STUDY TITLE: A PHASE 2 TRIAL OF INFLIXIMAB IN CORONAVIRUS DISEASE 2019 (COVID-19).

STUDY SPONSOR: Tufts Medical Center

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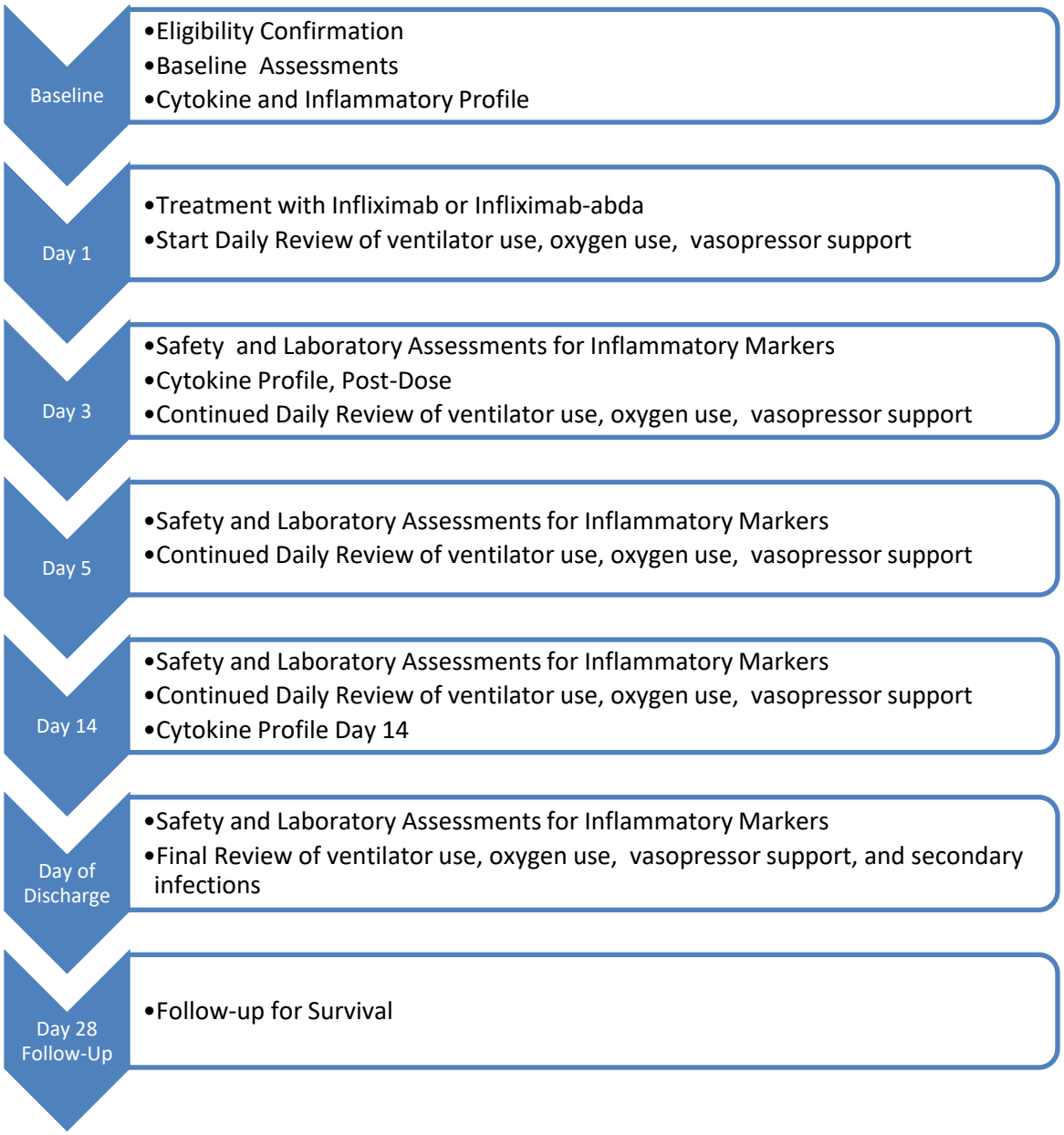
A. Study Schema



B. Introduction

B.1 Background, Rationale and Therapeutics

On March 30 2020, there were over 720,000 confirmed cases of SARS-COV2 infection (COVID-19) globally and 33,000 deaths, with a mortality rate that is10-fold higher than that of influenza.[1] In the United States, COVID-19 cases and deaths from COVID-19 are accelerating rapidly across the nation. Twenty percent of COVID-19 patients present with severe illness, defined by presence of dyspnea, respiratory rate ≥30/min, blood oxygen saturation ≤93%, partial pressure of arterial oxygen to fraction of inspired oxygen ratio (PaO2/FiO2) <300, and/or lung infiltrates >50% within 24 to 48 hours. Approximately 50% of admitted patients will require intensive care monitoring for cardiorespiratory support. Among those who are admitted to the intensive care unit and require intubation, the mortality rate approaches 85%. Death from cardiorespiratory

failure is principally due to acute respiratory distress syndrome (ARDS) and myocardial depression or cardiogenic shock.[2] With the COVID-19 pandemic threatening to overwhelm the US healthcare system, there is a dire need for treatment to mitigate disease severity and death and also to preserve and ration medical equipment and intensive care unit (ICU)-level interventions, including ventilator requirements.[3] Lack of ICU access and ventilator shortages alone threaten to contribute significantly to the excess mortality from COVID-19 disease.

Course of the Lethal Illness and the Role of Cytokine Storm.

In both inpatient and ambulatory settings, the typical clinical course includes stable or improving symptoms over the first 7 to 10 days after symptom onset. Beyond this time frame, some patients have a sudden, fulminant clinical decompensation requiring emergent intubation and ICU care.[4, 5] This rapid deterioration with progressive hypoxemia, myocardial depression, and multiorgan failure is thought to be secondary to cytokine storm, rather than direct viral damage to pneumocytes. Early intervention to prevent or mitigate this hyperinflammatory cytokine response may markedly reduce the need for ICU level of care, ventilator support, and vasopressors usage and improve survival rates in COVID-19 patients.[6]

Pathophysiology of the Cytokine Storm in COVID-19

Strong parallels have been noted between severe COVID-19 and hyperinflammatory states, such as cytokine storm and secondary hemophagocytic lymphohistiocytosis (sHLH). These illnesses are characterized by aberrant and exorbitant immune responses, which can be triggered and amplified by viral infections interacting with incompletely determined host factors. A hallmark of the illness is the influx of tissue macrophages that generate prodigious amounts of cytokines that suppress cytotoxic T-lymphocytes and NK cell function and inflict a diverse array of tissue damage.[7] Severe COVID-19-related multi-organ dysfunction is associated with a cytokine profile characterized by increased levels of tumor necrosis factor-alpha (TNFα), interleukin-2 (IL-2), interleukin (IL-7), interleukin-10 (IL-10), granulocyte colony stimulating factors (GCSF), interferon-gamma inducible protein (IP10) and macrophage inflammatory protein 1-a (MIP1A).[2] The cytokine profile of COVID-19 patients admitted to the ICU showed higher levels of TNFα, IL-2, IL-7, and IL-10 than non-ICU patients, correlating with disease severity.[2] In addition, CRP, IL-6 and ferritin levels were found to be 2-4 fold higher in patients who died of COVID-19 relative to those who recovered.[8] Moreover, flow cytometry analysis of peripheral blood from 28 patients with COVID-19 identified a unique monocyte population that expressed macrophage markers and secreted TNFα, IL-6 and IL-10. This monocyte population, otherwise absent in healthy controls, had readily detectable morphological and inflammation-related phenotypic changes, which were more pronounced in patients requiring prolonged hospitalization and ICU admission.[9] The aberrant and excess inflammatory activation appears to be detrimental rather than protective in COVID-19 patients. The hyperinflammatory state that is noted in COVID-19 patients resembles virally-triggered secondary HLH (Figure 1), which can also result in a fulminant life-threatening course with overlapping patterns of organ failure.[10]

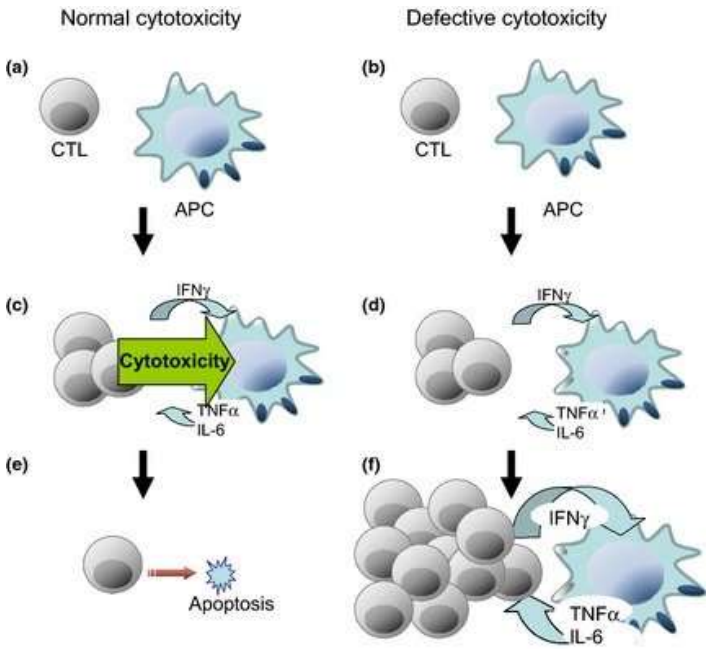


Figure 1: Pathophysiological model of the cytokine storm for COVID-19/HLH: NK and CTLs fail to eliminate activated macrophages, Activated macrophages continue to secrete excessive amount of cytokines and feed back to cause CTL proliferation, Excessive cytokine production (cytokine storm) by macrophages, NK cells, and CTLs resulting in tissue damage and multi-organ failure. [Adapted from Rosee et al, Blood 2019]

Active therapeutic trials for the Cytokine Storm of COVID-19 and their rationale:

A number of anti-viral drugs that inhibit viral replication are being investigated as treatment for COVID-19.[11] However, tissue injury in severe COVID-19 appears to be principally mediated by pro-inflammatory cytokines[6], which suggests that blocking cytokine activity at the cellular level is a valid and potent strategy to blunt the magnitude of damage from the viral illness. Given the diversity of cytokine elevation associated with the illness, identifying a driver or master-regulator cytokine to target would be essential to this approach. Cytokine directed therapies have been used for the treatment of cytokine storms of diverse origins, including IL-6, IL-1b and TNFα inhibitors.[12-14] Tocilizumab, an IL-6 inhibitor, has shown efficacy when used for cytokine release syndrome that occurs after CAR-T administration.[12] Use of tocilizumab for managing cytokine storm related to COVID-19 is now gaining mainstream acceptance after preliminary data from a study of 21 patients with severe or critical COVID-19 infection showed rapid defervescence and improved oxygenation after administration of a single dose of tocilizumab.[15] Current clinical guidance on management of severe COVID-19 infections recommends against the use of corticosteroids, unless indicated for another reason, and reports insufficient evidence to issue any recommendation on the use of any of the following: antiviral agents, recombinant interferons, chloroquine/hydroxychloroquine, or tocilizumab.[16] Clinical trials (NCT04317092, NCT04306705, NCT04320615) are currently ongoing to demonstrate the efficacy of IL-6 and IL-1beta antagonism in COVID-19.[17] In secondary HLH, TNFα-antagonists have demonstrated striking efficacy after failure of frontline steroids and etoposide-based chemotherapy.[14]

TNFα as a master regulator of the cytokine storm of COVID-19

There are diverse lines of evidence that implicate TNFα as a master regulator of the cytokine storm in COVID-19 and secondary HLH. TNFα is a cytokine produced by mononuclear cells, predominantly macrophages/monocytes and plays an integral role in the inflammatory cascade associated with infections and autoimmune diseases (Figure

2).[18] Its inflammatory and immuno-regulatory actions are mediated by its binding to TNF receptor-1 and TNF receptor-2. TNFα is thought to trigger a febrile reaction by a mechanism that involves release of IL-6 and is independent of IL-1β.[19] Fever is the most common symptom of COVID-19 infection[2] which suggests that TNFα may be an early mediator in the pathological cascade of the illness. TNFα levels have been found to be significantly higher in both ICU and non-ICU patients relative to healthy controls, whereas IL-6 levels were only elevated in ICU patients (Figure 3).[2] These findings suggest that TNFα levels may rise earlier in the disease course and play a critical role in the early pathogenesis of severe illness. Additionally, TNFα was found to up-regulate the production of ferritin in acute phase state human hepatocyte cultures, as opposed to IL-6 and interleukin-lα that did not.[20] As with HLH, ferritin elevation was noted in almost 80% of COVID-19 cases and hyperferritinemia was more common in patients who died from the infection.[2] Elevated ferritin levels have been identified as a predictor of mortality in COVID-19 and thus, indirectly linking TNFα with adverse disease outcomes.[2, 21] The role of TNFα in the pathogenesis of autoimmune disease such as rheumatoid arthritis (RA) is clear. In a disease model that represents RA, anti-TNF agents were shown to inhibit the production of several important pro-inflammatory cytokine such as IL-1, IL-6 and GM-CSF from the synovial culture.[22] Anti-TNF therapies have since become the mainstay of disease modifying therapies for RA.

Figure 2

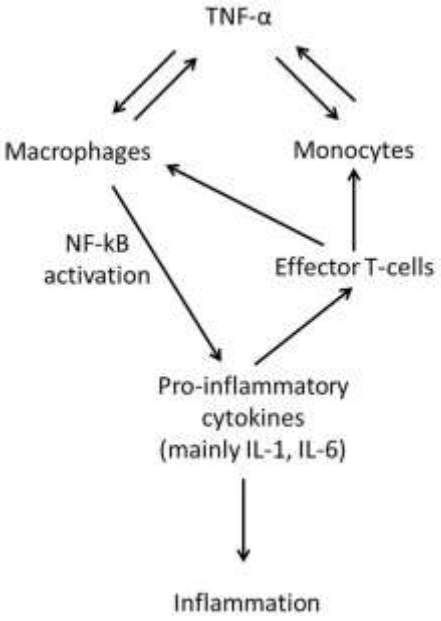
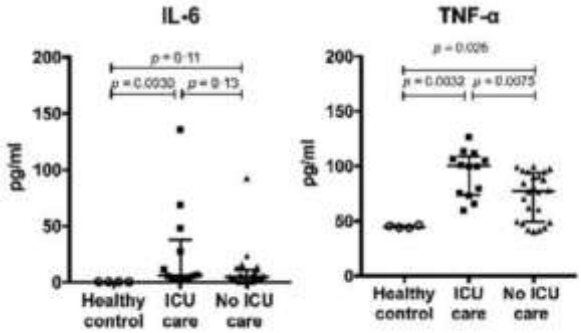


Figure 3



The detrimental role of excess TNFα has been established in influenza and severe acute respiratory syndrome (SARS)-Coronavirus (CoV). The levels of pro-inflammatory cytokines (IL-6, TNFα) in patients with influenza correlate with both the severity and duration of symptoms.[23, 24] Furthermore, single-nucleotide polymorphisms (SNPs) in the TNF gene have been associated with severity of influenza.[25] In preclinical mouse models of influenza, etanercept (which binds to circulating TNFα) used in H1N1 infected mice decreased inflammatory cytokines, including TNFα, IL-6 and IFN-gamma, reduced inflammatory cell infiltration including macrophages and neutrophils, reduced viral replication, and improved survival.[26] The improved virological outcomes suggest restoration of physiological immune surveillance, specifically cytotoxic T cell and NK-function. Similarly, early upregulation of key inflammatory mediators, including TNFα and

IL-6, in SARS-CoV infected mice was shown to promote lethal disease with all responses being significantly more robust early during infection in the lethal isolates.[27] This upregulation in TNFα, as well as IL-6, was also noted in SARS patients and found to be sustained for every length of the clinical courses investigated when compared to healthy controls.[28, 29] Infection of St. Jude porcine lung (SJPL) epithelial cells with different respiratory viruses significantly increased TNFα expression, indicating a role for TNFα in the pathogenesis of virus inflicted cellular injury.[30] These results indicate that viral lung damage is at least in part mediated by TNFα action and TNFα depletion via use of TNFα inhibitors may potentially be protective against this direct damage as well.[26, 31]

ARDS is characterized by diffuse alveolar damage (DAD) and lung capillary endothelial injury, and overdrive in the inflammatory response led by inflammatory cytokines has been implicated in its evolution. High TNFα levels have been detected in serum and bronchoalveolar lavage fluid of patients with ARDS and correlate with severity of lung damage.[32, 33] Post-mortem analysis of patients who died from COVID-19 shed light on the diffuse alveolar damage, cytopathic changes in the pneumocytes and presence of mononuclear inflammatory infiltrates, dominated by lymphocytes.[34, 35] Immune profiling by flow cytometry depicted a higher than expected fraction of pro-inflammatory CCR6+ Th17 cells in the blood.[35] Th17 cells produce cytokines, including interleukin (IL)-17, IL-6, IL-21, IL-22 and TNFα and have been identified as a pathogenic factor in RA. Use of TNFα inhibitors has been shown to suppress their activity and potentially control the inflammatory cascade.[36] As ARDS can develop in up to one-third of COVID-19 cases and high TNFα levels have been seen in these patients, the role of TNFα inhibitors to protect from lung injury should be further contemplated. Cardiac decompensation is another hallmark of COVID-19 with pre-existing hypertension and cardiovascular disease accounting for the highest risk of death.[2, 8] TNFα has been associated with myocardial depression and evolution of viral myocarditis and taken together may be implicated in the cardiac pathology of lethal COVID-19.[37, 38]

Taken together, the preclinical and clinical data strongly support the rationale for investigating the efficacy and safety of anti-TNFα therapy in patients at risk for rapid cardiorespiratory decompensation and early mortality in severe COVID-19 infections. Other groups have echoed the need for clinical trials to study this approach.[39] Fulfilling this need is urgent in the setting of a rapidly accelerating global pandemic with restricted availability of effective systemic therapies and access to critical care support.

We **hypothesize** that early institution of TNFα inhibitor therapy in patients with severe COVID-19 infections will prevent further clinical deterioration and reduce the need for advanced cardiorespiratory support and early mortality. To address this hypothesis, a prospective, single center, phase 2 trial is proposed to assess the efficacy of infliximab or infliximab-abda in hospitalized adult patients with severe or critical COVID-19. Observations from this study will inform the conduct of prospective randomized controlled studies to follow.

B.1.1 Therapeutic Agents – Infliximab and Infliximab-abda

Infliximab and Infliximab-abda are TNFα inhibitors currently FDA-approved for the treatment of autoimmune disorders, including Crohn's disease and rheumatoid arthritis. The risks and adverse reactions are described in the approved prescribing

information for infliximab (or infliximab-abda). Either infliximab or infliximab-abda may be used, at the discretion of the investigator. For the full prescribing information for infliximab, please click here. Full prescribing information for infliximab-abda can be found here.

Treatment with infliximab or infliximab-abda 5mg/kg IV should ideally be administered within 6 hours of enrollment, and no more than 24 hours following enrollment. Pre-medication with Tylenol 650 mg once 30 minutes prior to infusion would be recommended. Other pre-medications may be given at the discretion of the treating physician. These include diphenhydramine 50mg by mouth, as well as prednisone 20mg by mouth, both given 30 minutes prior to infusion. Pulse and blood pressure should be monitored every 30 minutes during the infusion, and patients should be monitored for at least 30 minutes following the infusion.

Patients will be monitored for safety through the infusions of infliximab or infliximab-abda. Common adverse drug reactions include infections and infusion related reactions. Signs of infusion related reactions include chest pain, chills, dyspnea, fever, hypertension, hypotension, pruritis, urticarial, abdominal pain, coughing, fatigue, headaches, nausea, and upper respiratory tract infections. In the event any of these occur during or following the infusion, the relation to infliximab or infliximab-abda will be determined by the investigator.

In the event of a hypersensitivity reaction, the infusion should be stopped and the treating physician should be alerted. For mild hypersensitivity reactions (localized hives, itching, warmth), diphenhydramine 25-50mg IV or orally and famotidine 20mg IV should be administered. For moderate hypersensitivity reactions (generalized hives or itching, flushing, diffuse erythema, or mild hypotension), hydrocortisone 100mg should be administered in addition to diphenhydramine 50mg IV or orally and famotidine 20mg IV. For severe reactions (angioedema, respiratory failure, hypotension), 0.3 mg of epinephrine intra-muscular injection should be given in addition to the mild and moderate reaction regimens listed above.

Retreatment with infliximab is permitted at treating physician discretion 7-21 days following primary therapy and based on initial response; the usual treatment schedule is every 2 weeks, this interval is not strictly enforced given the uncertainty of outcomes with primary therapy.

B.2 Risks to Subjects and Study Staff

Infliximab (or Infliximab-abda) is currently FDA-approved for the treatment of a number of autoimmune disorders, including Crohn's disease and rheumatoid arthritis. The risks and adverse reactions are described in the approved prescribing information for infliximab (or infliximab-abda). In addition to the possible side effects and toxicities associated with the use of infliximab (or infliximab-abda), the risks of this study include possible unique and unknown adverse events or complications related to the use of a TNFα inhibitor in the setting of COVID-19, a potential for worsening or accelerated disease progression and death, and possible exclusion from receiving other immunosuppressive or biologic agents or from enrollment in other trials evaluating other therapeutic options. However, reporting to date has indicated that cytokine-directed therapies have not resulted in disease worsening in

COVID-19 and TNF-directed therapies in bacterial sepsis have not resulted in adverse outcomes.

Infliximab may enhance the adverse/toxic effect of live vaccines and diminish the therapeutic effect of both live and inactivated vaccines. Live-organism and live-attenuated vaccines should not be given concurrently with infliximab. Live-attenuated vaccines should not be given for at least 3 months after infliximab. In the absence of any other contraindications, vaccinations per standard of care can be given after at least 3 months from the last dose of infliximab which also should include a COVID-19 vaccine in the event that becomes available.
Study investigators and staff may be at risk for contracting COVID-19 while performing study procedures. However, Tufts Medical Center has implemented numerous universal precautions to prevent the spread of COVID-19, including reducing the number of staff on-site, required use of facemasks at all times, and use of additional PPE when interacting with COVID-19 patients. Remote consenting processes have obviated the need for in-person interactions by study personnel.

B.3 Potential Benefits to Subjects

Potential direct benefits to an enrolled subject may include a shorter time to improvement in oxygenation and breathing, a shorter recovery time, decreasing the need for intensive care or ventilator support, and decreased mortality. Possible benefits to others include learning if the study drug is safe and effective in the treatment of COVID-19 and which subsets of patients with COVID-19 as defined by cytokine profile, benefit maximally from the study drug. Furthermore, If the trial meets its endpoints and TNFα inhibitor therapy is proven effective in severe cases of COVID-19, it would add a critically required therapeutic option, especially in the setting of limited resources, and restricted availability and expected shortages in other agents used such as remdesivir and IL-6 inhibitors. Repurposing an already approved and well-studied drug for a new indication is an efficient and attractive alternative in the setting of an outbreak of this magnitude. Given the shortage in critical care resources that will alone account for significant excess mortality, effective sparing of ventilator therapy for one patient may prove life-saving for two individuals rather than just one.

B.4 Alternatives

Patients may choose not to participate in this research study. There is currently no FDA approved treatment available for COVID-19. Alternative treatment options may include supportive care alone, palliative care, enrollment in other clinical trials or off-label treatment with one of the agents that are part of the Tufts Medical Center COVID-19 treatment guidelines. .

C. Objectives

Primary endpoint:

1. Time to improvement in oxygenation (increase in SpO2/FiO2 of 50 or greater compared to the baseline SpO2/FiO2) sustained for a minimum of 48 hours

Secondary endpoints:

1. 28-day mortality

2. Assessment of cytokine and inflammatory profile at baseline and at 48 hours after therapy (TNFα, IL-1b, IL-2, sIL-2r, IL-6, ferritin, ESR, CRP, CPK, troponin)
3. Qualitative and quantitative toxicity
4. Incidence and duration of supplemental oxygen administration by nasal cannula, simple face mask, or other similar oxygen delivery device
5. Incidence and duration of non-invasive ventilation or by non-rebreather mask or high-flow nasal cannula
6. Incidence and duration of mechanical ventilation
7. Incidence and duration of vasopressor support
8. Incidence and duration of extracorporeal membrane oxygenation (ECMO)
9. Duration of fever
10. Correlation of cytokine profile to clinical outcomes specified in primary and secondary objectives
11. Duration of hospitalization
12. Secondary infections

D. Enrollment and Withdrawal

D.1 Inclusion Criteria

Patients must meet <u>all</u> the following criteria to be eligible:

1. Age 18 years or older
2. Able to provide informed consent
3. Hospitalized adult patients with pneumonia evidenced by chest X-ray or CT scan
4. Laboratory (RT-PCR) confirmed infection with 2019-nCoV or strongly suspected to be infected with SARS-COV2 with confirmation studies pending
5. And at least one of the following:
   a) Respiratory frequency ≥30/min
   b) Blood oxygen saturation ≤93% on RA
   c) Partial pressure of arterial oxygen to fraction of inspired oxygen ratio (PaO2/FiO2) <300
   d) Worsening of lung involvement, defined as an increase in number and/or extension of pulmonary areas of consolidation, need for increased FiO2 to maintain stable O2 saturation, or worsening O2 saturation of >3% with stable FiO2

D.2 Exclusion Criteria

Patients who meet any of the following criteria will be excluded from the trial:

1. Treatment with any TNFα inhibitor in the past 30 days
2. Known hypersensitivity to any TNFα inhibitor, murine proteins, or any component of the formulation

3. Presence of any of the following abnormal laboratory values at screening: absolute neutrophil count (ANC) less than 1000 mm3, hemoglobin <7.0g/L, platelets <50,000 per mm3, or AST or ALT greater than 5 x ULN
4. Known active or latent Hepatitis B
5. Known or suspected active tuberculosis (TB) or a history of incompletely treated or latent TB.
6. Pregnancy
7. Patients with uncontrolled systemic bacterial or fungal infections (Patients with a history of positive bacterial or fungal cultures but on enrollment are on appropriate therapy with negative repeat cultures may be enrolled)
8. Serious co-morbidity, including:
    a) Myocardial infarction (within last month)
    b) Moderate or severe heart failure (New York Heart Association (NYHA) class III or IV)
    c) Acute stroke (within last month)
    d) Uncontrolled malignancy
    e) Stage 4 severe chronic kidney disease or requiring dialysis (i.e. estimated glomerular filtration rate (eGFR) < 30 ml /min/1.73 m^2) at baseline

Subject eligibility will be assessed using medical record review and clinical tests. The Principle Investigator and/or Co-Investigator will determine eligibility. Patients may participate in other therapeutic studies while participating in this trial but all concomitant cytokine-directed, anti-viral and other COVID-19 experimental therapeutics will be annotated in reporting.

D.3 Withdrawal of Subjects

Participants may withdraw consent for participation in the study at any time. Participants may also be withdrawn from the study if, in the opinion of the investigator, it is in the patient's best interest to do so. Participants who elect to discontinue study treatment may still be followed for clinical events as appropriate.

D.4 Recruitment and Retention

D.4.1 Local Recruitment Methods

All eligible COVID-19 patients will be approached as part of their care at Tufts Medical Center. All participants will be recruited from the COVID-19 patient population at Tufts Medical Center. All COVID-19 trial investigators collaborate on identifying potential subjects and will determine the best course of treatment for these patients.

D.4.2 Study-Wide Recruitment Methods

All subjects will be recruited by methods under the control of Tufts Medical Center.

D.4.3 Payment

Subjects will not receive money, gifts, or any other incentive for participating in this study.

D.4.4 Reimbursement

Subjects will not be reimbursed for their expenses, such as travel, parking, meals, or other related costs.

E. Costs to Subjects

The drug will be dispensed from the Tufts Medical Center inpatient pharmacy and may be billed to the subject or their insurance. Participants will not be charged for research-related laboratory tests.

F. Study Design

F.1 Study Timelines

Subjects will participate in the study for up to 28 days. Enrollment is expected to last for approximately 12 weeks. The estimated date for investigators to complete this study is September 2020.

The following procedures will be performed at each study visit:

<u>Baseline Visit</u>

- Informed Consent
- Covid-19 Diagnosis Confirmation
- Demographic information
- Chest X-Ray or CT Scan
- Medical History
- Vital Signs: blood pressure, heart rate, respiratory rate, temperature, oxygen saturation
- CBC with Differential
- CMP
- Coagulation Profile
- Inflammatory Profile
- Cytokine Profile
- Hepatitis Panel
- Pregnancy Test (for women of child-bearing potential)
- O_2 Requirements
- Mode of Ventilation
- Vasopressor Support

<u>Day 1</u>

- Infliximab or Infliximab-abda Dosing
- Start Daily AEs of Special Interest/SAE Assessment
- Start Daily Vital Signs
- Start Daily O_2 Requirements
- Start Daily Mode of Ventilation

- Start Daily Vasopressor Support
- Start Daily CBC, CMP

Day 3

- CBC with Differential
- Chem-1
- Coagulation Profile
- Inflammatory Profile
- Cytokine Profile (between 48 – 72 hours post-treatment)

Day 5

- CBC with Differential
- Chem-1
- Coagulation Profile
- Inflammatory Profile

Day 14

This visit will be completed only if the subject is still an inpatient at Tufts Medical Center.

- CBC with Differential
- Chem-1
- Coagulation Profile
- Inflammatory Profile
- Cytokine Profile
- Infliximab or Infliximab-abda Dosing (at discretion of PI/Co-I +/- 7 days)

Day of Discharge

Day of discharge is the date of discharge from hospital, date of death or day 28, whichever occurs first.

- AEs of Special Interest/SAE Assessment
- Vital Signs
- O_2 Requirements
- Mode of Ventilation
- Vasopressor Support
- CBC with Differential
- Chem-1
- Coagulation Profile
- Inflammatory Profile
- Cytokine profile (only if discharge is between Day 7 and Day 14)
- Secondary Infections

Day 28 Follow-Up

- Survival Follow-Up

F.2 Procedures

Patients will be identified as they are seen at Tufts Medical Center. Safety labs and procedures will be performed per standard of care. The results of all labs and procedures will be reviewed by the study team and the PI.

Source records that will be used to collect data about subjects include documents from the Tufts Medical Center Electronic Medical Record, in addition to study coordinator checklists and case report forms (CRFs).

The following procedures and tests will be performed solely for research purposes:

- Blood collection for cytokine profile
- Informed Consent
- Treatment with Infliximab or Infliximab-abda 5mg/kg IV (See Section B.1.1)

All other procedures performed on this study are standard of care at Tufts Medical Center.

Below is a description of all procedures that will take place during the study:

Informed Consent

Informed Consent will be obtained prior to participation in this study. Informed consent will be obtained according to the Remote Consent Guidelines.

Covid-19 Diagnosis Confirmation

Patients must have a diagnosis of pneumonia by imaging and laboratory confirmed COVID-19 or with strong suspicion of COVID-19 with laboratory study pending to be eligible. This will be confirmed at Baseline prior to enrollment.

Adverse Events of Special Interest/Serious Adverse Events

Adverse events of special interest and serious adverse events will be collected from Day 1 until Day of Discharge. For more information see protocol section H.1.

Demographic information

Demographic information will be collected at Baseline and includes date of birth, gender and race/ethnicity.

Chest X-Ray or CT Scan

An X-Ray or CT Scan of the chest will be performed at any point during the admission prior to enrollment to confirm diagnosis of pneumonia.

Medical History

Medical history will be documented at Baseline and includes any ongoing medical co-morbidities (i.e.: cardiovascular, pulmonary, endocrine, oncologic diagnosis).

Vital Signs

Vital signs will be collected at baseline and throughout the study while participants are inpatients at the hospital. Vital signs include: heart rate, blood pressure, O2 saturation, respiratory rate, temperature, weight, height (on screening only).On the day of discharge, the first and last day of fever during the hospitalization will be recorded.

Complete Blood Count with Differential (CBC with Differential)

CBC with Differential will be collected at Baseline, Day 3, Day 5, Day 14, and on date of discharge. These labs may also be collected daily whenever possible or as per standard of care.

Complete Metabolic Panel (CMP)

CMP will be collected at Baseline, Day 3, Day 5, Day 14, and on date of discharge. Complete metabolic profile (CMP) includes: sodium, potassium, bicarbonate, chloride, glucose, creatinine, calcium, magnesium, estimated glomerular filtration rate, lactate dehydrogenase, blood urea nitrogen, calcium, magnesium, phosphate, total protein, alkaline phosphatase, aspartate aminotransferase, alanine aminotransferase, total and direct bilirubin. These labs may also be collected daily whenever possible or as per standard of care.

Coagulation Profile

Coagulation Profile will be collected at Baseline, Day 3, Day 5, Day 14, and on date of discharge. Key elements of the coagulation profile includes D-Dimer and fibrinogen.

Inflammatory Profile

Inflammatory Profile will be collected at Baseline, Day 3, Day 5, Day 14, and on date of discharge. Inflammatory profile includes C-reactive protein, troponin, Creatine phosphokinase and ferritin.

Cytokine Profile

A blood sample for cytokine profile testing will be collected at **Baseline** and **at least 48 hours but no later than 72 hours** following the first dose of infliximab or infliximab-abda. This will be repeated at Day 14 OR between Day 7 and Day 14 if the patient is discharged before Day 14. This cytokine profile includes TNFα, IL2, IL6, and IL1-beta. Instructions for sample collection and processing can be found in Appendix B.

Hepatitis Panel

Hepatitis panel will be performed at Baseline and includes: HepBs Ag and Ab, HepBc Ab and HepC Ab.

Pregnancy Test

Women of childbearing potential will undergo pregnancy testing at Baseline. Either a urine or serum pregnancy test is acceptable. Both tests are available at Tufts Medical

Center and the choice of which test is used will be at the discretion of the treating physician.

Infliximab or Infliximab-abda Dosing

Treatment with infliximab or infliximab-abda 5mg/kg IV should ideally be administered within 6 hours of enrollment, and no more than 24 hours following enrollment. Pre-medication with Tylenol 650 mg once 30 minutes prior to infusion would be recommended. Retreatment with infliximab is permitted at treating physician discretion 7-21 days following primary therapy and based on initial response; the usual treatment schedule is every 2 weeks, this interval is not strictly enforced given the uncertainty of outcomes with primary therapy. See section B.1.1 for more information regarding treatment with infliximab and infliximab-abda.

O2 Requirements

Fraction of inspired O_2 will be calculated at Baseline, within 24 hours of the first dose of infliximab, and at 24 hour intervals (+/- 12 hours) thereafter until primary end point is met, discharge, death or day 28 (which ever occurs first).

Mode of Ventilation

Mode of ventilation will be assessed throughout the study and until the day of discharge, death or day 28 (which ever occurs first). Mode of ventilation includes: none (on room air), nasal cannula/simple face mask/other, non-invasive ventilation, non-rebreather mask, high-flow nasal cannula, mechanical ventilation, extra-corporeal membrane oxygenation (ECMO). On day of discharge, the incidence and day of initiation and discontinuation of each mode of ventilation will be recorded.

Vasopressor Support

The incidence and date of initiation and discontinuation of vasopressor use will be recorded throughout the study and on the day of discharge, death or day 28 (which ever occurs first).

Secondary Infections

Any secondary infections that occur while the subject is an inpatient at Tufts Medical Center will be documented on the day of discharge. Secondary infections include any documented bacterial, fungal, parasitic and/or other viral infections.

Survival Follow-Up

Patients will be contacted 28 days following the initial dose of infliximab to assess survival status. This can be done via phone call, clinic visit, or review of medical records.

F.3 Evaluations

All laboratory tests, excluding the cytokine profile, will be performed as standard-of-care at the Tufts Medical Center Laboratory. Blood samples for cytokine profiles will be processed per the lab manual (Appendix B) and stored in the Neely Center laboratory. These assays will be performed at an outside facility.

F.4 Collection and Storage of Human Biological Specimens

Specimens for cytokine assays will be stored in the NCCCR laboratory for up to 1 year. NCCCR staff and study investigators will have access to the specimens, which are stored in a locked laboratory in Tufts Medical Center.

All specimens will be labeled with study ID only, a link to the patient identifier will be kept on a secure file accessible only to the principal investigator and study staff. IRB approval will be required for use of any specimen obtained from this study for future use.

Subjects can withdraw permission for use of their stored specimens and PHI for future research in writing, by email or letter, or documented phone conversation. The banked specimen will be labeled with a study ID number and we will keep a separate record linking the study ID to the patient so we can identify a sample if a participant wants to withdraw consent for future research.

G. Ethics and Protection of Human Subjects

G.1 Informed Consent Process

All subjects will be required to provide informed consent. The consent process will take place in private areas within Tufts Medical Center. The Principle Investigator or Co-Investigator will carry out the informed consent process, with the assistance from a study coordinator or study nurse. All patients will have long as necessary to make a decision about participation in the study. However due to the nature of the disease, it may be limited to 24 to 48 hours. To ensure the safety of study staff and investigators, a remote consent process will be used. This will be distributed separately. Study staff will ensure ongoing consent at every opportunity. Non-English speakers will be enrolled using interpreters and IRB approved Short Forms per the IRB's Short Form policy.

G.2 Waiver or Alteration of Consent Process

There will not be a request for a waiver or alteration of consent process for this study.

G.3 International Research

This study will only be conducted at Tufts Medical Center. There will be no additional sites, nationally or internationally.

G.4 Confidentiality

All data and specimens will be stored in a secure location only accessible by the research team. The PI will ensure that study documents are stored in a manner that protects the privacy of subjects and the confidentiality of study data. Signed ICFs and source documents will be stored in locked file cabinets in the Neely Center. De-identified data will be entered into a RedCap electronic database. Research bio

specimens will be processed in the NCCCR laboratory. Stored bio specimens will be kept in a locked freezer in the NCCCR laboratory.

Study numbers will be assigned and a codebook stored in a locked cabinet. Only the research team will have access to the file.

All study records will be retained for the timeframe described in the record retention policy of the "SOP – Records Retention Timeframe – Investigators". Long-term storage of study materials will take place at SPRY Moving and Storage.

G.5 Screening Data Collection Form/Screening Log

An IRB approved screening log will be used in this research study. This will be an identifiable screening log that will not be distributed or viewed outside of the institution. Only the research study team will have access to this log. This log will be stored securely with all other study documents.

G.6 Provisions to Protect the Privacy Interests of Subjects

All discussions with participants will occur in private settings. Study questions will ask about general medical information. Participants can withdraw consent for the study at any time, or decline to answer any questions.

G.7 Provisions to Monitor the Study to Ensure the Safety of Subjects

This is treatment-based study and patients will receive standard of care treatment for COVID-19 symptoms in addition to treatment with infliximab or infliximab-abda. Safety data will be monitored continuously while subjects are inpatients in COVID-19 units at Tufts Medical Center. Additionally, patient data will be reviewed at each study visit. All data will be reviewed by the study PI. A senior physician investigator (Raymond Comenzo, MD or Kenneth Miller, MD) within the Hematology & Oncology Division at Tufts Medical Center will review the safety data for all subjects enrolled onto this trial.

Case report forms and any clinical findings (sourced from EMR) will be reviewed by the PI or Co-Is at each study visit, or as-needed. Data collection will begin as soon as participants sign the informed consent form. Adverse events will be collected until 28 days following the last dose of study drug. Cumulative data will be analyzed at the end of the study period. In the first stage, 17 patients will be accrued. If there are five or fewer patients exhibiting improvement in oxygenation in these 17 patients, the study may be stopped.

G.8 Vulnerable Populations

This study will not involve enrollment of vulnerable subjects, including pregnant women, neonates, minors, prisoners, or students or employees at Tufts Medical Center. Patients who are not able to personally provide consent due to cognitive impairment or reduced performance status related to a medical condition (i.e. intubated patients, etc) may still be enrolled with a Legally Authorized Representative, including those with durable power of attorney for health care, court appointed guardian for health care decisions, a spouse, a health care proxy, or an adult child. This will be performed in accordance with the local IRBs "SOP: Legally Authorized Representatives, Children, and Guardians (HRP-013)." Assent will also be obtained whenever possible. For these subjects, ability

to give consent will be monitored continuously by clinic and study staff, and full consent would be obtained as soon as the patient is identified as able to provide it. However, it may still not be possible to obtain a signature due to their medical state. Every effort will be made to obtain informed consent and assent as appropriate given the condition of the patient. We will keep both the LAR and patient informed throughout the study as appropriate. An individual is determined to be capable of consent if at least two physicians (the treating physician and one of the investigators or another physician team member) determine based on their clinical judgment that the individual is capable of making their own decisions. These patients may be withdrawn from the trial if they appear to be under undue distress, as determined by the investigator. The investigators in collaboration with the clinical care teams will work together to assess withdrawal for each patient throughout their participation in the trial.

H. Adverse Event Monitoring

H.1 Definitions

Only adverse events of special interest will be collected for the purposes of this trial. Adverse events of special interest include infusion reactions from infliximab treatment, anaphylaxis, and any other event deemed by the investigator to be directly related to infliximab treatment.

An adverse event or suspected adverse reaction is considered "serious" if, in the view of either the investigator or sponsor, it results in any of the following outcomes: Death, a life-threatening adverse event, inpatient hospitalization or prolongation of existing hospitalization, a persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions, or a congenital anomaly/birth defect. The PI will assess each SAE as they become aware. Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

Unanticipated problems (UPs) are defined as any incident, experience or outcome that meets all of the following criteria:

- Unexpected (unforeseen by the researcher or the research participant) in terms of nature, severity, or frequency, given the research procedures and the subject population being studied;
- Related or probably related to participation in the research, or if the event or problem probably or definitely affects the safety, rights and welfare of current participants;
- Suggests that the research places subjects or others at a greater risk of harm (including physical, psychological, economic or social harm) than was previously known or recognized.

H.2 Reporting Procedures

The PI will review all study data to assess for any AEs possibly related to the research procedures. The PI would report to the IRB within 48 hours if any deaths or SAEs occur which could be related or possibly related to the research procedures, or any unexpected AEs that may be related to the research procedures. Any AEs possibly related to the research procedures will be collected and any unexpected AEs possibly related to the research procedures will be reported to the IRB within five business days

per the Tufts Health Sciences IRB reportable new information policy. Any unanticipated problems possibly related to the research procedures will also be collected and reported to the IRB within five business days.

H.3 Reportable New Information

All reportable new information will be reported to the IRB per the Tufts Health Sciences IRB's Reportable New Information policy.

I. Statistical Considerations

I.1 Study Endpoints

Primary endpoint:

Time to improvement in oxygenation (increase in SpO2/FiO2 of 50 or greater compared to the baseline SpO2/FiO2) for a minimum of 48 hours

Secondary endpoints:

1. 28-day mortality
2. Assessment of dynamic changes in cytokine and inflammatory profile after therapy
3. Qualitative and quantitative toxicity
4. Incidence and duration of supplemental oxygen administration by nasal cannula, simple face mask, or other similar oxygen delivery device
5. Incidence and duration of non-invasive ventilation or by non-rebreather mask or high-flow nasal cannula
6. Incidence and duration of mechanical ventilation
7. Incidence and duration of vasopressor support
8. Incidence and duration of extracorporeal membrane oxygenation (ECMO)
9. Duration of fever
10. Correlation of cytokine and inflammatory profile to clinical outcomes
11. Duration of hospitalization
12. Secondary infections

I.2 Statistical Analysis:

The design of the study will be based on estimated 75% risk of worsening oxygenation for the spectrum of severe COVID-19 lung disease allowed in the study (25% probability of improvement). This estimate is based on a mixture of patients with non-invasive and invasive oxygenation support. We will target a 50% rate of improvement in oxygenation following therapy with the experimental strategy. A Simon's two-stage design will be used. The null hypothesis that the true improvement of oxygenation is only 25% will be tested against a one-sided alternative. In the first stage, 17 patients will be accrued. If there are 5 or fewer patients exhibiting improvement in oxygenation in these 17 patients, the study will be stopped. Otherwise, 20 additional patients will be subsequently accrued for a total of 37 patients. The null hypothesis will be rejected if 13 or fewer responses are observed in 37 patients. This design yields a type I error rate of 0.05 and power of 0.9 when the true response rate is 25%. Mean cytokine levels at baseline and the mean dynamic change in cytokines between responders and non-responders will be compared using a t-test. Other secondary endpoints will be annotated by descriptive markers.

I.3 Number of Subjects

In the first stage, 17 patients will be accrued. If there are five or fewer patients exhibiting improvement in oxygenation in these 17 patients, the study may be stopped. Otherwise, 20 additional patients will be accrued for a total of 37 patients.

I.4 Data Management

All research coordinators go through training on a regular basis. All paper records will be kept in a locked cabinet and all electronic records will be kept in a password protected shared drive and only those directly involved with the study will have access. All participants will be assigned a study ID and the RedCap database will have de-identified data only. The Neely Center study coordinators will input all data into the RedCap database. All data will be reviewed by the PI.

Only the research study team will have access to the data and specimens. The primary documents will be stored in a locked cabinet, the electronic files will be kept in a password protected shared file on the Tufts Medical Center server and the bio specimens and redcap database will use a de-identified study ID number. The key that associates the de-identified study ID number with the patient name will be kept separately from the database and bio specimens in a locked location.

Data will remain at Tufts Medical Center. Bio specimens will be transported by a research coordinator to the Neely Center laboratory for specimen processing and storage.

Data and/or specimens will be stored for at least 7 years.

I.5 Randomization

Subjects will not be randomized in this trial.

J. Drugs or Devices

This research will involve the use of drugs. The Tufts Medical Center pharmacy, delegated study staff, and the PI/Co-Is will be accountable for the drugs used for the

study. Orders will made by the study staff and PI/Co-Is, and will be reviewed and signed by PI/Co-I. This will be completed using the EMR and/or physical order forms.

Patients should not receive any live vaccinations that could lead to a clinical infection while on the study. Additionally, the use of other anti-cytokine agents, anti-viral agents or other experimental therapeutics for COVID-19 disease is not prohibited. However, all disease-modifying therapies that occur while the subject is on-study will be documented.

Women of childbearing potential should employ the use of effective contraception methods to avoid pregnancy for 6 months following the last dose of infliximab/infliximab-abda, per manufacturer recommendations.

K. Study Administration

K.1 Setting

All research related activities will take place at Tufts Medical Center.

K.2 Registration

Subject Eligibility Checklists will be used to ensure that a subject is appropriately enrolled in the study prior to receiving any study intervention. Eligibility checklists will be reviewed and signed by the PI or Co-I prior to registration. The Investigators and study staff will also ensure consent and other study procedures are documented appropriately.

K.3 Resources Available

All study staff will be trained on study procedures on a regular basis. The PI will ensure all study staff are qualified to perform the procedures to which they are delegated. The research coordinators will screen patient lists and approach patients for consent along with the PI/Co-I. The PI will supervise the informed consent discussions. Blood and/or urine will be collected at the time of standard of care labs for treatment in the inpatient setting by the phlebotomist and/or staff nurse. Research samples will be transported by the research coordinators to the NCCCR laboratory for processing and storage. All CRFs will be reviewed and confirmed by the study PI.

Dr. Andreas Klein, a Co-I on this study will be the acting PI in the event that the PI is away or unavailable.

Medical or psychological resources that subjects might need, such as for emergencies or medical issues, will be available for the study.

K.4 IRB Review

This study will be reviewed and approved by an appropriate IRB registered with the OHRP. Any amendments to the protocol or informed consent documents will be reviewed and approved by the IRB prior to use, unless required to eliminate an apparent immediate hazard to subjects.

K.5 Multi-Site Research

This is not a multi-site study where Tufts is the sponsor, primary grant recipient, or coordinating site.

K.6 Community-Based Participatory Research

This study will not involve community-based participatory research.

K.7 Sharing Results with Subjects

The results of clinical tests performed as part of standard of care and will be reported according to routine practice to the patients' treating physician. The results of exploratory biomarkers will not be given to the participants or treating physicians.

L. References

1. *WHO. Coronavirus disease 2019 (COVID-19) situation report – 70. March 30, 2020. https://www.who.int/docs/default-source/coronaviruse/situation-reports/20200330-sitrep-70-covid-19.pdf?sfvrsn=7e0fe3f8_2 (accessed March 30, 2020).*

2. *Huang C, Wang Y, Li X, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet 2020; 395: 497–506*

3. *Emanuel E.J, et al. Fair Allocation of Scarce Medical Resources in the Time of Covid-19. New England Journal of Medicine. DOI: 10.1056/NEJMsb2005114.*

4. *Lauer SA, et al. The incubation period of Coronavirus Disease 2019 from publicly reported confirmed cases: Estimation and Application. Ann Intern Med. Published online March 10, 2020 doi:10.7326/M20-0504*

5. *Liu Y, et al. Viral dynamics in mild and severe cases of Covid-19. Lancet Infect Dis. Published Online March 19, 2020 https://doi.org/10.1016/ S1473-3099(20)30232-2*

6. *Shi, Y., Wang, Y., Shao, C. et al. COVID-19 infection: the perspectives on immune responses. Cell Death Differ (2020). https://doi.org/10.1038/s41418-020-0530-3*

7. *Crayne CB, et al. The immunology of macrophage activation syndrome. Front Immunol 2019 Feb 1;10:119. doi: 10.3389/fimmu.2019.00119. eCollection 2019.*

8. *Ruan Q, Yang K, Song J. Clinical predictors of mortality due to COVID-19 based on an analysis of data of 150 patients from Wuhan, China. Intensive Care Med. 2020 Mar 3. doi: 10.1007/s00134 020-05991-x.*

9. *Zhang D, et al. COVID-19 infection induces readily detectable morphological and inflammation-related phenotypic changes in peripheral blood monocytes, the severity of which correlate with patient outcome. medRxiv preprint. Posted March 26, 2020. doi: https://doi.org/10.1101/2020.03.24.20042655*

10. *Tisoncik JR, Korth MJ, Simmons CP, Farrar J, Martin TR, Katze MG. Into the eye of the cytokine storm. Microbiol Mol Biol Rev. 2012;76(1):16-32*

11. *Wang M, Cao R, Zhang L, Yang X, Liu J, Xu M, et al. Remdesivir and chloroquine effectively inhibit the recently emerged novel coronavirus (2019-nCoV) in vitro. Cell Res. 2020;30(3):269-71*

12. *Maude SL, Barrett D, Teachey DT, Grupp SA. Managing cytokine release syndrome associated with novel T cell-engaging therapies. Cancer J. 2014;20(2):119-22.*

13. *Flammiger A, Fiedler W, Bacher U, Bokemeyer C, Schneider M, Binder M. Critical imbalance of TNF-alpha and soluble TNF receptor 1 in a patient with macrophage activation syndrome: potential implications for diagnostics and treatment. Acta Haematol. 2012;128(2):69-72.*

14. *Ideguchi H, Ohno S, Takase K, Hattori H, Kirino Y, Takeno M, et al. Successful treatment of refractory lupus-associated haemophagocytic lymphohistiocytosis with infliximab. Rheumatology (Oxford). 2007;46(10):1621-2.*

15. *Xu X HM, Li T et al. Effective treatment of severe COVID-19 patients with Tocilizumab. Available on chinaXiv website Accessed online 2020 Mar 19.*

16. *Poston J, et al. Management of critically ill adults with COVID-19. JAMA. Published online March 26, 2020. doi:10.1001/jama.2020.4914*

17. *Genentech I, South San Francisco, CA. Actemra use in Coronavirus Disease 2019 (COVID-19) standard reply letter. 2020 Mar 16*

18. *Fiers W. Tumor necrosis factor. Characterization at the molecular, cellular and in vivo level. FEBS Lett. 1991;285(2):199-212*

19. *Sundgren-Andersson AK, Ostlund P, Bartfai T. IL-6 is essential in TNF-alpha-induced fever. Am J Physiol. 1998;275(6):R2028-34.*

20. *Muntane-Relat J, Ourlin JC, Domergue J, Maurel P. Differential effects of cytokines on the inducible expression of CYP1A1, CYP1A2, and CYP3A4 in human hepatocytes in primary culture. Hepatology. 1995;22(4 Pt 1):1143-53.*

21. *Mehta P, et al. COVID-19: Consider cytokine syndromes and immunosuppression. Lancet. Published online March 12, 2020. https://doi.org/10.1016/ S0140-6736(20)30630-9*

22. *Brennan FM, Chantry D, Jackson A, Maini R, Feldmann M. Inhibitory effect of TNF alpha antibodies on synovial cell interleukin-1 production in rheumatoid arthritis. Lancet. 1989;2(8657):244-7.*

23. *Sladkova T, Kostolansky F. The role of cytokines in the immune response to influenza A virus infection. Acta Virol. 2006;50(3):151-62.*

24. *Fritz RS, Hayden FG, Straus Sec et al. Nasal Cytokine and Chemokine Responses in Experimental Influenza A Virus Infection: Results of a Placebo-*

Controlled Trial of Intravenous Zanamivir Treatment. The Journal of Infectious Diseases, Volume 180, Issue 3, September 1999.

25. *Garcia-Ramirez RA, Ramirez-Venegas A, Quintana-Carrillo R, Camarena AE, Falfan-Valencia R, Mejia-Arangure JM. TNF, IL6, and IL1B Polymorphisms Are Associated with Severe Influenza A (H1N1) Virus Infection in the Mexican Population. PLoS One. 2015;10(12):e0144832.*

26. *Shi X, Zhou W, Hai Huang, Ju D. Inhibition of the inflammatory cytokine tumor necrosis factor-alpha with etanercept provides protection against lethal H1N1 influenza infection in mice. Crit Care. 2013; 17(6): R301. doi: 10.1186/cc13171*

27. *Rockx B, et al. Early upregulation of acute respiratory distress syndrome-associated cytokines promotes lethal disease in an aged-mouse model of severe acute respiratory syndrome coronavirus infection. J Virol 2009, 83(14): 7062-7074*

28. *Beijing Group of National Research Project for SARS. Dynamic changes in blood cytokine levels as clinical indicators in severe acute respiratory syndrome. Chin Med J (Engl). 2003 Sep;116(9):1283-7.*

29. *Hsueh P, Chen P, Hsiao C, et al. Patient Data, Early SARS Epidemic, Taiwan. Emerging Infectious Diseases. 2004;10(3):489-493. doi:10.3201/eid1003.030571*

30. *Seo SH1, Webster RG. Tumor necrosis factor alpha exerts powerful anti-influenza virus effects in lung epithelial cells. J Virol. 2002 Feb;76(3):1071-6.*

31. *Hussell T1, Pennycook A, Openshaw PJ. Inhibition of tumor necrosis factor reduces the severity of virus-specific lung immunopathology. Eur J Immunol. 2001 Sep;31(9):2566-73*

32. *Zheng J, Shi Y, Xiong L, Zhang W, Li Y, Gibson PG, et al. The Expression of IL-6, TNF-alpha, and MCP-1 in Respiratory Viral Infection in Acute Exacerbations of Chronic Obstructive Pulmonary Disease. J Immunol Res. 2017;2017:8539294.*

33. *Hamacher J, Lucas R, Lijnen HR, Buschke S, Dunant Y, Wendel A, et al. Tumor necrosis factor-alpha and angiostatin are mediators of endothelial cytotoxicity in bronchoalveolar lavages of patients with acute respiratory distress syndrome. Am J Respir Crit Care Med. 2002;166(5):651-6.*

34. *Tian S, Hu W, Niu L, Liu H, Xu H, Xiao SY. Pulmonary Pathology of Early-Phase 2019 Novel Coronavirus (COVID-19) Pneumonia in Two Patients With Lung Cancer. J Thorac Oncol. Published online 2020 Feb 28. doi: 10.1016/j.jtho.2020.02.010*

35. *Xu Z, Shi L, Wang Y, Zhang J, Huang L, Zhang C, et al. Pathological findings of COVID-19 associated with acute respiratory distress syndrome. Lancet Respir Med. Published Online February 17, 2020 https://doi.org/10.1016/ S2213-2600(20)30076-X*

36. *Leipe J, et al. Role of Th17 cells in human autoimmune arthritis. Arthritis Rheum 2010 Oct;62(10):2876-85. doi: 10.1002/art.27622.*

37. *Heard SO, Perkins MW, Fink MP. Tumor necrosis factor-alpha causes myocardial depression in guinea pigs. Crit Care Med. 1992;20(4):523-7.*

38. *Mann DL. Tumor necrosis factor and viral myocarditis: the fine line between innate and inappropriate immune responses in the heart. Circulation. 2001;103(5):626-9*

39. *Feldmann M, et al. Trials of anti-tumour necrosis factor therapy for COVID-19 are urgently needed. Lancet. Published online April 9, 2020 https://doi.org/10.1016/S0140-6736(20)30858-8*

Appendix A – Tufts Medical Center COVID-19 Treatment Guidance Document for Inpatients Guideline on Anti-Inflammatory Management

Updated as of 10 April 2020

This is one of a series of collaborative documents developed by members of ID, Pulmonary/Critical Care, Hematology, Nephrology, Rheumatology, Pharmacy, Cardiology, Radiology, Dermatology, Pediatrics and Pathology Divisions and Departments.

Please note the date at the bottom of the page; this document will be updated as the COVID-19 pandemic evolves and new data becomes available. There will be changes.
Please see other existing documents located on intranet for current information on definitions of persons under investigation (PUI), testing guidelines, Algorithms for disposition and personal protective equipment (PPE), exposures, and FAQ.

There is currently no FDA-approved treatment for COVID-19 or agent with clear benefit at this time.

Changes since last version:

Addition of Corticosteroid Recommendations

Revisions to diagnostic criteria and treatment algorithm for cytokine storm

Requirement for Hematologist Approval for use of anti-cytokine therapy

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Corticosteroids

There is currently uncertain benefit and risk of corticosteroids in the treatment of COVID-19. Corticosteroids continue to have a controversial role in the treatment of non-infectious ARDS, and data show both detrimental and beneficial impacts on patients with other viral pneumonias including those caused by MERS and influenza (Russel CD, et al. *Lancet;* Li H, et al. *Influenza Other Respi Viruses*). Preliminary observational data suggests there may be a potential benefit of steroids in those with ARDS caused by COVID-19 (Wu C, et al. *JAMA Internal Medicine*) but this data is confounded by the lack of a true comparator.

Corticosteroids should not be withheld if underlying indication exists (i.e. adrenal insufficiency, COPD exacerbation).

For the management mechanically ventilated patients **without ARDS**, corticosteroids are not thought to be beneficial and may increase risk of mortality.

For the management of mechanically ventilated patients **with ARDS**, there is a potential role for corticosteroid therapy. A patient-specific risk vs. benefit decision should be made on a case by case basis by the critical care team.

Cytokine Storm

Definition:
Cytokine storm applies to a condition of dysregulated immune activation and cytokine release resulting in fever, vasogenic edema, and hypotension which may mimic severe sepsis. Clinical

entities associated with massive cytokine release include macrophage activation syndrome (MAS) and hemophagocytic lymphochistiocytosis (HLH). Cytokine storm is believed responsible for clinical deterioration and progressive pulmonary damage occurring in severe cases COVID-19 infection.

Diagnosis:
No formal diagnostic criteria for cytokine storm have been defined in the setting of COVID-19 but may be suspected on clinical grounds. The presence of the three criteria below may constitute sufficient evidence of cytokine storm to institute anti-cytokine treatment.

1. Appropriate clinical context: COVID+, progression of symptoms >= 3 days after onset of symptoms or positive test
2. Clinical deterioration: persistent fever and progressive hypoxemia:
   Escalation to ≥ 4-6L/min O2 via NC to maintain saO2 > 92%
   Transitioning from NC to NRB, BiPAP/HFNC or intubation
   BiPAP increased to FiO2 100%
   Increasing HFNC ≥ 60L/min, FiO2 ≥ 80%
   Intubated within 24h
3. Laboratory evidence of inflammatory state: elevation >2x ULN of any of ferritin, CRP or LDH

Laboratory Evaluation:
Patients suspected with cytokine storm should have the following lab work collected at the time of treatment initiation and repeated 48 to 72 hours later for evaluation of effect:

CBC with differential
PT, fibrinogen, D-dimer / fibrin split products
Ferritin, C-reactive Protein, CPK, troponin
sIL-2R, IL-6 (send out to Viracor)
Procalcitonin

Treatment:
Treatment of cytokine storm is aimed at blocking mediators of inflammation. Two targets have been evaluated and demonstrated responses in clinical practice: IL-6 and IL-1. In order of preference and subject to availability, the following interventions below are suggested. Anticytokine therapy may increase the risk of infection or impair the ability to fight concurrent bacterial, fungal or mycobacterial infection – use should proceed with caution.

Sarilumab on clinical trial
Tocilizumab 4mg/kg IV over at least 1 hour x 1 – give whole 400mg vial, repeat x1 in 6-12 hours if response inadequate; dose of 8mg/kg (max 800mg) may be considered
Infliximab 5mg/kg IV over at least 2 hours x 1 (may be preferred for patients with particularly high ferritin)
Unlikely to be available for use
Siltuximab 11mg/kg IV over 1 hour x 1, repeat x1 in 6-12 hours if response inadequate
Sarilumab 200mg SC x1, repeat x1 in 6-12 hours if response inadequate
Anakinra 100mg SC daily x 3-5 days, consider in setting of sepsis

Consultation:
Hematology consultation is available 24/7 for questions regarding these guidelines, for refractory cases, or where alternative diagnosis is suspected. Administration of anti-cytokine therapy according to the above guidelines does not require a formal hematology consultation, but requires approval from one of the following doctors: Andreas Klein, Jason Law, Tishi Shah, Hannah Fassel, Cathy Rosenfield, Laura Wiltsie, or Blythe Thompson.

Appendix B – Laboratory Manual

WHEN: Cytokine assays will be collected at baseline and at least 48 hours but no later than 72 hours following the first dose of infliximab or infliximab-abda. A Day 14 assay will also be drawn on all patients remaining hospitalized whereas for patients who are planned for discharge between Day 7 and Day 14, a repeat assay will drawn prior to discharge. Research staff will coordinate the orders to be integrated with routine clinical care.

WHERE: These samples will be drawn by qualified inpatient staff at the time of routine standard-of-care blood draws whenever possible.

WHAT: For the cytokine panel, approximately 6 mL of blood will be collected in an EDTA tube

HANDLING: A member of the study staff will then gently invert the tube 8-10 times, before transporting the specimen from the inpatient unit to the lab for processing.

PROCESSING:

f) Blood samples will then be centrifuged by study staff at 1000 x g for 10 min at 4°C, **within 30 minutes of blood collection and no later than 2 hours after collection**.
g) Plasma will be collected immediately into two collection tubes: Tube #1 will contain 0.5 ml of plasma and Tube #2 with 0.5 ml of plasma).
h) Tubes #1 and #2 will be 0.5 ml Eppendorf Safe-Lock Tubes.

i) Each tube will be labeled alpha numerically with T (T for Tufts) followed by consecutive numbers.
j) Both tubes will then be placed in a -20°C freezer until ready for shipment.

SHIPPING FOR ANALYSIS OF CYTOKINES:

An outside laboratory facility, Eve Technologies Corporation, will conduct the cytokine assays. Undiluted tube #1 will be shipped to EVE technologies for Human Cytokine Array / Chemokine Array 48-Plex (HD48). Samples will be batched and shipped weekly using FedEx. Samples can be shipped on Mondays and Tuesdays only. The contact and shipping information is listed below:

Eve Technologies Corporation
Address: 3415A - 3 Ave., N. W. Calgary, AB. Canada T2N 0M4
email: admin@evetechnologies.com
website: www.evetechnologies.com
Tel: 587-975-8850
Fax: 587-975-8899

All samples will be shipped frozen on dry ice. Prior to shipment, samples will be removed from the freezer and placed in a plastic or cardboard sample box. The box will then be placed in a leak proof bag with absorbent material, which will then be placed in a Styrofoam insulated shipping boxes with a corrugated fiberboard outer box with a **minimum of 5 kg of dry ice**. A Class 9 Dry-ice label (UN 1845) will be affixed on outer packaging (with weights and addresses filled in). As the samples are infectious category B, UN 3373 label will also be affixed on the outer box. Eve Technologies Corporation will be emailed on the day of shipment with the shipment manifest and tracking information.

Appendix C – Schedule of Assessments

Study Procedures	Baseline	Day 1[1]	Day 2	Day 3	Day 4	Day 5	Day 14	Day of discharge[2]	Day 28 Follow-up
Informed consent	X								
Diagnosis confirmation	X								
Demographic information	X								
Medical History	X								
Vital signs	X	X	X	X	X	X	X	X	
AEs of special interest/SAEs	X	X	X	X	X	X	X	X	
Chest Xray or CT scan	X								
Infliximab or Infliximab-abda Dosing[1, 4]		X					X[1, 4]		
Required laboratory tests									
Pregnancy Test[5]	X								
CBC with differential[3]	X			X		X	X	X	
CMP[3]	X			X		X	X	X	
Coagulation profile[3]	X			X		X	X	X	
Inflammatory profile[3]	X			X		X	X	X	
Cytokine profile[3]	X			X			X	X[6]	
Hepatitis panel	X								
Disease and response assessment									
O2 requirements	X	X	X	X	X	X	X	X	
Mode of ventilation	X	X	X	X	X	X	X	X	
Vasopressor support	X	X	X	X	X	X	X	X	
Secondary infections								X	
Follow-up									
Survival Follow-up									X

1- Day 1 when anti-TNF therapy is administered, within 6 hours of enrollment and no more than 24 hours following enrollment.
2- Day of discharge is the date of discharge from hospital, date of death or day 28, whichever occurs first.
3- It is strongly preferred that all laboratory samples be collected per the schedule above (+/- 24 hours).
4- Retreatment with infliximab at treating physician discretion 7-21 days following primary therapy and based on initial response; the usual treatment schedule is every 2 weeks, this interval is not strictly enforced given the uncertainty of outcomes with primary therapy. Follow-up of clinical and inflammatory markers will continue in this context as prescribed following Day 1of therapy.
5- Pregnancy testing is required at Baseline only for women of childbearing potential. Either serum or urine testing is acceptable.
6- Only if discharge date is greater than or equal to Day 7 and less than Day 14 following Day 1 infliximab treatment.