

Bio-Plex Pro Human IgG SARS-CoV-2 Serology Assays Protocol

Introduction

This semiquantitative protocol is for use with Bio-Plex Pro Human IgG SARS-CoV-2 N/RBD/S1/S2 Serology Assays. Recommendations are included about how to use the VIROTROL SARS-CoV-2 serological controls as a reference standard to generate U/ml quantitation values.

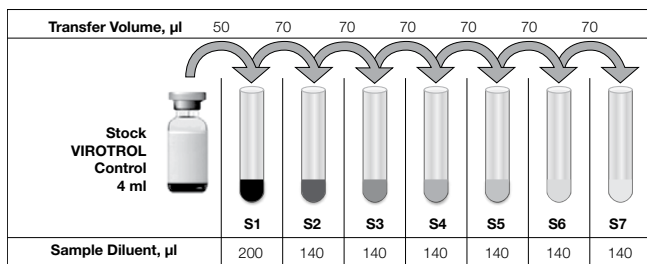
Required Materials

- VIROTROL SARS-CoV-2 Single Level Control (Bio-Rad Laboratories, Inc., catalog #200300A)
- Bio-Plex Pro Human IgG SARS-CoV-2 N/RBD/S1/S2 4-Plex Panel (Bio-Rad, #12014634)

Standard Curve Preparation

Run a seven-point standard curve starting at a 1:5 dilution of the stock VIROTROL. The control comes in liquid form at 4 ml, so one vial of VIROTROL should be enough for 83 assays when S1 is run in duplicate or triplicate.

- Dilute the contents of the vial fivefold up to a minimum volume of 250 μ l. To make S1, add 50 μ l of stock VIROTROL Control to 200 μ l serology sample diluent.
- Make threefold serial dilutions from S1 up to seven standard curve points. The volumes provided are for duplicate standard curve points.
 - Fill tubes for S2–S7 with 140 μ l serology sample diluent.
 - To make S2, add 70 μ l of S1 to 140 μ l serology sample diluent.
 - Repeat the serial dilution until you reach S7 by adding 70 μ l of the prior standard curve point dilution to 140 μ l of the following standard curve point.



VIROTROL Control standard curve dilution scheme.

Standard Curve Point	Threefold Serial Dilution	Value Assignment (U/ml)
S1	1:5	20,000
S2	1:15	6,666.7
S3	1:45	2,222.2
S4	1:135	740.7
S5	1:405	246.9
S6	1:1,215	82.3
S7	1:3,645	27.4

Sample Preparation

Dilute samples 1:1,000.

- Dilute 100-fold: Add 2 μ l of stock to 198 μ l serology sample diluent.
- Dilute tenfold: Add 20 μ l of 1:100 dilution to 180 μ l serology sample diluent.

Note: Adjust sample dilution scheme as needed for very high- and low-level anti-SARS-CoV-2 N/RBD/S1/S2 IgG samples. Dilutions of 1:500 and 1:300 have worked for some samples.

For bead, detection antibody, and streptavidin-phycoerythrin preparation, refer to the Bio-Plex Pro Human SARS-CoV-2 Serology Assays Quick Guide (1000133854) or Instruction Manual (10000133853).

For research use only.
Not for use in diagnostic procedures.

Visit [bio-rad.com/SARSCoV2Serology](https://www.bio-rad.com/SARSCoV2Serology) for more information.

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