What we can learn from convalescent COVID 19 patients: antibody responses and development of monoclonal antibodies

ID Grand Rounds
June 3, 2020
9am
Johanna Daily
Kartik Chandran
Background

Ongoing study *Studying natural immune response to emerging viral infections* to identify convalescent patients to develop monoclonal antibodies

**POWASSAN VIRUS**

mAB development underway

Ryan Malonis (Lai Lab)

Kartik Chandran

Jon Lai
Studying natural immune response to SARS-CoV-2

Healthy Convalescent Donor

Development of mAb to SARS-CoV-2

Identify and screen donors
Pilot data: antibody responses
Develop pseudoviral system to test neutralization
Develop clinical Antibody test

Approach and pilot data
Which antibodies are protective in COVID-19?
  - Against which antigens
  - Is there a titer threshold associated with protection
  - Neutralizing versus non-neutralizing function
  - Isotype

What is the longevity of antibody protection?
Survey to pre-qualify healthy volunteers:
- a. +SARS-CoV2 PCR test
- b. Well x 2 weeks
- c. Healthy

Advertise study to find Convalescent subjects

Invited into study, venipuncture

N=1716 survey respondents
N=294

Collect PBMCs for mAb program

Analyze Ab titers to Spike protein

Referred to NYBC to donate N=190

March 15, 2020

Amanda Mengotto, Duncan Kincaid
Finding COVID-19 convalescent patients early in the NYC epidemic

outbreak in New Rochelle

COVID19 survivors became donors

Amanda Mengotto
Duncan Kincaid
David Rosenthal

Photos: Lynda Shenkman Curtis

Chaim Lebovitz  Joseph Helft  Jon Shuter
Clone, express the spike protein in large quantity

Expressed spike protein March 26, 2020.

Develop, optimize and validate serology

Develop, optimize and validate neutralizing Ab assay

Four Convalescent sampling sites

Kartik Chandran and Jon Lai laboratories
# Convalescent Donors

## Table 1. Baseline Characteristics

<table>
<thead>
<tr>
<th>Variable</th>
<th>Median</th>
<th>IQR</th>
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</thead>
<tbody>
<tr>
<td>Age (yrs)</td>
<td>41</td>
<td>32,52</td>
</tr>
<tr>
<td>Gender (% Female)</td>
<td>32</td>
<td>NA</td>
</tr>
<tr>
<td>Days since symptom onset</td>
<td>33</td>
<td>24,39</td>
</tr>
<tr>
<td>Days symptom free</td>
<td>15</td>
<td>2,22</td>
</tr>
</tbody>
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N=281
Viral targets of antibody responses

Astuti et al. Diabetes & Metabolic Syndrome: Clinical Research & Reviews 2020
Nucleocapsid prt

Membrane prt

Spike

ssRNA

Binds to ACE2

Small Envelope prt

Serology tests based on different targets

<table>
<thead>
<tr>
<th>Chandran/Lai</th>
<th>Abbot</th>
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<tbody>
<tr>
<td>Spike protein</td>
<td>N protein</td>
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</table>

Astuti et al Diabetes & Metabolic Syndrome: Clinical Research & Reviews 2020
Total IgG (spike Protein)

- Number of values: 281
- Minimum: 90
- 25% Percentile: 4,860
- Median: 7,290
- 75% Percentile: 21,870
- Maximum: 590,038

Sent for Plasma Donation for RCT
Antibody titer not related to gender or days since symptomatic

P=0.777, Mann Whitney
Median, IQR

Days since symptom onset and IgG titer correlations

R= -0.167

P=0.0121, Pearson
CP neutralization using a pseudovirus system

Clone Spike protein into vesicular stomatitis virus (can be handled in BSL2), with a fluorescence protein (GFP)

1. CP inhibits invasion and viral growth in Vero Cells
2. Magnitude differs by donor

M. Eugenia Dieterle, in revision, Mol Cell Bio
High neutralizing titer in donors CP
SARS-CoV2 persistence in nasopharyngeal swabs in Convalescent patients

Median days from onset of symptoms to time of NP swab: 28 Days

No difference in age, or time from onset of symptoms and a positive test
SARS-CoV2 persistence in nasopharyngeal swabs in Convalescent patients

- Virus shedding patterns in nasopharyngeal and fecal specimens of COVID-19 patients (Gong et al)
  - Median duration of virus shedding was 10.0 days (IQR 8.0 to 17.0) in the upper respiratory swabs
  - N=66

- Single cases positive up to 6 months in other reports

Swabs sent to Army Dr. John Dyer
To determine if they are viable in culture (BSL3)
Donors referred to NYBC

Each unit is 200mls

NYBC: can donate 3 times

Dr. Chris Hillyer
Susan Kusic
Monika Paroder
Leana Serrano-Rahman

Montefiore Medical Hospitals Blood Banks

Expanded Access Program

Randomized Controlled Trial
Do antibody responses (titer, function, isotype) correlate with protection? Studies ongoing.
Duration of Anti-body responses: SARS (2002)

- Ab diminish over time (typical of coronaviral response 1-2yrs)

Symptomatic n=23

Tang et al JI 2011

Inactivated virus

6 yr study
Ab diminish over time (typical of coronaviral response-1-2yrs)

Symptomatic n=23

Duration of Anti-body responses: SARS (2002)

- Inactivated virus

SARS-CoV pneumonia, n=20

Nucleocapsid protein

Tang et al JI 2011

Woo et al Clin Diag Lab Imm 2004
Antibody changes over time:
compare screening antibody to plasma unit antibody titer

Measure antibody from Plasma segment (20% citrate)

16 days median
Antibody changes over time:
compare screening antibody to plasma unit antibody titer

- 25% Percentile: Screen 7290, Donation 7290
- Median: Screen 21870, Donation 21870
- 75% Percentile: Screen 65610, Donation 196829

Screening Plasma donation

16 days median

Measure antibody from Plasma segment (20% citrate)
Longitudinal study: CP donors

1st visit is follow up for out patient cohort

Complete the medical history

Case report form (symptoms)
Blood draw

60d 90d 120d 270d 340d 550d 750d

ongoing

Can contact us if illness in between visits
Studying natural immune response to SARS-CoV2

Healthy Convalescent Donor

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