Tocilizumab Use in COVID-19 Cytokine-release Syndrome: Retrospective Study of Two Centers
Materials And Methods-

Retrospective case-control analysis of all COVID-19 patients between March 15, 2020 to May 15, 2020 with severe to critical disease in ICU.

- Patients were evaluated for CRS, and 22 patients who met the criterion were given tocilizumab.
- The primary objective was to evaluate the effect of tocilizumab on escalation of respiratory support and ICU mortality.
- The secondary objectives were ICU length of stay, trends of inflammatory markers, and any adverse effects.
**Table 2: Criteria for diagnosis of CRS and tocilizumab administration**

### A. Clinical criteria

|                  | Definition                                                                 | Intervention                      |
|------------------|---------------------------------------------------------------------------|-----------------------------------|
| Grade I mild     | Pneumonia with no oxygen requirement                                       | No treatment with TCZ             |
| Grade II moderate| Fever, need for IV fluid (not hypotension), mild oxygen requirement (FiO\textsubscript{2} less than 40%) | No treatment with TCZ             |
| Grade III severe | New organ dysfunction: liver test dysfunction, acute kidney injury, sepsis: IVF for resuscitation, low-dose vasopressor, supplemental oxygen (HFNC, HFNBM, FiO\textsubscript{2} ≥ 40%, NIV) | Send IL-6                         |
| Grade IV critical| Life-threatening, mechanical ventilation, high-dose vaspressors            | Consider TCZ                      |

### B. Inflammatory markers

| Marker          | Definition                                    | Intervention                        |
|-----------------|-----------------------------------------------|-------------------------------------|
| Ferritin        | 300 µg/L with doubling within 24 hours        |                                     |
| LDH             | >600 µg/L at presentation                      |                                     |
| Elevated D-dimer| >250 U/L                                      |                                     |
| CRP             | >1 mg/mL                                       |                                     |
| CRP             | >150                                           |                                     |

### C. Interleukin-6 (IL-6): more than 10 times of upper limit of normal (<7 pg/mL)

### D. Exclusion criteria for tocilizumab exclusion

- Active TB
- AST/ALT values higher than 5 times the normal levels.
- Neutrophil value lower than 500 cells/mm\textsuperscript{3}
- Platelets value lower than 50,000 cells/mm\textsuperscript{3}
- Complicated diverticulitis or intestinal perforation.
- Confirmed systemic bacterial and/or fungal infection (i.e., bacteremia with pathogenic bacteria, fungemia)
- Pregnant women
- Skin infection in progress (e.g., dermohypodermatitis not controlled by antibiotic therapy)
- Immunosuppressive antirejection therapy
- Absence of overt bacterial or fungal infection.

IL-6, interleukin-6; AST, aspartate aminotransferase; ALT, alanine aminotransferase; TB, tuberculosis; LDH, lactate dehydrogenase; CRP, C-reactive protein; TCZ, tocilizumab; HFNC, high-flow nasal canula; HFNBM, high-flow (more than 10 ltr) nonrebreathing mask; NIV, noninvasive ventilation
Standard Treatment –

The management of the patients in severe and critical category included—

• hydroxychloroquine, 400 mg 12 hourly on day 1 followed by 200 mg 12 hourly for 10 days,
• lopinavir 400 mg + ritonavir 100 mg combination twice a day for 10 days, or
• favipiravir 1600 mg 12 hourly on day 1 followed by 600 mg 12 hourly for 2–5 day as per national guidelines.
• Other supportive treatments included low dose steroids, anti coagulation and oxygen support.
• Daily monitoring by ECG biochemistry and hemodynamic parameters.
• NIV/IMV if needed.
Data Collection-

Data collected of 22 patients who received two doses of tocilizumab for inflammatory markers, total leukocyte count (TLC) and lymphocyte percentage, oxygenation (PaO2/FiO2 ratio), and type of respiratory support for seven days.

The patient demographics and comorbidities, ICU length of stay (ICU-LOS), and mortality at day 7 and day 28 of all 85 patients were collected

Statistical Calculation-

• The continuous variables were expressed as means [standard deviation (SD)], medians (interquartile ranges). The categorical variables were expressed in counts and percentages. Categorical variables compared using Fisher’s exact or Chi square and paired t test and repeated measures ANOVA (trend in repeated measurements) for continuous variables. A p value less than 0.05 was taken as significant. IBM SPSS (version 26.0, Armonk, NY: IBM Corp.) was used for analysis. All investigations were conducted according to the principles expressed in the Declaration of Helsinki.
Objectives of the Study

Primary Objective

- Effect of tocilizumab on mortality at day 7 and 28 in patients with severe and critical illness as compared to standard treatment.
- Effect of tocilizumab on the need for escalation of respiratory support (invasive or noninvasive ventilation) as compared to standard treatment.

Secondary Objective

- Effect of tocilizumab on ICU-LOS vs standard treatment.
- To understand any significant trend in inflammatory markers in response to tocilizumab administration.
- To identify any adverse events including hepatotoxicity, allergic reaction, and secondary infections in tocilizumab group.
Results

• The data from all 85 patients were collected.

Demographic Characteristics:

• Most of the patients were male in both tocilizumab and standard treatment groups.

• The average age was 51 years in the tocilizumab group vs 52 years in the standard treatment group (p = 0.684).

• Obesity (BMI > 30 kg/m2) was a common risk among patients admitted to ICU, 63% in the tocilizumab group and 66.7% in standard treatment group (p = 0.799) (Table 3).

• Comorbidities Hypertension was the most common comorbidity in both the tocilizumab and standard treatment groups (72% vs 63%, p = 0.604) followed by diabetes mellitus (59% vs 55.6% p = 0.808).
Outcomes

Primary Outcome (Table 3)

• Mortality: The mortality was significantly lower in the tocilizumab group vs standard treatment group at day 7 (9.1% vs 39.7%, p = 0.007) and 28 (9.1% vs 57.1%, p = 0.001) (Fig. 1).
• Escalation of respiratory support: the need for escalation of respiratory support was also significantly lower in the tocilizumab vs standard treatment group (22.7% vs 65.1% p = 0.001) (Fig. 1).
• There was also a requirement of less invasive ventilation in the tocilizumab group (18.2% vs 61.95%, p = 0.000).

Secondary Outcome

• CRP reduction among the inflammatory markers showed a significant trend in the tocilizumab group by day 3 (p = 0.033) (Table 4 and Fig. 2).
• ICU length of stay was more in the tocilizumab group vs standard treatment group (11.31 vs 9.73, p = 0.35) (Table 3).
• There were two patients (9.1%) in the tocilizumab group with elevations of liver function tests (LFT)—five times the baseline and two patients (9.1%) developed secondary sepsis, both bacterial, within one week of administration requiring escalation or change of antibiotics.
• The tocilizumab administration from day of hospital admission was a median of three (1–8) days.
Fig. 2: Outcome between tocilizumab and standard treatment

| Variable                 | Tocilizumab (percentage of total) | Standard treatment (percentage of total) | p value | p ≤ 0.05, significant |
|--------------------------|-----------------------------------|-------------------------------------------|---------|-----------------------|
| Number of patients       | 22                                | 63                                        |         |                       |
| Gender (male/female)     | 22/0                              | 60/3                                      |         |                       |
| Average age (mean)       | 51                                | 52                                        |         | p = 0.684             |
| BMI (>30 kg/m²)          | 14(63%)                           | 42 (66.7%)                                |         | p = 0.799             |
| Comorbidities            |                                    |                                           |         |                       |
| Hypertension             | 16(22%)                           | 41 (63%)                                  |         | p = 0.664             |
| Diabetes mellitus        | 12 (59%)                          | 35 (56.6%)                                |         | p = 0.808             |
| Cardiovascular disease   | 5 (22.7%)                         | 19 (30.1%)                                |         | p = 0.590             |
| COPD or asthma           | 2 (8.6%)                          | 10 (15.9%)                                |         | p = 0.723             |
| Chronic kidney Disease   | 3(12.0%)                          | 8 (14.3%)                                 |         | p = 1.00              |
| Invasive ventilation     | 4(18.2%)                          | 39 (61.0%)                                |         | p = 0.000             |
| Outcome                  |                                    |                                           |         |                       |
| Escalation of treatment  | 5 (22.7%)                         | 41 (65.1%)                                |         | 0.001                 |
| Mortality at day 28      | 2 (9.1%)                          | 16 (57.1%)                                |         | 0.001                 |
| Mean ICU LOS (SD) (in days) | 11.31 (5.21)        | 6.73 (3.12)                               |         | 0.35                  |

BMI, body mass index; COPD, chronic obstructive pulmonary disease; LOS, length of stay; p values ≤ 0.05

| Variables     | Base      | Day 1      | Day 2      | Day 3      | Day 4      | Day 5      | Day 6      | Day 7      | p value |
|---------------|-----------|------------|------------|------------|------------|------------|------------|------------|---------|
| LDH           | Mean 563.13 | 666.23     | 671.53     | 555.55     | 760.02     | 845.90     | 755.88     | 654.08     | 0.082   |
|               | SD 223.97  | 273.25     | 199.44     | 288.25     | 254.93     | 271.89     | 296.50     | 196.41     |         |
|               | n 6        | 6          | 6          | 6          | 6          | 6          | 6          | 6          |         |
| CRP           | Mean 168.84 | 209.92     | 145.64     | 108.58     | 59.42      | 24.16      | 11.90      | 6.58       | 0.033   |
|               | SD 91.44   | 87.53      | 32.40      | 82.00      | 41.61      | 15.21      | 9.84       | 5.20       |         |
|               | n 5        | 5          | 5          | 5          | 5          | 5          | 5          | 5          |         |
| Ferritin      | Mean 1465.23 | 1623.42    | 1779.43    | 1766.50    | 1757.67    | 1568.17    | 1280.45    | 1309.50    | 0.245   |
|               | SD 835.01  | 582.81     | 469.66     | 571.96     | 590.17     | 697.04     | 657.22     | 627.01     |         |
|               | n 6        | 6          | 6          | 6          | 6          | 6          | 6          | 6          |         |
| D-clinr       | Mean 726.04 | 791.00     | 1395.00    | 934.00     | 1853.20    | 1911.20    | 1891.40    | 2277.60    | 0.438   |
|               | SD 479.45  | 505.73     | 2015.74    | 786.95     | 1993.19    | 1831.16    | 1251.44    | 1912.89    |         |
|               | n 5        | 5          | 5          | 5          | 5          | 5          | 5          | 5          |         |
| Lympho        | Mean 8.78  | 9.42       | 9.84       | 10.57      | 12.47      | 12.22      | 11.58      | 13.34      | 0.256   |
|               | SD 2.37    | 2.26       | 2.17       | 2.95       | 4.97       | 5.06       | 4.61       | 6.34       |         |
|               | n 6        | 6          | 6          | 6          | 6          | 6          | 6          | 6          |         |

LDH, lactate dehydrogenase (IU/L); CRP, C-reactive protein mg/dL; Ferritin (ng/L); Lymphocytes; D-clinr (ng/mL)
Discussion

• In our study, tocilizumab was started when IL-6 was ten times the upper limit of normal in the absence of any obvious bacterial or fungal infection as a marker of CRS.
• The mean IL6 in our patients was 205.26 and median 145.9.
• In our patients, there was male preponderance, and hypertension was the commonest associated comorbidity.
• The mortality at day 7 (p = 0.007) and day 28 (p = 0.001) and need for escalation of respiratory support (p = 0.001) were statistically significant lower in the patients who received tocilizumab vs standard treatment (Table 3, Fig. 1).
• Timely identification of CRS and administration of tocilizumab to the patients reduce the need for escalation of respiratory support such as invasive ventilation in these patients and hence reduced mortality.
• The ICU-LOS was higher in the tocilizumab group but was not statistically significant, which can be partly explained by the higher need for escalation in standard treatment group and early mortality.
• On reviewing the trends of inflammatory markers, only CRP showed a significant decline with the use of tocilizumab by day 3 (p = 0.033) as reported by other studies (Table 4 and Fig. 2).
• Hypercytokinemia is a hallmark of COVID-19; however CRS should be considered with SARS-CoV-2 infection only in cases of overly exuberant systemic inflammation leading to critical illness such as ARDS or MOF.
• The steroids were used in all our severe and critically ill patients for 7–10 days.
• In RECOVERY trial on COVID-19 patients, dexamethasone, statistically reduces the mortality by one-third in patients on mechanical ventilation (hazard ratio = 0.65, p = 0.003).
• The dramatic effect of dexamethasone in severe illness also proves the hypothesis that hyperinflammation significantly contributes to ARDS seen with COVID-19.
• The use of immunomodulator such as tocilizumab with specific action on IL6 and its possible synergistic action with steroids is likely responsible for good outcomes in our study.

The strength of this study-
• inclusion of only critically ill patients with IL6 value 10 times the upper limit of normal. (to separate the subset of CRS from hyperinflammation)
• The exclusion of secondary bacterial and fungal sepsis.

Limitations for our study-
• Retrospective cohort, small numbers of patients and effect of steroids on the outcomes.
• The effect size of outcome could have been exaggerated due to limited numbers of the patients in the study.

• Despite its limitations, this study proves use of simple pragmatic protocol for early identification of CRS and treatment with tocilizumab and steroids for 7–10 days can significantly improve the outcome in these patients.

**Conclusion**

• Tocilizumab can improve outcome by reducing the need for invasive ventilation and mortality when used timely in patients with CRS.

• We propose a double-blind randomized study involving a larger sample size and using IL6 and other inflammatory markers to evaluate its potential therapeutic role of tocilizumab in CRS seen with COVID-19.
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