Establishment of an anti-RBD threshold that predicts robust neutralizing activity

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BACKGROUND

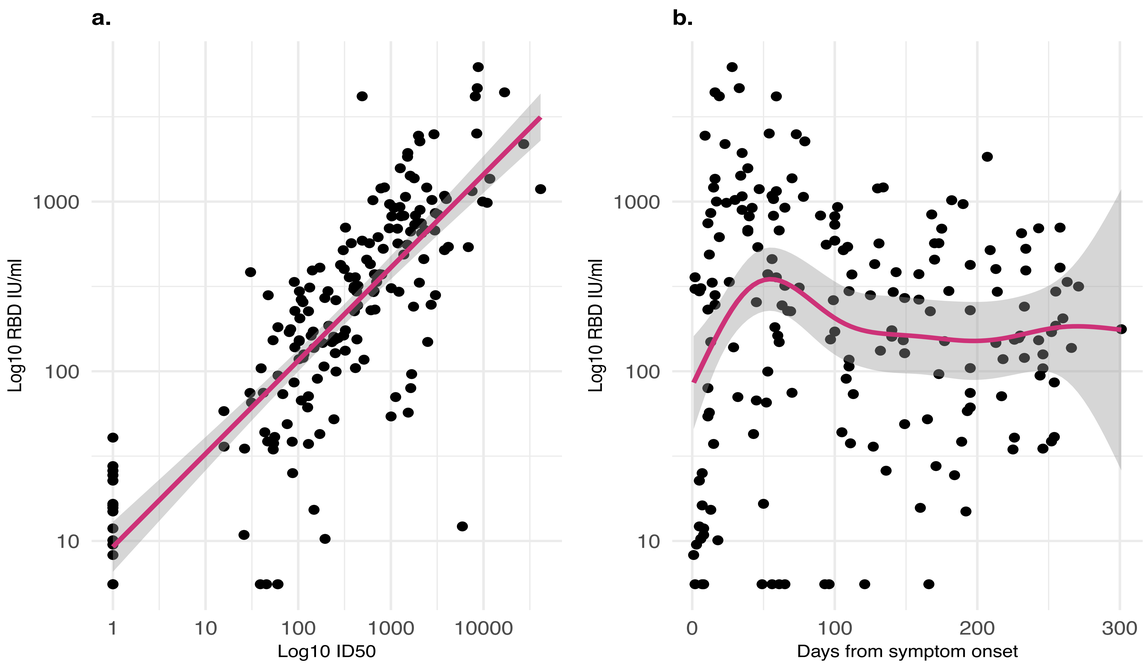
- Presence of SARS-CoV-2 neutralizing antibodies can provide protection against development of COVID-19[1,2].
- However, how reflective circulating anti-SARS-CoV-2 antibody levels are of underlying neutralizing capacity and whether a threshold exists to predict underlying sufficient neutralizing capacity remains unclear.

METHODS

- We measured anti SARS-CoV-2 IgG and neutralizing capacity in plasma from individuals with PCR-confirmed COVID-19 within the All Ireland Infectious Diseases Cohort Study.
- Binding IgG concentrations against RBD, Spike protein sub-unit 1 and 2 (S1, S2) and Nucleocapsid (NC) were measured using a multiplex electrochemiluminescence assay (IU/ml through normalization to World Health Organisation reference serum).
- Plasma neutralizing capacity was measured against live SARS-CoV-2 virus (Clinical isolate 2019-nCoV/Italy-INMI1) by determining the dilution of plasma that inhibited 50% of infection of Vero E6 cells, the 50% Neutralisation Titre (NT50), by flow cytometry-based micro-neutralisation assay.
- We estimated that a NT50 of 1:1000 against wild type SARS-CoV-2 would maintain neutralising activity against variants of concern, as the beta variant may reduce NT50 6 fold[3].
- Spearman correlation and linear regression was used to model relationships between neutralising capacity and IgG concentrations.
- Data are presented as median (IQR) unless specified.

RESULTS

- NT50 was highly correlated with RBD (Rho of 0.81 p <0.001), and less so with other IgG classes (S1 Rho of 0.8, S2 0.73, NC 0.72 all p<0.001)
- Median RBD IgG titre was 246 (71-662) IU/ml across all time points and trended lower over time, with an RBD IgG of 319 (61-1012) IU/ml at 0-90 days, 244 (86-523) IU/ml at 90-180 days and 157 (80-364) IU/ml at >180 days post symptom onset respectively (p=0.08)
- An RBD IgG concentration of 476 IU/ml corresponded to a NT50 of 1:1000.



- Sensitivity of RBD ≥476 IU/ml to predict NT50 1:1000 was 77 (95% CI 65-87)% and specificity 89 (95% CI 82-93)%.
- Restricting analysis to samples from convalescent subjects (>30 days post symptom onset, n = 148), sensitivity and specificity of RBD ≥476 IU/ml to predict NT50 ≥1:1000 increased to 88% (95% CI 74-96%) and 90% (95%CI 82-95%) respectively.

RESULTS

Patient characteristic (n = 190)	
Age	50 (40 - 64)
Female sex (n (%))	104 (55)
Time from symptom onset	98 (35 - 179)
Mild disease (n (%)	135 (71)
RBD IU/ml	246 (71-662)
S1 IU/ml	307 (85 – 770)
S2 IU/ml	165 (78 – 370)
NC IU/ml	65 (18 – 378)
NT50	323 (87 – 1483)

CONCLUSIONS

- In convalescent plasma, RBD IgG concentration ≥476IU/mL is sensitive and specific for predicting robust underlying neutralizing capacity.
- Further research is required to validate these findings in other cohorts and confirm these thresholds in vaccinated individuals.

REFERENCES

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