Journal Homepage: - www.journalijar.com

INTERNATIONAL JOURNAL OF ADVANCED RESEARCH (IJAR)

Article DOI: 10.21474/IJAR01/17784
DOI URL: http://dx.doi.org/10.21474/IJAR01/17784

RESEARCH ARTICLE

HOSPITAL BASED RETROSPECTIVE STUDY ON COVID-19 IN ANANTHAPUR, ANDHRA PRADESH

Dr. V. Aruna[1], Dr. T. Durga[2] and Dr. T. Amulya[3]

1. Associate Professor of Biochemistry, GMC, Machilipatnam, Andhra Pradesh, India.
2. Professor & HOD, Dept of Biochemistry, GMC, Ananthapuram, AP, India.
3. Postgraduate of Biochemistry, GMC, Ananthapuram, AP, India.

Manuscript Info

Manuscript History
Received: 28 August 2023
Final Accepted: 30 September 2023
Published: October 2023

Key words:-
COVID -19, LDH, CRP, Ferritin, D-Dimer

Abstract

Aim : To study the role of inflammatory markers in the diagnosis and prognosis of Covid-19. To understand correlation of Inflammatory markers in RTPCR +ve Covid-19 subjects

Study design: Retrospective study was conducted on Covid-19 cases admitted in Government Geeneral Hospital , Ananthapuram from June - August 2021.

Material and Methods: The case records were collected from medical records section after obtaining permission from authorities. RTPCR positive case records were included for the study. Case records with past history of heart, kidney or liver disease were excluded. Laboratory data belonging to 300 individuals was collected. Among the cases 143 were males and 157 were females.Acute phase inflammatory markers , LDH, CRP, Ferritin and D-dimer , evaluated for Covid-19 patients were gathered from 300 case records. FBS was noted to discriminate diabetes status of all individuals.

Results: Based on CRP values the subjects were divided into mild (0-26mg/L) , moderate (27-99 mg/L) and severe (>100mgs/L). LDH,CRP and Ferritin showed sharp rise from the normal in all subjects. The same was not detected in case of D-dimer. Statistical analysis expressed as Mean ±SD(Standard Deviation) for all biochemical parameters . One-way ANOVA(Analysis of Variance) of LDH.CRP, Ferritin and D-dimer of entire study group showed significant correlation , P-value <0.00001(Table 2) In moderate and severe cases based on CRP frequency peaked at 40 whereas in mild cases it was around 60. 68♂ and 55♀ were found to be diabetic . The inflammatory markers did not show much difference between males & females or in the nondiabetic community. ANOVA of means of CRP ,Ferritin in diabetics was significant <0.00001 and that of LDH , D-dimer followed suit. To understand the correlation of CRP and Ferritin in mild and severe cases based on CRP ,ANOVA was performed. It was statistically significant with P value <0.00001.(Table 5). Frequency distribution of cases based on ferritin values showed 72.6% have high circulating ferritin and are prone to coagulopathy.

Conclusion: Retrospective studies help us understand the outcome of disease based on biochemical parameters. Similar study , meta-analysis

Corresponding Author:- Dr. V. Aruna
Address:- Associate Professor of Biochemistry, GMC, Machilipatnam, Andhra Pradesh, India.

can be done by obtaining information from all tertiary care centres in the state and correlate information for better knowledge.

……

Introduction:-

The COVID-19 virus is a new virus linked to the same family of viruses as Severe Acute Respiratory Syndrome (SARS) and some types of common cold.The virus is transmitted through direct contact with respiratory droplets of an infected person (generated through coughing and sneezing), and touching surfaces contaminated with the virus.Symptoms include fever, cough and shortness of breath. In severe cases, infection caused pneumonia or severe acute respiratory syndrome(SARS).

The WHO declared the coronavirus disease (COVID-19) as a global pandemic on March 11, 2020[1].In India, the disease was first detected on January 30, 2020, in Kerala in a student who returned from Wuhan [2,3]. The total (cumulative) number of confirmed infected people is more than 44,949,671 to date (May , 2023) across India.Number of deaths in India as on May 2023 was 5,33,293 as on 31st October 2023. [3A]. In advertent exposure, advanced age , comorbid conditions like diabetes mellitus were considered leading causes of life threatening complications secondary to upper respiratory infection. Severity of the disease was identified by arterial blood gas analysis and capillary Oxygen saturation levels (through pulse-oxymeter). Inflammatory parameters evaluated in covid-19 cases were Lactate Dehydrogenase (LDH), C - reactive protein(CRP),Ferritin and D-dimer.

LDH is a glycolysis enzyme and it is released from all tissues upon damage to cytoplasmic membrane by infections. C-reactive protein (CRP) is acute-phase protein appears in blood within 6–10 hours of any tissue damaging event and has a plasma half-lifeof 19 hours. It is synthesized by liver. Ferritin is an iron storage form; it is one of the bio - markers of inflammation. In bacterial or viral infection,an increase in ferritin occurs due to cytokine storm such as in covid -19 infection .[4] and to a decline in its transport capacity due to spleen and liver damage. D-dimer is fibrin degradation product and when it's levels are elevated , indicates thromboembolic phenomenon and covid-19 is procoagulant condition[5].

Inclusion Criteria

In this retrospective study data from Government General Hospital ,Anathapuram was analysed to assess correlation of D-dimer, ferritin , CRP & LDH with morbidity and mortality of Real -time PCR (RT PCR) positive covid-19 cases. Other details recorded include renal parameters – Blood urea, serum creatinine , hepatic profile- total bilirubin, SGOT & SGPT, Diabetic profile – FBS and Hypertension . In most cases the presenting symptom was respiratory distress but few have fever, sore throat.

Exclusion criteria

Individuals with heart disease like CAD, known cases of renal disease or cirrhosis of liver or any malignant disorder were excluded .Incomplete case records and records belonging to individuals below 18yrs were excluded from the study.

Materials and Methods:-

Study design and procedure

Data was collected from medical records section of Government General Hospital,Anathapuram after obtaining permission from IEC. This is retrospective study pertaining to SARS-COVID-19 pandemic from June 2021 to August 2021.

Statistical analyses

The data was collected from approximately 300 case records. Continuous variables were expressed as Mean±SD(Standard deviation). Descriptive statistics were done for marker parameters to understand severity of the disease and it's correlation to outcome . Influence of comorbid conditions was studied using Pearson's correlation coefficient. One way ANOVA of LDH,CRP, D-dimer & Ferritin was done to understand correlation of these parameters.

Results:-

All the individuals were admitted between June -August 2021. Normal range considered for LDH – 200-400 IU/L, for CRP < 0.3mgs/L, for Ferritin 12 – 150 ng/ml and for D-dimer <0.5mg/L. Results were expressed as Mean±SD for continuous variables . The probability value of <0.05 at 95% confidence interval considered statistically significant.

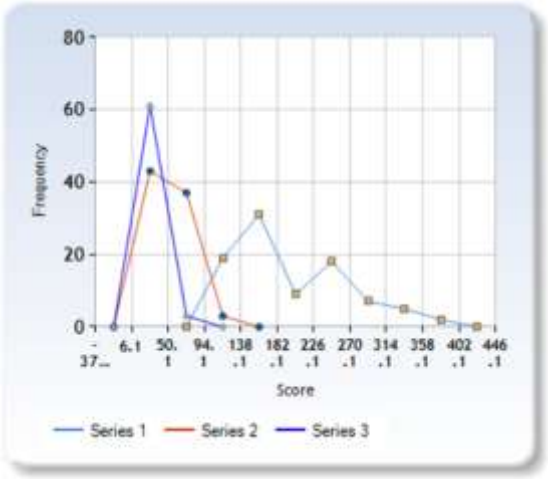
Table 1Meean ± SD of Biochemical parameters in study group

	Parameter						
	LDH	CRP	FERRITIN	D-DIMER	Total Bilirubin	SGOT	SGPT
Normal range	200-400 IU/L	<0.3mg/L	12-150 mg/ml	<0.5mg/L	0.2-1.2mg/dl	15-40IU/L	13-35IU/L
N	300	300	300	300	300	300	300
Mean	393.094	78.056	382.956	1.7702	0.7	28.56	44.37
S.D.	±355.84	± 89.25	±259.61	± 2.33	± 0.24	±0.165	± 2.86

N = Number of subjects

We observed mean ± SD of LDH,CRP, Ferritin and D-dimer in the study group was moderately elevated. The individuals were grouped into mild, moderate and severe based on the CRP values 7-26mg/L, 27-99mgs/L and >100mg/L respectively .

Figure 1:- Frequency polygon of CRP.



Series 1 >100

Series 2 26-99

Series 3 7- 26

Frequency of CRP levels in moderate and severe groups was clustered around 40 whereas mild elevation of CRP showed a peak frequency of approx. 60 indicating all the RTPCR positive individuals have elevated CRP values.

Table 2:- Oneway Anova Of Ldh,Crp, Ferritin & D-Dimer In All Subjects.

	LDH	CRP	FERRITIN	D-DIMER
Number of subjects	300	300	300	300
Mean±SD	393.094±355.84	78.056±89.25	382.96±259.61	1.77±2.33
Results				
Source	**SS**	**df**	**MS**	
Between-treatments	33766522.4249	3	11255507.475	F = 209.72727
Within-treatments	60268438.2592	1123	53667.3537	
Total	94034960.6841	1126		p- < 0 .00001

One – way ANOVA of mean ± SD of LDH,CRP, Ferritin and D-dimer of the study group (N=300) showed F-ratio of 209.72727 and P value of <0.00001 which is statistically highly significant(Table 2).

Out of 300 subjects 123(68 ♂ and 55 ♀) were diabetics and 143(75 ♂ and 68 ♀) have FBS within normal limits. 66 persons were not diabetic at admission , 1week after admission recorded elevated sugar values. The mean ± Standard Deviation (SD) of diabetics and nondiabetics was shown in table 2 gender wise.

Table 3:- Mean±SD of LDH,CRP,Ferritin and d-dimer in ♂ and ♀ diabetic and nondiabetic subjects.

	DIABETIC SUBJECTS			
	LDH	CRP	FERRITIN	D-DIMER
MALE (N= 68)	417.62 ± 270.83	77.0±79.84	408.15±260.41	1.92 ±2.52
FEMALE(N =55)	434.6 ± 659.075	69.68±72.22	356.63±242.14	1.34 ±1.585
NONDIABETIC SUBJECTS				
MALE (N=75)	384.92±219.19	64.54±87.59	386.72±262.57	1.42 ± 2.11
FEMALE (N =68)	389.91± 256.87	89.38±99.26	398.54±250.4	1.78 ± 2.22

In Table 3 mean ± SD values of LDH,CRP, Ferritin and D-dimer were shown to be 417.62 ± 270.83,77.0±79.84,408.15±260.41,1.92 ± 2.52 respectively in males with diabetes mellitus. Similarly in females with Diabetes mellitus the values were 434.6 ± 659.075, 69.68±72.22, 356.63±242.14, and 1.34 ±1.585. Significant difference was not observed observed between males and females having comorbid condition. The final outcome was not influenced by the presence of comorbid disease in our study, as death toll was negligible compared to total number of admissions.

Table 4:- One way ANOVA of CRP , Ferritin and LDH , D-Dimer in diabetics and non-diabetics, male Vs Female.

	CRP	**FERRITIN**	**F-RATIO**	**P-VALUE**
DIABETIC ♂ (68)	77.0±79.84	408.15±260.41		
NONDIABETIC ♂ (75)	64.54±87.59	386.72±262.57	67.506	< 0.00001
DIABETIC ♀ (55)	69.68±72.22	356.63±242.14		
NONDIABETIC ♀ (68)	89.38±99.26	398.54±250.4	53.834	< 0.00001
DIABETIC ♂ Vs ♀	77.0±79.84 (M) 69.68±72.22(F)	408.15±260.41(M) 356.63±242.14(F)	57.652	< 0.00001
	LDH	**D-DIMER**		
DIABETIC ♂(68)	417.62 ± 270.83	1.92 ± 2.52		
NONDIABETIC ♂(75)	384.92±219.19	1.42 ± 2.11	126.62145	< 0.00001
DIABETIC ♀ (55)	434.6 ± 659.075	1.34 ±1.585		
NONDIABETIC ♀ (68)	389.91± 256.87	1.78 ± 2.22	29.84428	< 0.00001
Statistically highly significant				

Analysis of Variance (ANOVA) of mean±SD of CRP and Ferritin of subdivisions in the study group like diabetics males (N=68) Nondiabetics males (N=75) and Diabetic females (N=55) , Nondiabetic females (N=68) was statistically significant with p value <0.00001 respectively. Similarly ANOVA of LDH and D-dimer showed F-ratio of 126.621(P value <0.00001) and 29.84428 (P value < 0.00001) for diabetic /nondiabetic males and females respectively (Table 4). Whereas ANOVA of CRP and Ferritin of Diabetic males and females showed F- ratio to be 57.652 (P value <0.00001). These findings correlate with the study of Liu **et al**[6]. Elevated CRP levels were confirmed biomarkers for severity of COVID-19 and the same was stated by other studies[7,8, 9]

Based on CRP values the cases were divided into 2 groups , group 1 CRP > 100 mg/L(N =94) and group 2 CRP < 100mgs/L (N = 68).Pearson's correlation coefficient was done to to study the effect CRP on Ferritin in both groups . The r value for group 1 was 0.1698 and P-Value < 0.1018 and the same for group 2 was 0.2147 P-Value < 0.0787. CRP being acute inflammatory marker was elevated in all COVID-19 cases ,but the same did not modify ferritin values either in mild or severe cases. Between the groups based on CRP one-way ANOVA showed F-ratio 120.95026 p-value < 0 .00001, significant at p< .05.(Table 5)

Table 5:- One-way ANOVA of CRP,Ferritinin mild and severe cases.

	GROUP 1 (CRP >100)		Group 2 (CRP<100)	
	CRP	FERRITIN	CRP	FERRITIN
Number	94	94	68	68
Mean± SD	192.25±72.01	496.97±210.69	48.83±19.28	416.925±248.72
Results				

	SS	df	MS	F-ratio
Regression	9956043.342	3	3318681.114	
Residual	8780286.4258	320	27438.3951	120.95026
Total	18736329.7677	323	p-value < 0.00001	

This indicates severity of infection correlates with high CRP values and the same was proven by other studies.[10,11]

In this study Ferritin values were classified into 3 categories viz. deficient , normal and high and frequency distribution was calculated as shown in Table 6. Elevated Ferritin was detected in 72.67% of cases and most the individuals were on ventilatory support. ((Izcovich et al., 2020) Similar study was done by Nabaz Fisal Shakir Agha et al in 2020 and they found ferritin in the normal range in 60.3% of cases.

Table 6:- Frequency Distribution Tablefor Ferritin.

Class	Count	Percentage
0 – 14 mgs/dl Deficiency	04 (1st Qtr)	1.3%
15 150mgs/dl Normal	78 (2nd Qtr)	26%
Above 150mgs/dl High	218 (3rd Qtr)	72.67%

Discussion:-

In the retrograde analysis of data from 300 medical records from Government General Hospital ,Anathapuram we found very high levels of CRP and mild elevation of ferritin levels in all infected individuals ,but LDH and D-dimer were moderately elevated. We found elevated CRP was neither related to disease severity or mortality. 0.2% is case fatality rate in Andhra Pradesh during 2021.[12]. We observed 2-fold rise in ferritin levels and 72.6% individuals showed definite rise (Table 6) which is consistent with the study of Zhou **et al**.[13]. The mean D-dimer levels in the study group, males, females, associated with diabetes in males and females were 1.77±2.23, 1.42 ± 2.11, 1.78 ± 2.22, 1.92 ± 2.52 and 1.34 ±1.585. The values were around to be 1.7 in males but in females there was drop by 0.4mg/L indicating presence of comorbid condition or disease severity did not influence it's concentration. In a study done by Zhang L raised D-dimer levels were observed in clinically worsening individuals.[14]. In a retrospective study conducted by Raja, Sangeetha et al in TamilNadu high D-dimer levels (4.62 ± 3.28) were associated with clinically worsening Covid-19 disease. In diabetic males & females CRP, Ferritin and LDH, D-dimer showed statistically significant correlation as shown in table 4 p-value of <0.00001. In several studies it was ascertained that covid -19 induces Diabetes Mellitus(DM) and also worsens preexisting DM because of cytokine storm, stress, cytotoxic destruction of β-cells of pancreas or drugs (Steroids) . [15]

We found elevated LDH, CRP and Ferritin levels in all cases and in both males and females as well as in diabetics and nondiabetics. These elevated levels were not related to disease outcome. Clinically disease severity and prognosis was evaluated during pandemic. Death rate in Andhra Pradesh in 2020 was 0.9% and the same dropped to 0.7% in April -August 2021.Means the data obtained from case records represents general scenario of SARS COVID -19 in the state of Andhra Pradesh.

Conclusions:-

Combination of routine laboratory biomarkers (CRP, LDH and ferritin ±D dimer) can be used to predict the diagnosis of viral infections. Few studies included Interleukins, Tumor Necrosis Factors, Immunoglobulin G and M . In individuals with elevated LDH,CRP , Ferritin or D-dimer levels the necessity for anticoagulant therapy may be considered to prevent diseases secondary to thromboembolic phenomena.

Acknowledgements:-

My sincere thanks to Dr.T.Durga ,Prof & HOD of Biochemistry, GMC, Ananthapur for her support.

Conflict of interest :
None.

Financial support:
None.

References:-

1.Mannan, A. et al. (2021). 'A multi-centre,cross-sectional study on coronavirus disease2019 in Bangladesh: clinical epidemiologyand short-term outcomes in recoveredindividuals', New Microbes and NewInfections, 40, p. 100838.doi:10.1016/j.nmni.2021.100838

2. Docherty, A.B. et al. (2020). 'Features of 20133 UK patients in hospital with covid-19using the ISARIC WHO ClinicalCharacterisation Protocol: prospectiveobservational cohort study, BMJ, p. m1985.doi:10.1136/BMJ.m1985.

3.De Prost, N. et al. (2021). 'Plasma Exchangeto Rescue Patients with Autoantibodiesagainst Type I Interferons and Life-Threatening COVID-19 Pneumonia',Journal of Clinical Immunology, 41(3), pp.536–544. DOI: 10.1007/s10875-021-00994-9

3A.Ministry of Health and Family Welfare. Government of India.Available from: https://www.mohfw.gov.in. [Last accessed Sep 30]

4. Suvvari TK, Kutikuppala LV, Babu GK, Jadhav MJ. Understanding the unusual viral outbreak: Coronavirus disease 2019 Curr Res Sci Med. 2020;6:3–10Cited Here

5. He X, Yao F, Chen J, Wang Y, Fang X, Lin X, et al The poor prognosis and influencing factors of high D-dimer levels for COVID-19 patients Sci Rep. 2021;11:1830.Cited Here

6.Liu F, Li L, Xu M, et al. Prognostic value of interleukin-6, C-reactive protein, and procalcitonin in patients with COVID-19. J Clin Virol Off Publ Pan Am Soc Clin Virol. 2020;127:104370. [PMC free article] [PubMed] [Google Scholar]

7. Canovi S, Besutti G, Bonelli E, et al. The association between clinical laboratory data and chest CT findings explains disease severity in a large Italian cohort of COVID-19 patients. BMC Infect Dis. 2021;21:157. [PMC free article] [PubMed] [Google Scholar]

8. Kazemi E, Soldoozi Nejat R, et al. The laboratory findings and different COVID-19 severities: a systematic review and meta-analysis. Ann Clin MicrobiolAntimicrob. 2021;20:17. [PMC free article] [PubMed] [Google Scholar]

9. Nadeem R, Elhoufi AM, Iqbal NE, et al. Prediction of Cytokine Storm and Mortality in Patients with COVID-19 Admitted to ICU: Do Markers Tell the Story? Dubai Med J. 2021;4:142–150. [Google Scholar]

10. Gandini O, Criniti A, Ballesio L, Giglio S, Galardo G, Gianni W, et al Serum ferritin is an independent risk factor for acute respiratory distress syndrome in COVID-19 J Infect. 2020;81:979–97Cited Here

11. Bozkurt FT, Tercan M, Patmano G, BingolTanrıverdi T, Demir HA, Yurekli UF. Can ferritin levels predict the severity of illness in patients with COVID-19? Cureus. 2021;13:e12832.Cited Here

12.Ministry of Health and Family Welfare. Government of India.Available from: https://www.mohfw.gov.in. [Last accessed2023Nov 03]

13. Zhou B, She J, Wang Y, Ma X. Utility of ferritin, procalcitonin, and C-reactive protein in severe patients with 2019 novel coronavirus disease Res Sq. 2020Cited Here

14. Zhang L, Yan X, Fan Q, Liu H, Liu X, Liu Z, et al D-dimer levels on admission to predict in-hospital mortality in patients with COVID-19 J ThrombHaemost. 2020;18:1324–9Cited Here

15. Ghosh A, Anjana RM, Shanthi Rani CS, Jeba Rani S, Gupta R, Jha A, Gupta V, Kuchay MS, Luthra A, Durrani S, Dutta K,Tyagi K, Unnikrishnan R, Srivastava BK, Ramu M, Sastry NG, Gupta PK, Umasankari G, Jayashri R, Mohan V, Misra A.Glycemic parameters in patients with new-onset diabetes during COVID-19 pandemic are more severe than in patients withnew-onset diabetes before the pandemic: NOD COVID India Study. Diabetes Metab Syndr 2021; 15: 215-220 DOI: 10.1016/j.dsx.2020.12.033].