

Receptor-Binding Domain (RBD) Antibodies Contribute More to SARS-CoV-2 Neutralization When Target Cells Express High Levels of ACE2

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Neutralization assays are experimental surrogates for the effectiveness of infection- or vaccine-elicited polyclonal antibodies and therapeutic monoclonal antibodies targeting SARS-CoV-2.

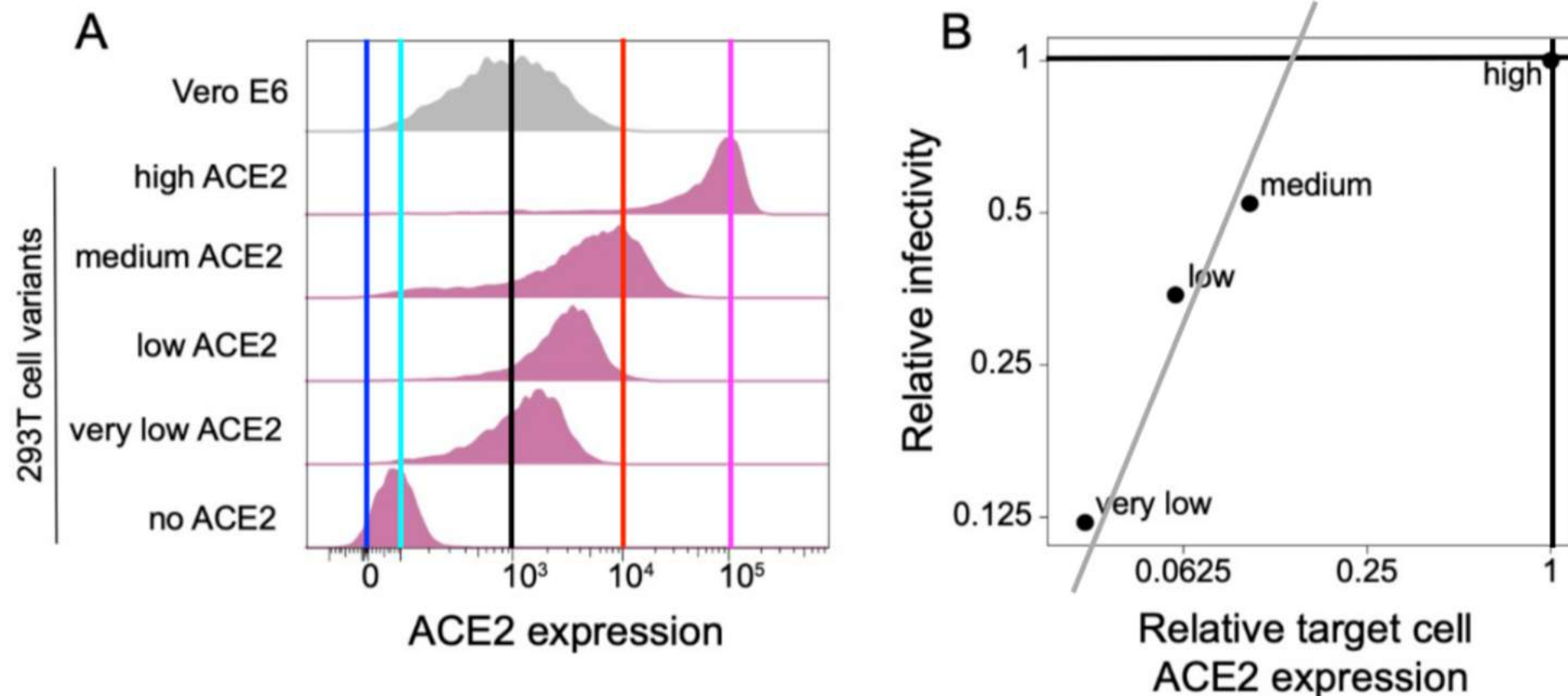


Figure 1. 293T cell clones expressing ACE2 at different levels. (A) ACE2 expression in 293T cells engineered to express different levels of ACE2. ACE2 surface expression was measured by flow cytometry, and the histograms show the distribution of expression levels over a population of cells. Vero E6 cells are included for comparison. (B) Relationship between ACE2 expression in the four 293T target cell clones and infection by lentiviral particles pseudotyped with the SARS-CoV-2 D614G spike.

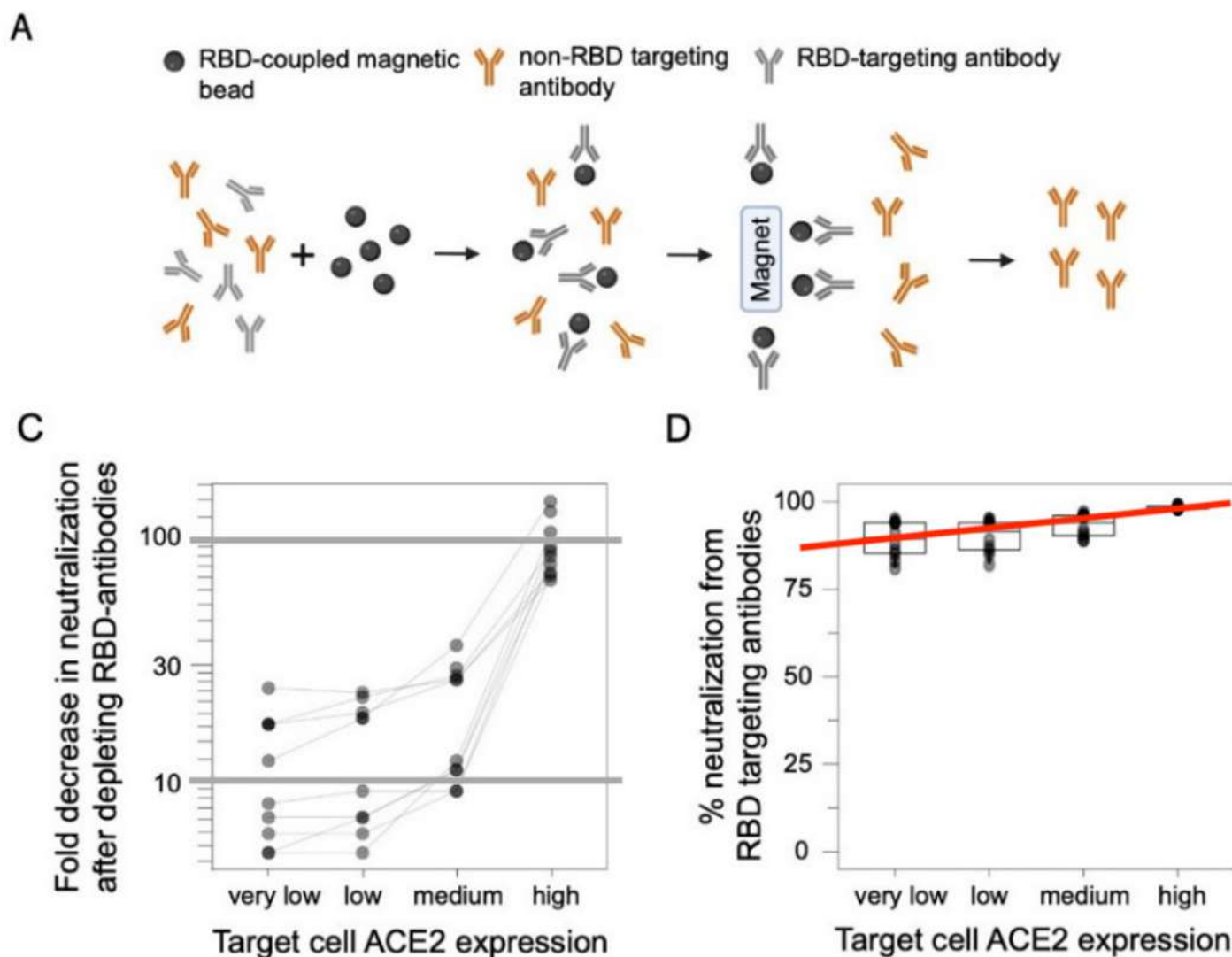


Figure 2. RBD-targeted antibodies make a larger contribution to serum neutralization when target cells express higher levels of ACE2. (A) Process for depleting RBD-targeted antibodies from polyclonal human serum. RBD-coupled magnetic beads are incubated with sera. The RBD-targeting antibodies bind the beads and are then removed from sera by magnetic separation. RBD-targeting antibody depletion was confirmed by ELISA (Figure S2). This figure was created with BioRender.com. (B) Neutralization of D614G spike-pseudotyped lentiviral particles by polyclonal human sera from ten different individuals with or without depletion of RBD-targeting antibodies, as measured on target cells expressing different levels of ACE2. Neutralization is reported as the neutralization titer 50% (NT_{50}), which is the reciprocal serum dilution that neutralizes half of the virus. The dashed red line represents the limit of detection (NT_{50} of 25), and values less than this limit are assigned a value of 25. (C) Fold decrease in neutralization after depleting the RBD antibodies and (D) percent of neutralization due to RBD-targeting antibodies for the sera shown in panel (B), calculated by subtracting NT_{50} values for depleted sera from non-depleted sera and expressed as percentage of non-depleted sera neutralization.