109
552	Sino Biological 40150-D003	AbCellera clone 525	-	378
553	Sino Biological 40150-D003	Creative Diagnostics CABT-CS031[BIB112]	6.5	-
554	Sino Biological 40150-D003	Creative Diagnostics CABT-CS033[BIB114]	47	-
555	Sino Biological 40150-D003	Leinco clone 2136	-	286.5
556	Sino Biological 40150-D003	Leinco clone 2143	-	191.5
557	Sino Biological 40150-D003	Leinco clone 2146	-	320.5
558	Sino Biological 40150-D003	Leinco clone 2197	-	366.5
559	Sino Biological 40150-D003	Leinco clone 2215	-	294
560	Sino Biological 40150-D003	Leinco clone 2355	-	120.5
561	Sino Biological 40150-D003	Leinco clone 2381	-	120.5
562	Sino Biological 40150-D003	Leinco clone 2838	-	169
563	Sino Biological 40150-D003	Sino Biological 40150-D001	4.5	156.5
564	Sino Biological 40150-D003	Sino Biological 40150-D002	1	302
565	Sino Biological 40150-D003	Sino Biological 40150-D003	34.5	-
566	Sino Biological 40150-D003	Sino Biological 40150-D004	15.5	408.5
567	Sino Biological 40150-D003	Sino Biological 40150-R007	60.5	-
568	Sino Biological 40150-D004	AbCellera clone 131	42.5	-
569	Sino Biological 40150-D004	AbCellera clone 277	-	187
570	Sino Biological 40150-D004	AbCellera clone 283	-	191.5
571	Sino Biological 40150-D004	AbCellera clone 291	-	71
572	Sino Biological 40150-D004	AbCellera clone 308	-	64
573	Sino Biological 40150-D004	AbCellera clone 310	-	331
574	Sino Biological 40150-D004	AbCellera clone 313	-	55.5
575	Sino Biological 40150-D004	AbCellera clone 353	-	247
576	Sino Biological 40150-D004	AbCellera clone 397	-	510.5
577	Sino Biological 40150-D004	AbCellera clone 429	-	95
578	Sino Biological 40150-D004	AbCellera clone 459	-	26.5
579	Sino Biological 40150-D004	AbCellera clone 525	-	184.5
580	Sino Biological 40150-D004	Creative Diagnostics CABT-CS031[BIB112]	8	-
581	Sino Biological 40150-D004	Creative Diagnostics CABT-CS033[BIB114]	51	-
582	Sino Biological 40150-D004	Leinco clone 2146	-	209.5
583	Sino Biological 40150-D004	Leinco clone 2197	-	93.5
584	Sino Biological 40150-D004	Leinco clone 2215	-	107
585	Sino Biological 40150-D004	Leinco clone 2355	-	39
586	Sino Biological 40150-D004	Leinco clone 2381	-	32
587	Sino Biological 40150-D004	Leinco clone 2838	-	42.5
588	Sino Biological 40150-D004	Sino Biological 40150-D001	4	-
589	Sino Biological 40150-D004	Sino Biological 40150-D002	2.5	-
590	Sino Biological 40150-D004	Sino Biological 40150-D003	63.5	-
591	Sino Biological 40150-D004	Sino Biological 40150-D004	15.5	289.5
592	Sino Biological 40150-D004	Sino Biological 40150-R007	24.5	-
593	Sino Biological 40591-MM43[43]	AbCellera clone 277	-	381
594	Sino Biological 40591-MM43[43]	AbCellera clone 283	-	445
595	Sino Biological 40591-MM43[43]	AbCellera clone 291	-	105
596	Sino Biological 40591-MM43[43]	AbCellera clone 308	-	81

Index	Capture antibody	Detector antibody	Average rank round 1	Average rank round 2
597	Sino Biological 40591-MM43[43]	AbCellera clone 310	-	510.5
598	Sino Biological 40591-MM43[43]	AbCellera clone 313	-	57
599	Sino Biological 40591-MM43[43]	AbCellera clone 353	-	470
600	Sino Biological 40591-MM43[43]	AbCellera clone 397	-	548
601	Sino Biological 40591-MM43[43]	AbCellera clone 429	-	153.5
602	Sino Biological 40591-MM43[43]	AbCellera clone 447	-	278
603	Sino Biological 40591-MM43[43]	AbCellera clone 459	-	51
604	Sino Biological 40591-MM43[43]	AbCellera clone 525	-	173.5
605	Sino Biological 40591-MM43[43]	Leinco clone 2136	-	341.5
606	Sino Biological 40591-MM43[43]	Leinco clone 2143	-	27
607	Sino Biological 40591-MM43[43]	Leinco clone 2146	-	292
608	Sino Biological 40591-MM43[43]	Leinco clone 2197	-	230
609	Sino Biological 40591-MM43[43]	Leinco clone 2215	-	310.5
610	Sino Biological 40591-MM43[43]	Leinco clone 2355	-	11
611	Sino Biological 40591-MM43[43]	Leinco clone 2381	-	68
612	Sino Biological 40591-MM43[43]	Leinco clone 2838	-	53.5
613	Sino Biological 40591-MM43[43]	Sino Biological 40150-D004	-	76.5
614	Sino Biological 40150-R007	AbCellera clone 131	53.5	-
615	Sino Biological 40150-R007	Creative Diagnostics CABT-CS031[BIB112]	22	-
616	Sino Biological 40150-R007	Creative Diagnostics CABT-CS033[BIB114]	59.5	-
617	Sino Biological 40150-R007	Sino Biological 40150-D001	58	-
618	Sino Biological 40150-R007	Sino Biological 40150-D002	40	-
619	Sino Biological 40150-R007	Sino Biological 40150-D003	26	-
620	Sino Biological 40150-R007	Sino Biological 40150-D004	19	-
621	Sino Biological 40150-R007	Sino Biological 40150-R007	61	-