Production of Anti-Coronavirus Disease 2019 Hyperimmune Globulin from COVID-19 Convalescent Plasma

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Grifols, Since 1909

Headquartered in Barcelona, Grifols is a global healthcare company with more than 110 years of legacy dedicated to improving the health and well-being of people around the world

Grifols has over 24,000 employees located in 30 countries with commercial sales in over 100 countries

Operates under four specialized divisions

- Biosciences – Diagnostics – Hospital – Bio Supplies

Divisions within Grifols Involved with COVID-19 Initiatives



Bioscience

Producer of essential plasmaderived therapies worldwide

- Large network of plasma donation centers
- Manufacturing capacity of more than 15 million liters of plasma per year
- Leading producer of human IgG therapies (Gamunex-C, Flebogamma DIF, Xembify, GamaSTAN, HyperRAB)



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- Comprehensive portfolio for disease detection and managing laboratory operations
- Specialized diagnostics for detection and treatment monitoring for infectious, autoimmune, and neurodegenerative diseases



Diagnostic

Transfusion medicine from donation to transfusion

Convalescent Plasma

The first antibody-based therapy



Convalescent Plasma for Treating Infectious Diseases

 The passive transfer of convalescent plasma, collected from patients who have recovered from an infectious disease, is the fastest antibody-based path to provide a breath of antibodies that can recognize and neutralize the pathogen as well as other components that may contribute to an immune response



Image Credit: Kateryna Kon



Antibody Neutralization

Convalescent Plasma Has a Long History

- Earliest treatments were for diphtheria and tetanus in 1891 (Emil von Behring, the founder of serum therapy) and the influenza pandemic (Spanish flu) of 1918
- More recent treatments include severe acute respiratory syndrome (SARS), influenza A (H1N1), avian influenza A (H5N1), and several hemorrhagic fevers (Hantaan, Junin, Ebola)



- Early into the COVID-19 pandemic, many clinicians, epidemiologists, and researchers were investigating the COVID-19 convalescent plasma (CCP) with variable results
- Convincing results achieved with an early symptom onset, high-titer (≥12 in the Ortho VITROS IgG) study in older adults (R. Libster et al. N Engl J Med, Jan 6, 2021)
- Risk/benefit analysis demonstrated the benefit of CCP for the treatment of COVID-19 and on August 23, 2020, the FDA issued an Emergency Use Authorization for the use of CCP for the treatment of hospitalized patients

CCP Antibody Titers to SARS-CoV-2 - Donor-to-Donor Variability Ortho Vitros IgG (Spike Protein, S1) vs Abbott Architect (Nucleocapsid Protein)



Architect S/CO

- While a correlation exists in antibody titer to nucleocapsid protein and spike protein, there is a high variability from donor to donor
- A relatively small fraction (<15%) of CCP considered high antibody titer (≥12 in the Ortho VITROS IgG)
- A significant number of donations have an antibody titer at or below S/CO thresholds
- To best utilize CCP in treatment of COVID-19, we need a consistent plasma source that maximizes the use of all neutralizing antibodies

A Potentially Improved CCP Product for Treating COVID-19

- A high potency, well-defined IgG product manufactured from a consistent source of CCP
 - Polyvalent and highly diverse monomeric IgG as it is comprised of many convalescent donors
 - Defined product with greater consistency in antibody neutralization
 - **o** A well established safety and tolerability profile
 - Higher antibody neutralization capacity per unit volume
 - Reduced risk of adverse events
 - No blood type matching required
 - o Better pathogen safety profile with dedicated steps with high virus clearance capacity
 - \circ Improved product storage (2 8 °C vs -30 °C)
 - Potential source of neutralizing antibodies that evolves as the virus mutates



Creating an Anti-COVID-19 Hyper Immunoglobulin

A partnership with the Biomedical Advanced Research and Development Authority (BARDA) and the United States Department of Defense (DoD)



Plasma Collection and Screening Millions of potential donors, but with many challenges



Early Challenges with CCP Collection

Donor Criteria and Internal Systems

- Establishing criteria for donor selection
 - Symptomatic vs asymptomatic donors?
 - Any test vs EUA approved tests?
 - Molecular vs antigen test?
 - Time between symptom resolution and donation
- Verification of test results
- Creating systems to manage results
- Regulatory approvals
- Staff training and education

Donor Engagement

- Public awareness of a critical need for CCP
- Managing fear (CCP and non-CCP donors)
- Education about plasma donation (destigmatized)
- Managing misinformation from social media

Donor Recruitment During a Pandemic

Elevating plasma awareness and promoting plasma donations



- Campaign: The "Give Your Light" campaign used for greater donor engagement and recruitment
- Marketing: Posters, personal stories, mailings, Facebook, Instagram, Google Ads
- External Communications: Press release, radio station interviews.....



- "The Fight Is In Us" campaign encourages plasma donations from those recovered from COVID-19 in the U.S
- Supported by a coalition that includes plasma manufacturers, medical and research organizations, blood centers, philanthropic organizations and COVID-19 survivor groups

Grifols' Extensive Network of Donation Centers



- Grifols operates over 260 FDA and EU approved blood plasma donor centers under the names of Biomat USA Inc., BPC Plasma Inc., Interstate Blood Bank Inc (IBBI), and GCAM Inc.
- Plasma tested in San Marcos and Austin, Texas, with a plasma-testing capacity of over 15 million samples per year
- Centers and their donors are considered part of the "critical infrastructure industry" according to the President's Coronavirus Guidelines for America

GRIFOLS

*Includes whole blood donation centers

CCP Donor Criteria and Plasma Testing

Donor Criteria

Laboratory evidence of COVID-19 infection (NAT), positive antigen test, or by SARS-CoV-2 antibody test prior to enrollment

Must meet criteria to ensure they are **no longer infective**, including:

- Symptomatic donors must have resolution of symptoms at least 14 days before donation if negative by follow-up NAT, or 28 days without follow-up test
- Asymptomatic donors must wait 14 days after the initial positive test if negative by follow-up NAT, or 28 days without follow-up test

PLASMA COLLECTION

DONOR RECOVERED FROM COVID-19







CONVALESCENT PLASMA

Plasma Testing

Plasma collected **by plasmapheresis**, meeting all requirements for **source plasma** which includes stringent donor screening for a number of common viruses, now including SARS-CoV-2

Additional Requirements

- Donors are negative for anti-HLA antibodies
- All donations tested by NAT (Procleix SARS-CoV-2 assay) to confirm absence of SARS-CoV-2 (virus)
- Each donation verified to have antibodies to SARS-CoV-2 at or above an established cut-point, critical to maintain antibody titer with batch-to-batch consistency

Quickly Addressing the Need to Screen Plasma for SARS-CoV-2

Grifols' Procleix SARS-CoV-2 Assay on the Panther System 2020

- Procleix nucleic acid technology (NAT) used to develop test platform for screening plasma and whole-blood donations, CE-marked on 6 May 2020
- Designed for specific detection of SARS-CoV-2 virus
 - To confirm absence of SARS- CoV-2 RNA in plasma donations (for transfusion or for manufacture)
 - Detection of SARS-CoV-2 RNA in nasal, nasopharyngeal (NP) and oropharyngeal (OP) swab specimens





Manufacturing an Anti-COVID-19 Hyper Immunoglobulin Leveraging the Gamunex®-C Process and Multi-Purpose Facility



Manufacturing Process Based on Gamunex®-C

Gamunex®-C, an approved 10% immune globulin injection [human] with long, well established safety and efficacy profile



- Purified IgG is concentrated to 10% (100 mg/mL), formulated in an isotonic glycine solution, nanofiltered, and sterile filtered into a bulk bag
- The drug substance is filled into 100 mL Gri-bag using terminal sterile filtration



MANUFACTURING

Anion chromatography for protein purification (reduce IgM and IgA), generating an immunoglobulin material of **not less than 98% IgG**

High virus clearance capacity with dedicate and validated virus inactivation and/or removal steps

- Caprylate precipitation / depth filtration
- Caprylate incubation
- Nanofiltration
- Low pH incubation

Grifols' Response to Global Pandemic in Record Time

Specialized clinical-scale, isolated facility dedicated to the manufacture of hyper immunoglobulins against emerging infections



MPF (Multi-Purpose-Facility), initially designed to manufacture hyper immunoglobulin for the Western African Ebola virus epidemic (2013 – 2016), allowed Grifols to manufacture **anti-COVID-19 hyper immunoglobulin in less than 3 months**

- Multipurpose facility (MPF) Located in Clayton, North Carolina
- Operated by small, highly trained team
- Validated facility operates independently from systems for licensed products
- Specialized systems for handling solid and liquid waste
- Integrated QC lab for in-process & environmental monitoring testing

Consistency of the CCP Manufacturing Plasma Pools (N=17)

Antibody titer criteria of CCP helps create a consistent plasma pool, while maximizing utilization of CCP



Ortho VITROS (IgG)

Manufacturing Plasma Pool Series

Range	6.0 – 9.2
Mean	7.0
RSD	13%



Alpha Diagnostic (IgG) Spike Antigen

Manufacturing Plasma Pool Series

Range	3024 - 6741	
Mean	4831	
RSD	21%	GRIFOLS

Serologic Titers of Anti-COVID-19 hIVIG Product (Alpha ELISA)



IgG Antibody Titer in Pool and Product

- Mean **pool** antibody titer (1:X) at 4831
- Mean product antibody titer (1:X) at 78936
- Anti-SARS-CoV-2 product shows ~ 15x increase in serologic titer to SARS-CoV-2 (spike antigen), corresponding to the purification/concentration of IgG

Antibody Neutralization with Anti-COVID-19 hIVIG Product (N=17)

Antibody neutralization testing performed at the National Institutes of Health (NIH) Integrated Research Facility (IRF), Frederick, MD



Antibody Neutralization (Potency) with

Final Product Series

Mean IC ₅₀	486
Range	327 - 759
RSD	22%

- Fluorescent microneutralization method quantifies the anti-SARS-CoV-2 neutralization titer by inhibition of infection of cultured Vero (CCL-81) cells by SARS-CoV-2 (Washington isolate, CDC)
- Data are reported based on a 4-parameter regression curve (using a constrained fit) as a 50% neutralization titer (IC₅₀)

Grifols' Anti-COVID-19 Hyper Immunoglobulin



Anti-COVID-19 hIVIG meets all licensed IVIG (Gamunex-C) specification, with additional criteria for antibody titer to SARS-CoV-2

- Ready-to-use sterile, preservative-free solution filled in a 100 mL Gri-bag
- Consists of 9%–11% protein in 0.16–0.24 M glycine, pH 4.0 4.3
- Anti-SARS-CoV-2 IgG titer enrichment of ~15X (consistent with antibody concentration) with established antibody neutralization capacity
- Contains not less than 98% IgG
- IgG subclass distribution similar to that found in normal human serum
- Anti-A and anti-B levels ≤1:64
- Trace levels of IgM and IgA
- Storage at 5±3°C

Clinical Study with Anti-COVID-19 hIVIG

Unprecedent Times Bring Unprecedent Collaboration



Inpatient Treatment With Anti-Coronavirus Immunoglobulin (ITAC)

An International Multicenter, Adaptive, Randomized Double-Blind, Placebo-Controlled Trial of the Safety, Tolerability and Efficacy of Anti-Coronavirus Hyperimmune Intravenous Immunoglobulin for the Treatment of Adult Hospitalized Patients at Onset of Clinical Progression of COVID-19

Sponsor and Collaborators

- University of Minnesota
- National Institute of Allergy and Infectious Diseases (NIAID), part of the National Institutes of Health (NIH)
- The International Network for Strategic Initiatives in Global HIV Trials (INSIGHT)
- Biomedical Advanced Research Development Authority (BARDA)
- The Food and Drug Administration (FDA)

Investigators

- Principal Investigator James Neaton, PhD, University of Minnesota
- Study Chair Mark Polizzotto, MD, The Kirby Institute, University of New South Wales
- 58+ International Clinical Sites

ITAC Study Design (ClinicalTrials.gov Identifier: NCT04546581)

The study seeks to determine whether the administration of anti-COVID-19 hIVIG at the onset of symptoms can increase the response of patients' immune systems against the virus, reducing the progression of the disease and the risk of mortality



hIVIG dose is a single infusion at 400 mg/kg (capped at 100 kg)

Primary endpoint based on the patient's clinical status on Day 7 which includes 7 mutually exclusive categories capturing a range of organ dysfunction

Inclusion Criteria

- Documented infection by NAT, symptomatic COVID-19 disease
- Duration of symptoms attributable to COVID-19 ≤ 12 days

Study start date Oct 8, 2020

Four Companies – One Goal

ITAC includes anti-COVID-19 hIVIG products manufactured by four Companies









- Each Company manufactures a commercial 10% product from CCP, meeting all compendial specifications for an IVIG
- Each Company has a long, demonstrated history of safety and efficacy with their 10% IVIG products
- All Anti-COVID-19 hIVIGs are manufactured using a validated and approved process
- The most relevant attribute for treating COVID-19 patient, antibody neutralization (potency) of SARS-COV-2, is measured for each product by the same lab with the same method (NIAID)

It begins with a donors' generosity



Every person that donates is a hero because you are able to impact someone's life

Thank you!

