ORIGINAL ARTICLE ASIAN JOURNAL OF MEDICAL SCIENCES

Role of C-reactive protein, lactate dehydrogenase, Vitamin D, and interleukin-6 in the prognosis of COVID-19 – A retrospective study in a tertiary care hospital in Tamil Nadu

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Submission: 19-03-2022 Revision: 29-04-2022 Publication: 01-06-2022

ABSTRACT

Background: SARS-COV-2 is also known as coronavirus causes an infectious disease has caused a global concern due to its pandemic nature. In India, nearly 36 million cases and 5.1 lakh deaths have been recorded so far. Laboratory biomarkers play an important role in improving the clinical situational awareness by predicting risk stratification and by discriminating between severe and non-severe cases. There is no sufficient information available regarding the role of C-reactive protein (CRP), lactate dehydrogenase (LDH), Vitamin D, and interleukin-6 (IL-6) in the prognosis of COVID-19 patients in Southern part of India. **Aims and Objectives:** We aim to retrospectively analyze CRP, LDH, Vitamin D, and IL-6 in COVID-19 patients. **Materials and Methods:** The observational and retrospective study was carried out from the case records of 103 patients from November 2021 to February 2022 in which CRP, LDH, Vitamin D, IL-6, COVID-19 Reporting and Data System (CO-RADS), and CT sore were analyzed during admission and follow-up period in the hospital. Mean, mean difference, standard deviation, standard error difference, and P value were calculated. $P<0.05$ was considered to be significant. **Results:** CRP, LDH, IL-6, Vitamin D, CO-RADS, and CT score were decreased during the follow-up period when these levels were compared at the time of admission in the hospital, while there is an increase in Vitamin D levels during the follow-up period. **Conclusion:** The use of reliable and easy to use assays such as CRP, LDH, Vitamin D, and IL-6 plays an important role in the diagnosis and prognosis of COVID-19.

Key words: COVID-19; C-reactive protein; Interleukin-6; Lactate dehydrogenase; Vitamin-D

Access this article online

Website: http://nepjol.info/index.php/AJMS

DOI: 10.3126/ajms.v13i6.43913

E-ISSN: 2091-0576

P-ISSN: 2467-9100

INTRODUCTION

SARS-CoV-2 virus causes an infectious disease which is known as coronavirus.[1] Globally, over 229,000,000 cases and 470,5111 deaths have been recorded.[2] In India, over 335,000,00 cases and 446,000 deaths have been recorded.[3] Disease such as severe pneumonia, acute respiratory distress syndrome (ARDS), and multiorgan failure develops in a small proportion of patients although the majority of the patients with COVID-19 are asymptomatic or can develop mild influenza like virus.[4]

The pathogenesis of COVID-19 involves two phases. In Phase 1, there is a suppression of innate immune response which is increased in oxidative stress followed by hyperinflammation in Phase 2.[5] Based on recent studies, uncontrolled inflammation along with high viral load has contributed to the severity of COVID-19.[6,7]

Laboratory biomarkers play an important role in predicting risk stratification, in discriminating between severe and non-severe cases which plays an important role in improving the clinical situational awareness.[8]

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C-reactive protein (CRP), a biomarker of inflammation, is synthesized in liver in response to interleukin-6 (IL-6).[9] Recent studies have reported an association between higher CRP levels and severity of COVID-19.[10] Due to its influence on several immune pathways, Vitamin D plays an important role in reducing the risk of viral respiratory infections.[11] According to the available data, increased lactate dehydrogenase (LDH) levels are seen in patients with COVID-19.[8]

Since the available information is insufficient in this part of the country, we did a retrospective study in a designated COVID-19 tertiary care center in Tamil Nadu, South India, on the role of CRP, LDH, Vitamin D, and IL-6 in the prognosis of COVID-19.

Aims and objectives

To analyze the role of CRP, LDH, Vitamin D, and IL-6 in COVID-19 patients.

MATERIALS AND METHODS

The medical records of COVID-19 patients admitted from November 2021 to February 2022 were retrospectively analyzed in a COVID-19 designated tertiary care hospital in Tamil Nadu, South India and deidentified data were obtained after getting approval from the Institutional Ethics Committee. The case records containing insufficient data and without any blood investigations were excluded from the study.

Case records of 103 COVID-19 infected patients were retrospectively analyzed for the following parameters, namely, CRP, LDH, Vitamin D, and IL-6. Patient characteristics (age and sex), the CO-RADS, and CT score were also analyzed. Independent t-test was employed for the statistical significance between the observed parameters. The obtained data were analyzed using SPSS software version 25.0.

RESULTS

Out of 103 case records of hospitalized COVID-19 patients' percentage of male was 66.01% and that of female was 33.98%. In age-wise distribution, significant proportion (49.51% of patients) was in the age group of 26–50 years followed by (34.95%) patients in the age group of 51–75 years. There was a decrease in the mean value of COVID-19 Reporting and Data System (CO-RADS) (1.30±0.46) during follow-up when compared with the mean value of (4.76±1.34) during admission of the hospitalized COVID-19 patients. The mean value of CT score (10.33±3.72) during admission decreased to (6.17±1.42) during follow-up of the patients. The mean value of SpO_2 which was (95%±0.05) during admission improved to (97%±0.008) (Table 1).

The mean value of CRP which was (22.72±6.74) during admission decreased to (12.31±15.85) during follow-up. There was a decrease in the mean value of LDH levels from (466.33±161.73) during admission to (253.31±96.08) during follow-up of the patients. There was an increase in the mean value of Vitamin D levels from (29.89±18.09) during admission to (37.88±12.47) during the follow-up of the patients while there was a decrease in the mean value of IL-6 from (56.36+80.96) to (10.41±15.57) during the follow-up of the patients (Table 2).

In independent T test employed for CRP levels during the admission and follow-up of the patients, the P value (0.05) was found to be significant (Table 3).

In independent T test employed for LDH levels during the admission and follow-up of the patients, the P value (0.05) was found to be significant (Table 4).

In independent T test employed for Vitamin D levels during the admission and follow-up of the patients, the P value (0.01) was found to be significant (Table 5).

Table 1: Base line characteristics of patients during admission and follow-up

Variable	COVID-19 patients			
Demographics				
Total patients	103			
Male	68 (66.01%)			
Female	35 (33.98%)			
Age range				
0–25	9			
26–50	51 (49.51%)			
51–75	36 (34.95%)			
>75	7			
Clinical findings	**During admission**		**During follow-up**	
	Mean	**SD**	**Mean**	**SD**
CO-RADS	4.76	1.34	1.30	0.46
CT-Score	10.33	3.72	6.17	1.42
SpO_2	95%	0.05	97%	0.008

CO-RADS: COVID-19 reporting and data system

Table 2: Laboratory findings of patients during admission and follow-up

Laboratory findings	During admission		During follow-up	
	Mean	**SD**	**Mean**	**SD**
CRP	22.72	6.74	12.31	15.85
LDH	466.33	161.73	253.31	96.08
VITAMIN-D	29.89	18.09	37.88	12.47
IL-6	56.36	80.96	10.41	15.57

CRP: C-reactive protein, LDH: Lactate dehydrogenase, IL-6: Interleukin-6

In independent T test employed for IL-6 levels during the admission and follow-up of the patients, the p value (0.05) was found to be significant (Table 6).

DISCUSSION

In our study, we retrospectively analyzed the role of CRP, LDH, Vitamin D, and IL-6 from the case records of COVID-19 infected patients. From the results of our study, we found out that there is a marked decrease in the levels of CRP, LDH, IL-6, and an increase in the levels of Vitamin D during the follow-up period of COVID-19 infected patients.

CRP an acute phase reactant enhances phagocytosis and facilitates clearance by binding to phosphocholine in pathogens and membrane of host cells.[12] Recently, reports have shown that increase in CRP concentrations in COVID-19 patients has a five-fold greater risk of acquiring ARDS than those with lower CRP values.[13,14]

LDH is an intracellular enzyme and has five separate isoenzymes among which LDH-3 is present in pneumocytes. Greater amounts of LDH can be expected to be released in the circulation of the patients with severe COVID-19 infections, since LDH is present in lung tissue. According to the previous studies, elevated LDH levels play an important role in influencing the clinical outcomes in patients with COVID-19 as it reflects the multiple organ injury and failure.[15,16]

In our study, there is an elevation in CRP and LDH levels in COVID-19 infected patients on admission to the hospital and it markedly decreased during the follow-up period in the hospital.

From the previous studies, it has been reported that there is a decrease in Vitamin D levels in COVID-19 infected patients when compared with COVID-19 negative patients which is suggestive of a relationship between Vitamin-D levels and COVID-19 positivity.[17,18]

The previous experimental studies have demonstrated that activation of Vitamin D in the lung tissues has a preventive effect on interstitial pneumonia.[19]

The possible role of Vitamin D is characterized by the inverse association between Vitamin D and IL-6. IL-6 which is crucial in regulating the inflammatory response has been related to the severity and prognosis of COVID-19.[20,21]

IL-6 plays an important role in cell proliferation and differentiation in addition in controlling the immune response. Since it is elevated in many inflammatory diseases, its control plays an important role during the course of the disease.[22,23] Our results showed that there is an increase

Table 3: Independent samples test of CRP

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. 2-tailed	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
0	Equal variances assumed	49.52	0.05	6.13	204	0.00	10.40	1.69	7.05	13.75
	Equal variances not assumed			6.13	137.74	0.00	10.40	1.69	7.05	13.76

CRP: C-reactive protein

Table 4: Independent samples test for LDH

		Levene's Test for Equality of Variances		t-test for Equality						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
0	Equal variances assumed	19.96	0.05	11.49	204	0.00	213.02	18.53	176.48	249.57
	Equal variances not assumes			11.49	166	0.00	213.02	18.53	176.43	249.62

LDH: Lactate dehydrogenase

Table 5: Independent samples test for Vitamin D

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	Df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
0	Equal variances assumed	6.32	0.01	−3.68	204	0.00	−7.98	2.16	−12.25	−3.71
	Equal variances not assumed			−3.689	18	0.00	−7.98	2.16	−12.26	−3.71

Table 6: Independent samples test of IL-6

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
0	Equal variances assumed	32.33	0.05	5.65	204	0.00	45.94	8.12	29.93	61.96
	Equal variances not assumes			5.65	109.54	0.00	45.94	8.12	29.84	62.05

IL-6: Interleukin-6

in IL-6 levels during admission of COVID-19 infected patients, followed by decrease in the levels of IL-6 during the follow-up period which was in accordance with the previous studies conducted.[24,25]

CO-RADS is based on the features of unenhanced chest CT for the pulmonary involvement in COVID-19 patients. It is categorized into very low (category I) to very high (category V). We analyzed the CO-RADS of COVID-19 positive patients in our study in which there is a decrease in the score of CO-RADS of the patients during the follow-up period when compared with the score of CO-RADS during admission.[26]

In our study, we also analyzed the CT score of the COVID-19 patients during admission and follow-up in which there is a decrease in the CT score of the patients during the follow-up period which was in accordance with the previous studies.[27]

Limitations of the study

Large scale studies are needed to confirm these findings as the study is a retrospective study and the sample size is small.

CONCLUSION

The rapid spread of COVID-19 especially in developing countries is a great concern. The use of reliable and easy-to use assays plays an important role in diagnosis of COVID-19. In our study, there is an elevation in the levels of CRP, LDH, and IL-6 patients and there is a decrease in the levels of Vitamin D levels during admission of COVID-19 patients, followed by decrease in the levels of CRP, LDH, and IL-6, increase in Vitamin D levels during follow-up of COVID-19 patients. Therefore, these biomarkers can be used a diagnostic as well as prognostic tool in COVID-19 patients as it is easy to use and reliable.

ACKNOWLEDGMENT

We thank all the individuals included in the study

REFERENCES

1. Available from: https://www.who.int/health-topics/coronavirus#tab=tab_1 [Last Accessed on 2022 May 05]
2. Available from: https://covid19.who.int [Last Accessed on 2022 May 05]
3. Available from: https://www.worldometers.info/coronavirus/country/india [Last Accessed on 2022 May 05]
4. Chen N, Zhou M, Dong X, Qu J, Gong F, Han Y, et al. Epidemiological and clinical characteristics of 99 cases of 2019novel coronavirus pneumonia in Wuhan, China: A descriptive study. Lancet. 2020;395(10223):507-513. https://doi.org/10.1016/S0140-6736(20)30211-7
5. Mrityunjaya M, Pavithra V, Neelam R, Janhavi P, Halami PM and

Ravindra PV. Front Immunol. 2020;11:570122.
https://doi.org/10.3389/fimmu.2020.570122

6. Merad M and Martin JC. Pathological inflammation in patients with COVID-19: A key role for monocytes and macrophages. Nat Rev Immunol. 2020;20(6):355-362.
https://doi.org/10.1038/s41577-020-0331-4
7. Vabret N, Samstein R, Fernandez N, Merad M and Sinai Immunology Review Project; Trainees; Faculty. Advancing scientific knowledge in times of pandemics. Nat Rev Immunol. 2020;20(6):338.
https://doi.org/10.1038/s41577-020-0319-0
8. Henry BM, de Oliveira MH, Benoit S, Plebani M and Lippi G. Hematologic, biochemical and immune biomarker abnormalities associated with severe illness and mortality in coronavirus disease 2019 (COVID-19): A meta-analysis. Clin Chem Lab Med. 2020;58(7):1021-1028.
https://doi.org/10.1515/cclm-2020-0369
9. Morley JJ and Kushner I. Serum C-reactive protein levels in disease. Ann N Y Acad Sci. 1982;389:406-418.
https://doi.org/10.1111/j.1749-6632.1982.tb22153.x
10. Petrilli CM, Jones SA, Yang J, Rajagopalan H, O'Donnell L, Chernyak Y, et al. Factors associated with hospital admission and critical illness among 5279 people with coronavirus disease 2019 in New York City: prospective cohort study. BMJ. 2020;369:m1966.
https://doi.org/10.1136/bmj.m1966
11. Parlak E, Ertürk A, Çağ Y, Sebin E and Gümüşdere M. The effect of inflammatory cytokines and the level of Vitamin D on prognosis in Crimean-Congo hemorrhagic fever. Int J Clin Exp Med. 2015;8(10):18302-18310.
12. Volanakis JE. Human C-reactive protein: Expression, structure, and function. Mol Immunol. 2001;38(2-3):189-197.
https://doi.org/10.1016/s0161-5890(01)00042-6
13. Wu C, Chen X, Cai Y, Xia J, Zhou X, Xu S, et al. Risk factors associated with acute respiratory 0.distress syndrome and death in patients with coronavirus disease 2019pneumonia in Wuhan, China. JAMA Intern Med. 2020;180(7):934-943.
https://doi.org/10.1001/jamainternmed.2020.0994
14. Goyal P, Choi JJ, Pinheiro LC, Schenck EJ, Chen R, Jabri A, et al. Clinical characteristics of Covid-19 in New York city. N Engl J Med. 2020;382(24):2372-2374.
https://doi.org/10.1056/NEJMc2010419
15. Martinez-Outschoorn UE, Prisco M, Ertel A, Tsirigos A, Lin Z, Pavlides S, et al. Ketones and lactate increase cancercell "stemness," driving recurrence, metastasis and poor clinical outcome in breastcancer: Achieving personalized medicine via metabolo-genomics. Cell Cycle. 2011;10(8):1271-1286.
https://doi.org/10.4161/cc.10.8.15330
16. Erez A, Shental O, Tchebiner JZ, Laufer-Perl M, Wasserman A, Sella T, et al. Diagnostic and prognostic value of very high serum lactate dehydrogenase in admitted medical patients. Isr Med Assoc J. 2014;16(7):439-443.
17. Kaufman HW, Niles JK, Kroll MH, Bi C and Holick MF. SARS-CoV-2 positivity rates associated with circulating 25-hydroxyvitamin D levels. PLoS One. 2020;15(9):e0239252.
https://doi.org/10.1371/journal.pone.0239252
18. Meltzer DO, Best TJ, Zhang H, Vokes T, Arora V and Solway J. Association of Vitamin D status and other clinical characteristics with COVID-19 test results. JAMA Netw Open. 2020;3(9):e2019722.
https://doi.org/10.1001/jama networkopen.2020.19722
19. Tsujino I, Ushikoshi-Nakayama R, Yamazakj T, Matsumoto N and Saito I. Pulmonary activation of Vitamin D3 and preventive effect against interstitial pneumonia. J Clin Biochem Nutr. 2019;65(3):245-251.
https://doi.org/10.3164/jcbn.19-48
20. McGonagle D, Sharif K, O'Regan A and Bridgewood C. The role of cytokines including interleukin-6 in COVID-19 induced pneumonia and macrophage activation syndrome-like disease. Autoimmun Rev. 2020;19(6):102537.
https://doi.org/10.1016/j.autrev.2020.102537
21. Cummings MJ, Baldwin MR, Abrams D, Jacobson SD, Meyer BJ and Balough EM. Epidemiology, clinical course, and outcomes of critically ill adults withCOVID-19 in New York city: A prospective cohort study. Lancet. 2020;395(10239):1763-1770.
https://doi.org/10.1016/S0140-6736(20)31189-2
22. Heinrich PC, Castell JV and Andus T. Interleukin-6 and the acute phase response. Biochem J. 1990;265(3):621-636.
https://doi.org/10.1042/bj2650621
23. Kumar H, Kawai T and Akira S. Pathogen recognition by the innate immune system. Int Rev Immunol. 2011;30(1):16-34.
https://doi.org/10.3109/08830185.2010.529976
24. de Jong MD, Simmons CP, Thanh TT, Hien VM, Smith GJ, Chau TN, et al. Fatal outcome of human influenza A (H5N1) is associated with high viral load and hypercytokinemia. Nat Med. 2006;12(10):1203-1207.
https://doi.org/10.1038/nm1477
25. Wang W, Ye L, Ye L, Li B, Gao B, Zeng Y, et al. Up-regulation of IL-6 and TNFalpha induced by SARS-coronavirus spike protein in murine macrophages via NFkappaB pathway. Virus Res. 2007;128(1-2):1-8.
https://doi.org/10.1016/j.virusres.2007.02.007
26. Prokop M, van Everdingen W, van Rees Vellinga T, van Ufford HQ, Stöger L, Beenen L, et al. CO-RADS: A categorical CT assessment scheme for patients suspected of having COVID-19 definition and evaluation. Radiology. 2020;296(2):E97-E104.
https://doi.org/10.1148/radiol.2020201473
27. Francone M, Iafrate F, Masci GM, Coco S, Cilia F, Manganaro L, et al. Chest CT score in COVID-19 patients: Correlation with disease severity and short-term prognosis. Eur Radiol. 2020;30(12):6808-6817.
https://doi.org/10.1007/s00330-020-07033-y

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SJ – Concept and design of the study and prepared first draft of manuscript; **AG –** Interpreted the results and reviewed the literature and manuscript preparation; and **PK –** Concept, coordination, statistical analysis and interpretation, preparation of manuscript, and revision of the manuscript.

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Source of Support: Nil, **Conflicts of Interest:** None declared.