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Determination role of some biomarkers tests for severe SARS-COV-2 infections in babylon province/IRAQ

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Abstract
In a series of 30 SARS-COV-2 infected patients whom clinically proven as severe pulmonary infection form. These were found with male/female ratio of 1:1. The age range of below 50 years old account for 60% and those of above 50 years old constitute the remaining 40%. They were the residents of Merjan Teaching Hospital/ Babylon Province/Iraq, to the period of March to April 2021 and primary screen by PCR for Sars-cov-2 RNA genes, in public health central laboratory found to be positive. The over- all laboratory investigation were; D –dimer, Ferritin, LDH, acute phase reactant C, and IL6. LDH was tempted to probe the immune mediated pulmonary tissue injury (367.48 U/L.), ferritin response may indicate hemolytic and acute phase reactant expressed as hyper-inflammation (331.1 ng/L.). |The D-dimer shed a light on the fibrino-lytic responses (6049 ng/L.) post to the immune-thrombotic overre- actions, where IL6 levels give a clue to the state of hyper-cytokininemia (171.92 pg/L.). The overall immune status of these patients was as; Hyper-inflammatory and immune overreaction. The inflammatory and immune herd plots were of skewed distribution types.

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1. Introduction
Sars-cov-2 infections in man may six forms [1]. Among which the pulmonary form that gains wide pandemic spread all over the world [2]. The duration of the infection can be of acute [2] or chronic [3,4]. As when different peoples contracts this infection have different disease settings as asymptomatic, mild, moderate and severe symptomatic disease [5]. Though, such range of disease setting variations, there an in-common diagnostic markers usable for laboratory investigation in addition to other markers devoted for severe cases [6]. The objective of the present work was to use some biomarkers to delineate severe infection state in this province and to deduce the immune state as well as to check for nature of the inflammatory and immune herd plots.

2. Materials and methods

2.1. Patients
The elected disease subjects were 30 to the period of March to April 2021 at Hillah Babylon Province. All of them checked for RNA genes of Sars-cov-2 virus by PCR and be positive in Babylon Central Public Laboratory. They were interviewed by respiratory disease specialists in Mergan Medical Teaching Hospitals with primary diagnosis of covid-19 and hospitalized initially in infectious disease unites. On need to ventilating settings for assisted respiration they referred to Intensive care unit (ICU). Actually they ICU resident patients. The blood samples were collected from the brachial vein without anticoagulant for the laboratory investigations [7].

2.2. Laboratory biology

For the determination of both D-dimer and Ferritin a French made vidas with the eligible accessories using their recommendation for serum application was tempted. Acute phase reactants C was determined using a Mispi2 Swiss made apparatus providing...
it with its special Accessories needed for serum application. LDH was estimated using a Chinese made Minividas. IL6 was done as per ELISA apparatus.

3. Results

3.1. Demographic

The Male-Female ratio was 1:1. The 30 patients were subgrouped into; One, 10, seven, five, one and one to the age ranges of; 20–29, 30–39, 40–49, 50–59, 60–69, 70–79 and 80–89 years old respectively. Patients whom ranged below 50 years old were accounting for 60%. While those whom above 50 years old constitute 40%.

3.2. Biomarkers

3.2.1. Pro versus inflammatory markers

One sixth of the 30 patients were tested for IL6. Four of five of which have shown IL6 cytokine ranging from 171 to 457 pg/L as compared to the normal value of 7 pg/L, Table 1.

3.2.2. Inflammatory markers

Other one sixth of the 30 patients were tested for the acute phase reactant C. They showed Levels of 11.7 to 96.8 mg/L as compared to the normal value of 6 mg/L. A case of Hyper inflammatory responses, Table 2.

LDH was investigated to explore the immune mediated pulmonary tissue damage in the test patients. It was ranged from 197 to 665 U/L as compared to the normal values of 140–280 U/L, Table 3.

3.2.3. Hyperferritnemia

Ferritin responses among these SARS-COV-2 infected patients indicates haemolytic and acute phase reactants expressed as hyper inflammation. They showed ferritin levels ranging from 126 to 1181.75 ng/L as compared to normal values of 25–350 ng/L in males and 13 to 233 ng/L in females, Table 3.

3.2.4. Immunothrombotic marker

One of the facets of the immune overreaction in severe sars-cov-2 infection is the immune-thrombotic reactions. D dimer is the accepted mapper to the sequela of the immune-thrombotic events. It was ranged between 510 and 6040 ng/L as compared to the normal value of 500 ng/L, Table 3.

3.3. Severity Index

The LDH, Ferritin and D-Dimer higher levels all indicating severity of sars-cov-2 infection among these patients, Table 3.

3.3.1. Patients response variations

Four patients showed both elevated CRP and D –dimer. Other four were showing both elevated LDH and D-dimer. Three have both elevated ferritin and D –dimer. Two were showing only elevated LDH response, and two other were with elevate Ferritin, LDH and D-dimer.

3.3.2. Immune herd plots

The D-Dimer and LDH herd plots represents the immune herd plots. While the ferritin indicates the hyper-inflammatory herd plots. The plots for D-dimer, LDH and ferritin were of skewed distribution types, Figs. 1-3.

4. Discussion

The SARS-COV-2 human infection have shown heterogeneous clinical presentation, and complex pathophysiology and wide range of biomarker response variation depending on the severity and the infection time course [8]. Severe infection is that risky or life-threatening infection. Such severity can be deduced by clinical symptoms and by laboratory biomarkers. A battery of; IL 6, CRP, LDH, ferritin and D –dimer the sounded generally agreed in practice for delineation of severe SARS-COV-2 human infections [9,10].

Ferritin is the key mediator of immune dysregulation as in the case of hyperferritinemia via direct immune suppressive and pro-inflammatory effects through hypercytokinemia [6,11]. During the virus infection, increased ferritin levels represent host defense mechanisms that deprive the growth of the pathogen and protect the immune cell function [12] and be a marker of severe pulmonary involvement [13]. Ferritin is also able to activate macrophage that take part in the innate immunity of the body. When the macrophage activated begin to secret cytokines. Cytokine secretion at high levels will cause hypercytokinemia syndrome [14]. The hyper ferritinemia and hypertcytokinemia seen among these 30 patients Tables 2 and 3. Becomes confirmation to these holdings. Interleukine 6 was an accepted biomarker for mapping hyper inflammation and hyper cytokinemia [6,15]. The acute phase reactants C is a general immune biomarker for inflammation [9].

LDH is a biomarker for various inflammatory states like infections and sepsis and it is elevated in SARS-COV-2 infected patients, Table 2 [15]. Szarpak et al. [16] have the holdings that LDH is a

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### Table 1

| Statistical features | Ferritin ng/L | LDH U/L | IL6 pg/L |
|---------------------|--------------|---------|----------|
| Minimum             | 282          | 168     | 2.1      |
| Maximum             | 1228         | 883     | 457      |
| Mean                | 507          | 253.4   | 171.92   |
| Median              | 1000         | 863     | 321      |
| Range               | 282–1228     | 168–883 | 2.1–457  |

### Table 2

| Statistics | Ferritin ng/L | LDH U/L | CRP mg/L | D-Dimer |
|------------|---------------|---------|----------|---------|
| Minimum    | 106.1         | 269     | 11.7     | 268     |
| Maximum    | 450           | 575     | 96.8     | 2512    |
| Mean       | 257.62        | 342     | 55.96    | 268.95  |
| Median     | 276           | 269     | 53.1     | 268     |
| Range      | 106.1–276     | 269–575 | 11.7–96.8| 268–2512|

### Table 3

| Statistical features | LDH U/L | D-Dimer ng/L | Ferritin ng/L |
|---------------------|---------|--------------|---------------|
| Minimum             | 197     | 510          | 126           |
| Maximum             | 668.33  | 2074.033     | 1181.75       |
| Mean                | 367.48  | 6049         | 331.1         |
| Median              | 605     | 1160         | 295.075       |
| Range               | 197–665 | 510–2074.033 | 126–1181.75   |
potential marker for vascular permeability in immune mediated lung injury. D –dimer is a degradation product of cross-linked fibrins resulting from plasmin cleavage. During the fibrinolysis plasmin may degrade fibrin monomers, cross-linked fibrin polymers and possibly fibrinogen during systemic fibrinolysis following alpha depletion. All these fragments are collectively called degradation products. D-dimer constitute two adjacent fibrin D domains. Because D –dimer is a product of cross-linked fibrin, it is considered as a sensitive biomarker to rule out venous thromboembolism [17]. D –dimer levels, Table 3, correlate, with severity and are relatively prognostic marker for the in hospital morbidity in patient admission for covid-19 [18,19].

The immunology of Sars-cov-2 infections is rather complex [20], with heterogeneous clinical presentation as well as complex pathophysiology and wide range of biomarkers variations as noted in ferritin, LDH and D –dimer results of our patients depending on the severity and the infection time course [8].

The inflammatory and immune herd plots noted in this study, the skewed plot types parallel with some other bacterial infectious disease and contradicts with others [21–24].

5. Conclusion

i- SARS-COV-2 human virus infections are being proved on laboratory basis.

ii- Patients responses to the infectious agent, the virus are heterogeneous.

iii- There were an individual and group wise variations in these responses.
iv-LDH, ferritin and D-dimer were found to be reliable test battery for diagnosing the severe infection forms together with identification of hyper ferritinemia. Hyperinflammation, elevated immune based thromboembolic lysis and hypercytokinemia.

v-Inflammatory and immune herd plots were found to be of skewed plot types.

CRediT authorship contribution statement

Ibrahim Ms. Shnawa: Writing – original draft. Rusul Hayder Alfatlawi: Software, Validation, Writing - review & editing. Assel Hashim Nemah: Conceptualization, Methodology. Ahmed S. Abed: Visualization, Investigation, Supervision.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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