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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 overreactions, where IL6 levels give a clue to the state of hyper-cytokinemias (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 Mispil2 Swiss made apparatus providing

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**Table 1**  
IL6 Hypercytokinemia among SARS-COV-2 severe infected patients.

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

it with its special Accessories needed for serum application. LDH was estimated using a Chinese made Minividus.IL6 was done as per ELISA apparatus.

### 3. Results

#### 3.1. Demographic

The Male-Female ratio was 1:1. The 30 patients were sub-grouped into; One, 10, seven, five, 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. Hyperferritinemia

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 sequelae 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](#).

**Table 2**  
Acute Phase Reactant C levels indicating hyper inflammatory responses among sars-cov-2 severe infected patients.

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**  
Sars-cov-2 Severity Index.

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

#### 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 elevate 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 secrete cytokines. Cytokine secretion at high levels will cause hypercytokinemia syndrome [14]. The hyper ferritinemia and hypercytokinemia 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

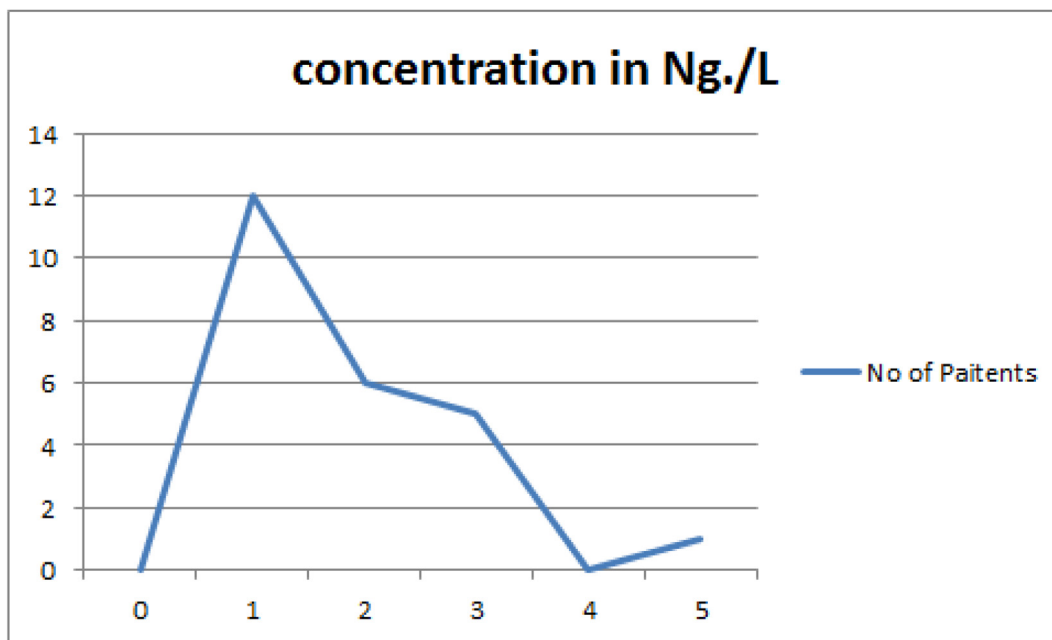


Fig. 1. D Dimmer Herd Plot.

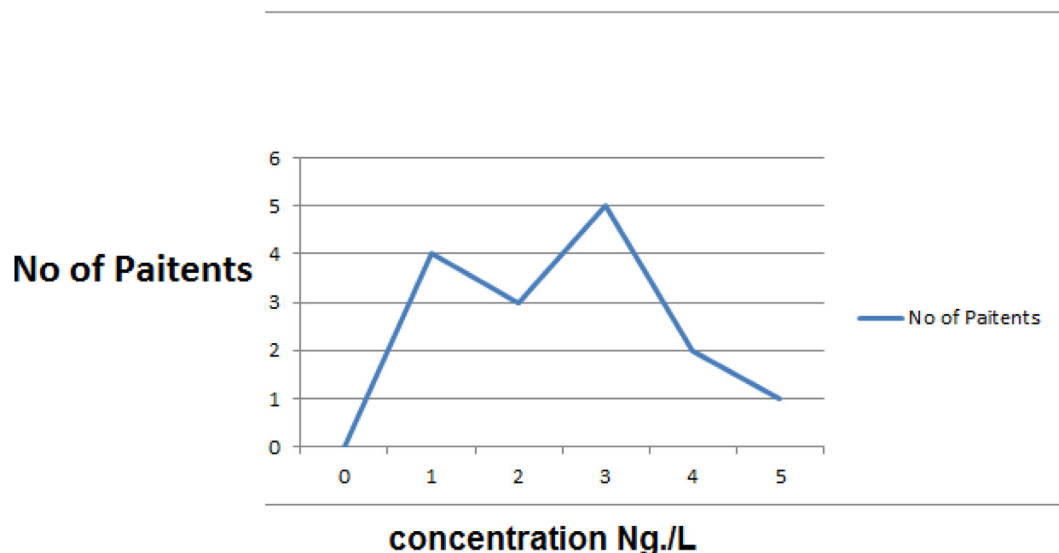


Fig. 2. Ferritin determination.

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.

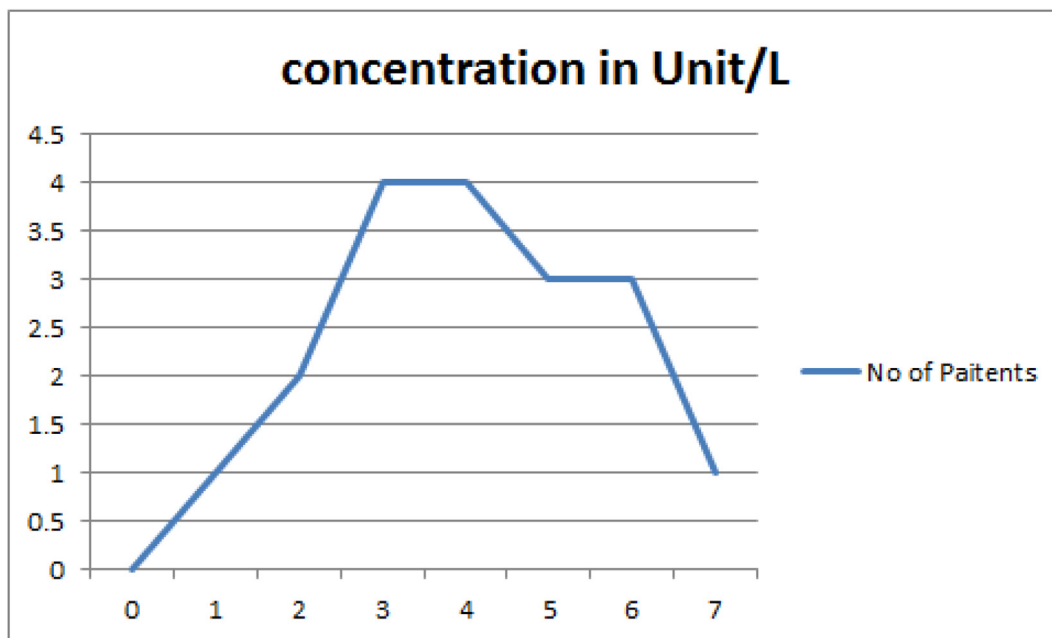


Fig. 3. LDH Level in study patients.

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 swed 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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#### References

- [1] C.H. Sudre, et al. Six distinct types of covid-19 identified, symptom cluster in covid-19: a potential clinical prediction tool from covid-19 symptom study group. doi.org/ 101191/ 2020.00.1220056.
- [2] B.W. Massey, K. Jayathilake, H.Y. Meltzer, Respiratory microbial co-infection with sars-cov-2, Front. Microbiol. 11 (2020), <https://doi.org/10.3389/fmicb.2020.02079>.
- [3] M.C. Dalakas, Gullain Barre syndrome. The first documented covid-19 triggered neurologic disease, Neurol. Neuroimmunol. Neuroinflammol. 7 (2020) e781, <https://doi.org/10.1212/NXL.000000000000781>.
- [4] M. Staff, Covid-19 long term effect, Myoclinic (2020).
- [5] V. Castelli, A. Cimini, C. Ferri. Cytokine storm in covid-19: when you come out of the storm, you would not be the same person who walked in. Front. Immunol. 2020.11. Article number 2132. doi.10. 3389.front. Immunol. 2020.02132.
- [6] Badawi, Hypercytokinemia and pathogen host interaction, J. Inflamm. Res. 13 (2020) 255–261.
- [7] C.D. Stevens, Clinical Immunology and Serology: A Laboratory Perspective, 3rd ed., F A Davis Company, Philadelphia, 2010.
- [8] F. Carrubi, L. Salvati, F. Maggi, E. Borghi, R. Mariuni, C. Ferri, et al. Ferritin is associated with the severity of lung involvement but not worse prognosis in patients with covid-19; data from two covid-19 units. Sci. Rep. le.2021. No.411; Article number 4863.
- [9] T. Greenhaigh, M. Knight, C.A. Court, M. Buxton, L. Husian. Management of post-acute covid-19 in primary care, BMJ 2020.370m30261. doi.10.1136/bmj.m3026.
- [10] E. Fraser. Long term respiratory complication of covid-19. BMJ. 2020. 30m3001. doi.1956 /bmjm3021.
- [11] M. Vargas-Vargas, C. Cotes-Rojo. Ferritin levels and covid-19. Revista. Panamerica de. Salud. Publica. 2020. Science. Open.Com.
- [12] K.F. Kernan, J.A. Carcillo, Hyperferritinemia and inflammation, Int. Immunol. 29 (9) (2017) 401–409.
- [13] C. Perricone, E. Bartoloni, R. Bursi, G. Cafaro, G.M. GuideLLi, Y. Shoenfeld, et al., Covid-19 of as a part of hyperferritinemia syndrome: the role iron depletion therapy, Immunol. Res. 68 (4) (2020) 413–424.
- [14] P. Ogorodinkova. High ferritin levels may indicate severe covid-19. EurekAlert 1.2020. Science News.
- [15] L. Szarpsk, K. Reuzier, K. Safiejko, M. Pruc, L. Kanezugo-Koda, K.J. Filipiak et al. Lactate dehydrogenase levels as covid-19 severity markers, Am. J. Emerg. Med. 2020 doi. 10.1016/j. ajem.20.11. e5.
- [16] E. Danese, M. Montagana. An historic approach to the diagnostic biomarkers of acute respiratory syndrome, Annal. Transl. Med. 4 (10) (2016) 194–196.
- [17] M.R. Gil, A. Lee, D. Sabath, C. Leissenger, O. Volod, G. Wool, et al. Covid-19 frequently asked questions. 2020. Covid-19 Q & A.
- [18] H. Yu, C. Qin, M. Chen, O. Wang, D.-S. Tian, D-dimer levels associated with severity of Covid-19, Throm. Res. (2020) 195–225.
- [19] Y. Yao, J. Cao, O. Wang, O. Shi, K. Liu, S. Hen, et al. D-dimer as biomarker for disease severity and mortality in covid-19 patients: a case control study, J. Inten. Care 8 2020 Article No.49.291k.
- [20] D.E. Speiser, M.E. Bachman. Covid-19. Mevhanisms of vaccination and immunity. Vaccines 8 2020 404 doi.10.3390/vaccines.8036404.
- [21] I.M. Shnawa, I.A. AL-janabi, A. AL-Mohanna, A.A. Buses, Functional Lapin-Human simulations of knee joint synovial mucosal cytokine responses in Staphylococcus aureus septic arthritis, Ind. J. Pub. Health. Res. Develop. 11 (3) (2020) 1242–1247.
- [22] I.M. Shnawa, B.H. AL-Amedei, The human rthropathy inflammatory and immune herd plots, Ind. J. Pub. Health. Res. Develop. 10 (9) (2019) 1067–1070.
- [23] S.B. Omer, L. Yildirim, H.P. Forman, Herd immunity and implications for sars-cov-2 control, JAMA 324 (20) (2020) 2095, <https://doi.org/10.1001/jama.2020.20892>.
- [24] Y. Zhou, N. Ding, G. Yang, W. Peng, F. Tang, C. Guo, et al. Serum lactate dehydrogenase level may predict acute respiratory distress syndrome of patients with fever infected with sars-cov-2. Annaals Transl. Med. 18(7) 2020 doi.10.21073:atm20-2411.