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EDITORIAL

The Multisystem Inflammatory Syndrome in Adults with SARS-CoV-2 Infection (MIS-A)- Another Great Puzzle in Evolution

Though Multisystem Inflammatory Syndrome in Children (MIS-C) has been well characterised, recently evidence is emerging of a parallel syndrome in adults. A case series of 27 patients was published in Morbidity and Mortality Weekly Report (MMWR) on Oct 9, 2020[1]. These patients had cardiovascular, gastrointestinal, dermatologic and neurologic symptoms without severe respiratory illness and had positive test results for SARS-CoV-2 by RT-PCR or antibody assays indicative of recent infection. Although hyperinflammation and extrapulmonary organ dysfunction have been described in hospitalised adults with severe Covid-19, there conditions are generally accompanied by respiratory failure but patients in this series had minimal respiratory symptoms, hypoxaemia or radiographic abnormalities. Majority of patients with MIS-A survived – similar to those with MIS-C.

A systematic review of 221 patients was published in *JAMA Network Open* in September 2021[2] which found that 70% of the patients were men, 58% had no underlying comorbidity, and 68% had a previous symptomatic Covid-19 like illness. Most patients presented with fever (96%), hypotension (60%), cardiac dysfunction (54%), shortness of breath (52%) and/or diarrhoea (52%) Median number of organ system involved was 5, 57% were admitted to ICU and 47% needed respiratory support; 7% died. Most patients had elevated markers of coagulopathy and/or inflammation and positive SARS-CoV-2 serologic findings. The syndrome presented approximately 4 weeks after acute covid-19 with hyperinflammation and extrapulmonary multi-organ involvement.

CDC proposed a case definition of MIS-A[3] as a patient more than 21 years of age hospitalised for more than 24 hours or with an illness resulting in death who meets the proposed clinical and laboratory criteria in absence of more likely alternative diagnosis. In this issue, AK Agarwal and others report a 28-year-old case with a history of Covid around an year back who had received a dose of Covid vaccine 2 weeks back and who fulfilled all the criteria for diagnosis as MIS-A as per CDC definition. He had very high levels of anti spike protein neutralizing IgG antibodies which probably led to hyperinflammation and MIS-A. Hence further studies on immunopathogenesis of this syndrome are needed. If MIS is post-infectious or antibody mediated then there could be important implications for vaccination policy such as delaying vaccination in patients who have recently recovered from Covid-19.

Although the clinical presentation in some patients with severe Covid-19 could overlap with MIS-A, the pathophysiology may be different and hence distinguishing between the two syndromes has implications for treatment and long-term follow-up. Interim recommendations for MIS-A include corticosteroids, IVIG or possibly other immunomodulators.

References

1. Morris SB, Schwartz NG, Patel P *et al*. Case Series of Multisystem Inflammatory Syndrome in Adults Associated with SARS-CoV-2 infection-UK and US, March-August 2020, MMWR, October 2, 2020.
2. Patel P, DeCuir J, Abrams J *et al*. *JAMA Network Open* 2021; 4 (9): e2126456.
3. Centers for Disease Control and Prevention. Multisystem Inflammatory Syndrome in Adults (MIS-A) Case Definition Information for Healthcare Providers. Available from: https://www.cdc.gov/mis/mis-a/hcp.html.

– MPS Chawla
Editor

ORIGINAL ARTICLE

JIACM 2021; 22 (3-4): 87-92

Six Minute Walk Test (6MWT) in The Assessment of Severity of Interstitial Lung Disease Secondary to Systemic Sclerosis

BM Indu, Ulka Kamble**, Desh Deepak***, Brijesh Sharma*****

Abstract

***Background:** Interstitial lung disease (ILD) and pulmonary artery hypertension account for 60% of systemic sclerosis (SSc) related deaths. Impaired gas exchange that worsens with exercise is central to the pathophysiology of SSc-related ILD (SSc-ILD). Six minute walk test (6MWT) is a simple and cost-effective tool to assess lung function and has been proven to be reproducible. It has the potential to be employed as a tool to assess and monitor severity of pulmonary involvement in SSc.*

***Aims and objectives:** This study evaluates the correlation between 6MWT results with the other parameters of disease severity like forced vital capacity (FVC), diffusion capacity of lung for carbon monoxide (DLCO) from pulmonary function tests, right ventricular systolic pressure (RVSP) by echocardiography, HRCT findings like pulmonary artery dilatation and honeycombing and clinical findings like modified Rodnan skin score (mRSS) in patients of SSc-ILD.*

***Materials and methods:** 30 patients with SSc-ILD were subjected to two 6MWTs. Six minute walk distance (6MWD) < 400 m and fall in saturation during 6MWT (△Sat) ≥ 4% were considered abnormal. 6MWD of the two tests were compared. If the variability was more than 15%, a third test was planned. The two tests with 6MWD within 15% variability were considered for our studies. Then, 6MWD and △Sat were compared with FVC, DLCO, RVSP, HRCT and clinical findings like mRSS.*

***Results:** There was no statistically significant correlation between 6MWD and △Sat (p = 0.51). On univariate analysis, there was no statistically significant correlation of 6MWD < 400 m with mRSS(p = 0.07), %FVC (p = 0.59), %DLCO (p = 0.68), RVSP (p = 0.35) and pulmonary artery dilatation on HRCT (p = 0.713). There was statistically significant positive correlation between 6MWD < 400 m and pre-test Borg index (p = 0.04) and post-test Borg score (p = 0.02). On multi-variate logistic analysis, no parameters had statistically significant correlation with 6MWD < 400 m. However, on univariate analysis, there was statistically significant negative correlation of △Sat ≥ 4% on 6MWT with % FVC (p = 0.04) and % FEV1 (p = 0.027) and statistically significant positive correlation pre-test Borg score (p = 0.02), post-test Borg dyspnoea score (p < 0.0001), honey combing on HRCT (p = 0.044) and pulmonary artery dilatation on HRCT (p = .01). There was no statistically significant correlation between △Sat ≥ 4% and % DLCO (p = 0.24), RVSP (p = 0.74) or mRSS (p = 0.79). On multivariate logistic analysis, only pulmonary artery dilatation on HRCT had statistically significant positive correlation with △Sat ≥ 4% (p = 0.04).*

***Conclusions:** 6MWT is a highly reproducible test. Desaturation during 6MWT is more reflective of pulmonary involvement and is an adjunct to pulmonary function tests in evaluation of patients with SSc-ILD. 6MWD is subjective and depends on patient motivation.*

Introduction

Systemic sclerosis is a multisystem disorder with varying manifestations. There is a strong female preponderance in SSc with female: male ratio of 3:1[1]. Pulmonary involvement remains the most common cause of morbidity and mortality in cases of SSc. SSc-ILD and pulmonary artery hypertension (PAH) account for 60% of deaths related to SSc[2].

Early diagnosis and regular follow-up is the keystone to prevent the morbidity and mortality associated with these diseases. Central to the pathophysiology of SSc-ILD is the impaired gas exchange that worsens with exercise[3]. Interstitial lung disease can be assessed with high resolution computed tomography (HRCT) and pulmonary function test including diffusion study. HRCT chest is the reference tool to detect pulmonary abnormalities in SSc[4]. Additionally, it provides an accurate assessment of the extent of lung involvement. Evaluation of PAH, which is also a marker of disease severity in SSc-ILD, requires right heart catheterisation or an echocardiography. However, these modalities require expertise and specialised equipment. This demands for a simple, cost-effective and reproducible test that may help to study the disease severity.

Six minute walk test (6MWT) is a simple tool that does not need sophisticated equipment or training and has been validated in various respiratory diseases. It has been suggested that 6MWT can be used to assess the central

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pathophysiological mechanism in SSc which is the impairment of gas exchange that worsens with exercise[3]. Desaturation at the end of 6MWT is as informative as the decrease in walk distance for pulmonary involvement in SSc[5]. However, 6MWT has not been validated in SSc in Indian patients. In this study, we evaluated the utility of 6MWT as a measure of disease status in SSc-ILD.

Material and Methods

A cross-sectional observational study was done in a teaching hospital in New Delhi. The study included patients above 18 years of age with SSc-ILD. All patients were first subjected to detailed history, examination and laboratory investigations. SSc was diagnosed based on ACR/EULAR criteria and ILD on HRCT. HRCT chest is the reference tool to detect the pulmonary abnormalities in SSc[4]. Additionally, it provides an accurate assessment of the extent of the lung involvement. Pulmonary artery hypertension (PAH) was diagnosed on echocardiography. Right ventricular systolic pressure (RVSP) of 40 mm Hg or more was used as the diagnostic criterion for PAH. Pulmonary artery dilatation on HRCT was defined by the ratio of diameter of main pulmonary artery to ascending aorta greater than one. This radiological sign of PAH was also compared with 6MWT results.

Then, they were subjected to pulmonary function test (PFT) with diffusion capacity of lungs for carbon monoxide (DLCO) and echocardiography. Those patients with forced vital capacity (FVC) < 40%, DLCO < 40% and RVSP > 50 mm Hg, SpO2 < 84% were excluded from our study as they had severely compromised cardiopulmonary functions. These subjects may develop life-threatening complications during 6MWT. Patients with history of pulmonary tuberculosis (fibrotic changes in lungs secondary to tubercular sequelae may affect the 6MWT results), severe musculoskeletal problems (unable to perform 6MWT), non-steady or inadequate SpO_2 (due to Raynaud's phenomenon in SSc) were excluded from our study as they may give spurious 6MWT results despite a normal pulmonary function. Patients with contraindications to 6MWT were also excluded. Thus, a total of 30 SSc-ILD patients were included in our study.

The patients were then classified into diffuse and limited SSc variants. Clinical features, antibody profile, HRCT, echocardiographic, and PFT findings were studied.

Six minute walk test (6MWT)

A seldom travelled 100 feet long straight hallway with hard surface was chosen. Patients were asked to wear comfortable clothes and shoes. They were allowed to use their walking aids and usual medications. Repeat test was done about the same time of the day and in the same location. The patients were asked to sit at rest in a chair at the starting point for at least 10 minutes before the test starts. Baseline heart rate, blood pressure were measured. Pulse oximetry was done. Baseline dyspnoea index and fatigue were rated using the Borg scale.

Patients were asked to walk as far as possible for 6 minutes. They could walk back and forth the hallway and that they are permitted to slow down, stop and to rest as necessary and can resume walking as soon as they are able to do so. Distance covered in 6 minutes, post-walk heart rate, oxygen saturation and blood pressure, post-test Borg dyspnoea index and fatigue were noted. 6MWT was terminated before 6 minutes if the patient got exhausted, develop chest pain or intractable leg cramps.

Two 6MWTs were performed between a minimum of 2 hours and a maximum of 4 weeks interval. To ensure the consistency of the tests, 6MWD of the two 6MWTs was required to be within 15% of each other. In the event of >15% variability, a third test was done, to be within 15% variability of the first or the second 6MWT or the patient was excluded. The results of the 6MWT with the best 6MWD were considered for all analyses.

Statistical analysis

For the purpose of analysis, desaturation was defined as $\triangle$Sat $\geq$ 4% from baseline and 6MWD was considered abnormal when it was < 400 m. These cut-off values for 6MWT were considered as predictors of severe pulmonary involvement. These were set based on prior studies as the one by Villaba *et al*[5]. We then divided patients into two categories: a) 6MWD $\geq$ 400 m and 6MWD < 400 m and b) $\triangle$Sat $\geq$ 4% and $\triangle$Sat < 4%.

Bland Altman plot was used to find the difference in measurement of 6MWD and $\triangle$Sat at two different time intervals. Multivariate logistic regression was used to find the independent risk factors predicting the change in 6MWD and $\triangle$Sat.

The relationship between various 6MWT parameters like 6MWD, $\triangle$Sat, pre-test and post-test Borg dyspnoea index were analysed. The relation of 6MWT parameters with PFT parameters like DLCO, FVC, the echocardiographic parameter RVSP, HRCT findings and clinical findings like mRSS were analysed.

Results

Majority of our patients, 29 out of 30 (96.67%) were females (This may be attributed to the strong female preponderance of SSc[1]). The mean age of our study population was 38.03 ± 11.28 years. Majority of them

(56.67%) were below 40 years of age. The median disease duration was 5.5 years (6 months to 30 years). The majority of our patients (76.67%) had localised systemic sclerosis (LSSc). Out of these LSSc, two had LSSc-Rheumatoid arthritis overlap. Three of the patients had combined SSc-ILD-PAH (10%). mRSS was higher in the 6MWD < 400 m and △Sat ≥ 4% group.

Anti-nuclear antibody (ANA) was positive in 20 out of 30 patients (66.67%). Anti Scl-70 antibodies were positive in 13 patients (43.33%). Anti-centromere antibodies (ACA) were positive in 5 patients (16.67%). Antibodies like PM-Scl (10%), anti SSA/Ro (30%), anti SSB/La (30%), anti U1RNP (13.33%), anti Ku (3.33%) were also seen in our study population. Out of the 13 patients positive for anti-Scl-70 antibody, six had PSS and the rest had LSSc.

All our patients had ILD diagnosed on HRCT. The common findings in HRCT were ground glass opacities (86.67%), interlobar septal thickening (76.67%) and honeycombing (13.33%). ILD pattern on HRCT was non- specific interstitial pneumonia (NSIP) in 86.67% (26) and usual interstitial pneumonia (UIP) in the rest. Of the patients with UIP, two had progressive systemic sclerosis (PSS) and the other two had RA-LSSc overlap. Pulmonary artery dilatation, another useful sign of PAH was present in 13 (43%) of our study population.

3 out of 30 (10%) had echocardiographic evidence of pulmonary artery hypertension. Of the 13 who had CT evidence of PAH, only two had PAH by echocardiography.

DLCO < 50% was present in 33.33% (10) of our population; all of them except three had pulmonary artery dilatation on HRCT. This however did not have a statistical significance. PAH by echocardiography did not correlate with DLCO. Reduced DLCO without any other abnormalities was the only abnormality detected in 3 (10%) of our patients; the spirometry, RVSP and 6MWT were normal in them.

6MWT is a highly reproducible test. Bland Altman plot was used to find out the difference in measurement of 6MWD and △Sat of the two 6MWTs. We had done a third 6MWT in three of our patients. This was due to lack of steady SpO_2 reading secondary to Raynaud's phenomenon (during winter months) in all of them when the second 6MWT was done. However, the third 6MWT done later in the absence of Raynaud's phenomenon was within 15% variability of the first. The two 6MWTs within 15% variability were named 6MWT1 and 6MWT2 for study purpose. Only 6MWD1 and 6MWD2 (Fig. 1), also △Sat1 and △Sat2 (Fig. 2) did not exceed the maximum allowed difference between the two test results (i.e., within mean ± 1.96 SD) and hence the two 6MWT results were in agreement and could be used interchangeably.

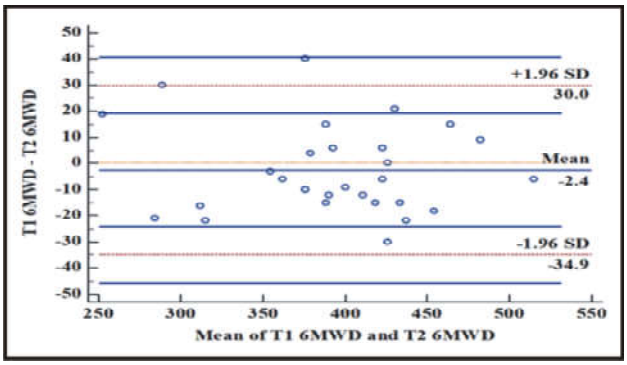

Fig. 1: *Bland Altman plot for 6MWD1 and 6MWD2.*

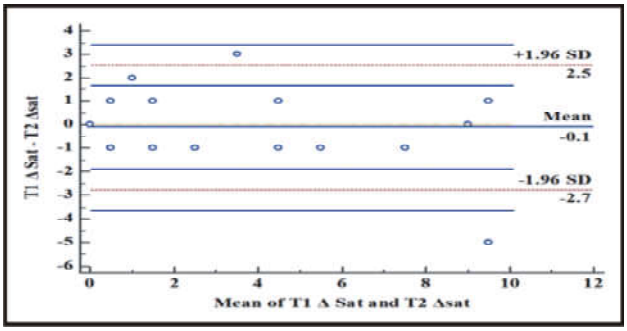

Fig. 2: *Bland Altman plot for △Sat 1 and △Sat 2.*

The mean 6MWD in our study was 400.97 ± 57.77 m. 50% had 6MWD < 400 m. The mean 6MWD in the group with △Sat ≥ 4% was 387.75 ± 45.99 m (304 to 441 m) and that in △Sat < 4% group was 407.59 ± 58.87 m (262 to 518 m).

8 out of 30 (26.67%) patients had desaturation during the 6MWT. The mean △Sat in our study population is 2.07 ± 3.03%. The mean △Sat in the group with 6MWD < 400 m was 2.4 ± 3.01% and that in 6MWD ≥ 400 m group was 1.73 ± 2.91%.

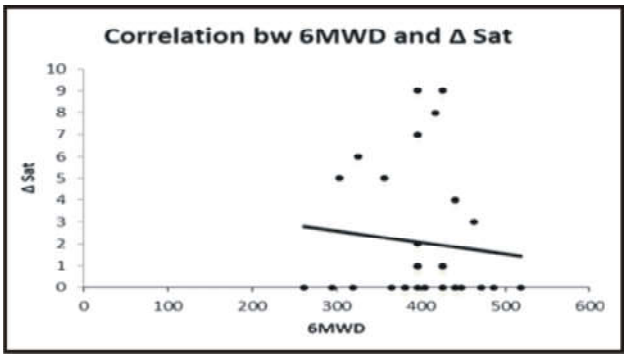

Fig. 3: *Correlation between 6MWD and △Sat.*

Fig. 3 shows the correlation between 6MWD and △Sat. There was no statistically significant correlation between the two variables (p = 0.5058).

Age and disease duration were lower, mRSS and weight were higher in the 6MWD < 400 m group. % FVC, % DLCO and % RVSP were lower in this group. On univariate analysis (Table I), there was no statistically significant correlation between 6MWD < 400 m and % FVC, % DLCO, RVSP or other parameters discussed above. There was statistically significant correlation between 6MWD < 400 m and pre-test Borg index and post-test Borg score. On multi-variate logistic analysis, none of the above parameters had a statistically significant correlation with 6MWD < 400 m.

Table I: Variables associated with 6MWD.

Variable	Mean value in 6MWD < 400 m	Mean value in 6MWD ≥ 400 m	P value in univariate analysis	Odds Ratio
Weight (Kg)	50.4 ± 7.47	49.4 ± 7.9	0.713	1.018
mRSS	23.27± 7.43	18.67 ± 5.27	0.077	1.129
FVC (%)	59.88 ± 11.61	62.99 ±20.15	0.595	0.988
FEV1 (%)	62.15 ± 12.77	65.12 ± 20.22	0.621	0.989
DLCO (%)	64.08 ± 19.32	67.49 ± 26.09	0.676	0.993
RVSP (mm Hg)	27.73 ± 8.46	30.47 ± 7.55	0.347	0.956
Pre-Borg dyspnoea index (n)	2.33±1.19	1.27±1.18	0.036	2.079
Post-Borg dyspnoea index (n)	5.47± 1.45	3.33±2.12	0.016	1.879
Honeycombing on HRCT (n)	2	2	1.00	1.00
Pulmonary artery. dilatation on HRCT (n)	6	7	0.713	0.762

Age, weight, disease duration, mRSS were higher in the group with △Sat ≥ 4%. %FVC, %DLCO and 6MWD were lower and RVSP was higher for the group with △Sat ≥ 4%. On univariate analysis (Table II), △Sat ≥ 4% had statistically significant correlation with %FVC, pre-test Borg score and post-test Borg dyspnoea score. Honey combing on HRCT and pulmonary artery dilatation on HRCT also had significant correlation with △Sat ≥ 4%. There was no statistically significant correlation between △Sat ≥ 4% and % DLCO or RVSP. On multivariate logistic analysis, only pulmonary artery dilatation on HRCT had statistically significant correlation with △Sat ≥ 4% (p = 0.04).

Of the three cases of PAH diagnosed on echocardiography, one had △Sat ≥ 4% with 6MWD ≥ 400 m. One had both △Sat and 6MWD normal as per our set criteria. Third patient had 6MWD < 400 m with △Sat < 4%. RVSP had no statistically significant correlation between either of the 6MWT parameters.

In our study, almost all cases with pulmonary artery dilatation in HRCT had reduced % DLCO. So, we analysed its correlation with RVSP and % DLCO. There was no statistically significant correlation between pulmonary artery dilatation and the above two parameters.

Table II: Variables and △Sat.

Variable	Mean value in △Sat ≥ 4%	Mean value in △Sat < 4%	P value in univariate analysis	Odds ratio
Weight	54.25 ± 6.98	48.32 ± 7.29	0.069	1.112
mRSS	21.5 ± 7.1	20.77 ± 6.77	0.79	1.016
%FVC	50.72 ± 10.69	65.33 ± 16.32	**0.044**	0.923
%FEV1	51.69 ± 12.53	67.98 ± 16.06	**0.027**	0.914
%DLCO	57.68 ± 19.02	68.73 ± 23.49	0.243	0.975
RVSP (mm Hg)	29.88 ± 8.31	28.82 ± 8.07	0.745	1.017
Pre-Borg dyspnoea scale(n)	3 ± 0.87	1.36 ± 1.15	**0.016**	4.00
Post-Borg dyspnoea scale(n)	6.75 ± 0.83	3.54 ± 1.75	**<0.0001**	51.32
Honeycombing on HRCT(n)	3	1	**0.044**	12.6
Pulmonary artery dilatation on HRCT(n)	7	1	**0.012**	18.67

Discussion

Lung involvement is a leading cause of morbidity and mortality in SSc patients. The prevalence of ILD in SSc is difficult to predict as patients are asymptomatic earlier in the disease. Early screening for pulmonary involvement and follow-up in initial years prevents morbidity. 6MWT is a submaximal exercise test that can be used to monitor disease progression in SSc-ILD. This cost-effective test can be handy especially in a low resource country.

The study characteristics of our study population and HRCT findings were similar to studies conducted earlier by Villaba *et al*[5]. Majority of our population were females (29 out of 30). This is attributed to the female preponderance of SSc (female: male ratio of 3:1)[1]. Most of them were below 40 years of age and had disease duration of 5.5 years. Organ involvement in SSc occurs within the first 4 years of the disease.

Extent of disease by HRCT is a marker of disease severity, so is honeycombing pattern. Honeycombing was present in 4 of our patients. UIP pattern in our PSS cases may be due to the more extensive disease that takes on a characteristic reticulo-nodular appearance and associated with fine honeycomb airspaces and ultimately large airspaces[6].

PAH as defined by echocardiography was present in three

of our patients but when defined by HRCT, it was present in thirteen. The dilatation of pulmonary artery trunk is one among the CT findings in PAH. This is also a marker of severity of lung disease. Distal main pulmonary artery dilatation exceeding that of aorta has a positive predictive value of › 95% and a specificity of more than 90% for the diagnosis of PAH[7,8].

DLCO was reduced in almost all patients with other PFT abnormalities in our study. This is in agreement with the study by Bourous *et al*[9]. Of the four patients with FVC > 80%, three had DLCO < 80%. Thus, reduced DLCO was the only abnormality in 10% of our study population. This is similar to the study by Steen *et al*[10] wherein isolated reduction in DLCO was present in 19% of their study population. Thus, low DLCO without reduced FVC is the earliest and most sensitive pulmonary functional abnormality in systemic sclerosis.

The two 6MWTs were highly reproducible both in terms of 6MWD and desaturation when compared using the Bland Altman plot. This is in agreement to studies by Gregory Pugnet *et al*[11] and Buch *et al*[12]. Thus, 6MWT is a highly reproducible test.

Clinical findings like extent of skin involvement as calculated by modified Rodnan Skin Score, HRCT findings like honeycombing and pulmonary artery dilatation, PFT parameters like % FVC, % FEV1 and % DLCO, echocardiographic parameter RVSP, etc., are also measures of disease severity in SSc-ILD. 6MWT being a simple easily reproducible tool to assess the cardiopulmonary function, we assessed the correlation between the two 6MWT results with the various other parameters of disease severity.

% FVC and % FEV1 had statistically significant correlation with △Sat. There were trends that % DLCO was lower and RVSP was higher in the △Sat ≥ 4% group. But, there was no statistical significance. This may be due to the small sample size. Thus, △Sat is not completely reflective of DLCO and hence 6MWT may not be able to identify SSc-ILD in the initial stages. More studies with large sample size may be needed to establish this relation.

Of the other parameters analysed, honeycombing and pulmonary artery dilatation on HRCT and Borg dyspnoea index (both pre- and post-test) too had statistically significant correlation with △Sat.

Reduced DLCO may be the initial manifestation of pulmonary involvement in SSc-ILD. But, none of the 6MWT parameters has a correlation with % DLCO. In our study, PAH by echocardiography neither correlated with % DLCO nor with △Sat. Almost all cases of pulmonary artery dilatation had reduced DLCO and vice-versa. However, there was no statistically significant correlation between %DLCO and pulmonary artery dilatation. This may be due to the small sample size and hence, more studies need to be conducted to fully validate this.

Pulmonary artery dilatation was the only parameter which had a statistically significant relation with △Sat on multivariate analysis. Thus, pulmonary artery dilatation on HRCT is more predictive of % DLCO and △Sat than echocardiographic evidence of PAH. None of the previous studies had looked into this finding. Since we did not perform right heart catheterisation (RHC) to diagnose PAH in these cases, more studies need to be done to prove the superiority of CT over echocardiography to diagnose PAH.

The mean desaturation was higher in the 6MWD < 400 m group and the mean 6MWD was lower for △Sat ≥ 4% group. But, there was no statistically significant correlation between 6MWD and △Sat, unlike the study by Villaba *et al*[5].

There were trends that higher disease activity as indicated by mRSS and lower PFT values were associated with 6MWD <400. But, none of the PFT parameters or RVSP on echocardiography had a statistical significance when compared with 6MWD <400 m.

It was also noted that the highest and the lowest 6MWD is recorded in the group with △Sat ≥ 4%. Systemic sclerosis being a multisystem disease, the various non-pulmonary aspects like musculoskeletal pain can affect 6MWD. Thus, 6MWD is more subjective than △Sat and depends on patient motivation.

Of the two 6MWT parameters, △Sat is more correlating to PFT parameters than 6MWD. Musculoskeletal involvement does not affect the △Sat. Thus, △Sat is more predictive of pulmonary involvement than 6MWD. The drawbacks of our study were the small sample size. We did not confirm PAH by RHC.

Conclusion

6MWT is a highly reproducible test and can be used in the follow-up of patients with SSc-ILD. Of the two 6MWT parameters, 6MWD is more subjective. △Sat is more predictive of lung involvement and can be used to assess the progression of disease and aids in further evaluation and treatment modification. Being simple and cost effective, 6MWT can be used to detect trends in disease progression. However, more studies have to be done to fully validate its usefulness in SSC-ILD.

References

1. Peoples C, Medsger Jr TA, Lucas M *et al*. Gender differences in Sysytemic sclerosis: relationship to clinical features, serologic status and outcomes. *J Scleroderma Relat Disord* 2016; 1 (2): 177-240.
2. Steen VD, Medsger TA. Changes in causes of death in systemic

sclerosis, 1972-2002. *Ann Rheum Dis* 2007; 66 (7): 940-4.

3. Morelli S, Ferrante L, Sgreccia A *et al*. Pulmonary hypertension is associated with impaired exercise performance in patients with Systemic sclerosis. *Scand J Rheumatol* 2000; 29: 236-42.
4. Pignone A, Matucci-Cerinic M, Lombardi *et a*l. High resolution tomography in systemic sclerosis. Real diagnostic utilities in the assessment of pulmonary involvement and comparison with other modalities of lung investigation. *Clin Rheumatology* 1992; 11: 465-72.
5. Villaba WO, Sampaio-Barros PD, Pereira MC *et al*. Six minute walk test for the evaluation of pulmonary disease severity in scleroderma patients. *Chest* 2007; 131 (1): 217-22.
6. Desai SR, Veeraraghavan S, Hansell DM *et al*. CT features of lung diseases in patients with systemic sclerosis: Comparison with idiopathic pulmonary fibrosis and non specific interstitial pneumonia. *Radiology* 2004; 232 (2): 560-7.
7. Tan RT, Kuzo R, Goodman LR *et al*. The utility of CT scan in the evaluation for predicting pulmonary artery hypertension in patients with parenchymal lung disease. Medical college of Wisconsin Lung Transplant Group. *Chest* 1998; 113 (5); 1250-6.
8. Pandey AK, Wilcox P, Mayo JR *et al*. Predictors of pulmonary hypertension on High Resolution Computed Tomography of the Chest in Systemic sclerosis. A retrospective analysis. *Can Assoc Radiol J* 2010; 61 (5): 291-600.
9. Bourous D, Wells AU, Nicholson AG *et al*. Histopathologic subsets of fibrosing alveolitis in patients with systemic sclerosis and their relationship to outcome. *Am J Respir Crit Care Med* 2002; 165 (12): 1581-6.
10. Steen V, Medsger TA. Predictors of isolated pulmonary artery hypertension in patients with systemic sclerosis and limited cutaneous involvement. *Arthritis Rheum* 2003; 48 (2): 516-22.
11. Pugnet G, Marjanovic Z, Delingny C *et al*. Reproducibility and utility of the six minute walk test in systemic sclerosis. *J Rheumatol* 2018: jrheum.170994; doi: https://doi.org/10.3899/jrheum.170994.
12. Buch MH, Denton CP, Furst DE *et al*. Submaximal exercise testing in the assessment of interstitial lung disease secondary to systemic sclerosis: Reproducibility and correlations of the 6 minute walk test. *Ann Rheum Dis* 2007; 66 (2): 169-73.
13. Someya F, Mugii N, Hasegawa M *et al*. Predictors of Exercise induced oxygen desaturation in systemic sclerosis patients with interstitial lung disease. *Respir care* 2014; 59 (1): 75-80.

ORIGINAL ARTICLE

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Clinical Profile, Including Complications, in Patients with *Vivax* Malaria Mono-Infection

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Abstract

***Background and objectives:** Vivax malaria was long considered to have a benign course. However, in recent times there has been a remarkable increase in case studies describing complicated disease with vivax malaria. We report findings from a study conducted at a tertiary care hospital aiming at analysing the clinical manifestations, complications, and outcome of patients infected with P. vivax malaria.*

***Methodology:** This hospital-based cross-sectional observational study was carried-out on 100 patients diagnosed with P. vivax infection from January 2019 to June 2020. Detailed history, clinical findings, and relevant investigations were recorded and analysed.*

***Results:** 100 patients with malarial parasite rapid diagnostic test and peripheral smear positive for P. vivax were selected. Mean age was 30.59 ± 13.35 years, 62% were in age group of 20 - 30 years, 55% were males. Clinical features seen were fever (100%), vomiting (53%), abdominal pain (44%), headache (44%), cough (18%), breathlessness (12%) and altered sensorium (1%).*

Pallor was seen in 53% of cases, icterus in 32%, pedal oedema in 7%, splenomegaly in 61%, hepatomegaly in 36% and chest crepitations in 12%. Severe anaemia was seen in 11% of cases, thrombocytopenia in 85%, azotaemia in 29%, hyperbilirubinaemia in 76%, altered liver transaminases in 50%, and ARDS in 2%. 5% patients required dialysis support. All patients recovered and were discharged without residual features.

***Conclusion:** P. vivax malaria may be a potentially life-threatening disease which has excellent prognosis if diagnosed early and treated appropriately. Parameters like severe anaemia, thrombocytopenia and hepato-renal dysfunction serve as early indicators for progression into rather severe disease.*

Introduction

Malaria is a major public health problem, endemic in over a hundred countries across the world[1]. *P. vivax* malaria currently has the widest geographical distribution among all malaria parasites with about 35% of the world population living at risk of this physically debilitating infection[2-4]. A total of 130 - 435 million people are estimated to get *P. vivax* infection annually[5].

During recent years, many surveillance studies[6-9], case series[10-13] and reviews[14-16] have linked *vivax* malaria with severe manifestations similar to those seen in *P. falciparum* infection; observations that conflict the notion that *vivax* malaria is a benign disease.

This observational study was conducted with the aim of exploring varied clinical features of *vivax* malaria. Comparison of clinical profile of patients with *vivax* malaria with regards to demographic, clinical, haematological, and biochemical features was done in this study.

Material and methods

This was a cross-sectional, observational study conducted at the Department of Medicine in a tertiary care teaching institute in Greater Noida, Uttar Pradesh, over a period of one year and six months from January 2019 to June 2020. The study was approved by the institutional ethics committee.

Inclusion criteria

1. Patients admitted in hospital having fever (≥ 38.5° C) with peripheral smear and rapid malaria diagnostic test positive for *P. vivax* malaria.
2. Patients willing to give written consent.
3. All patients >20 years of age.

Exclusion criteria

1. Patients with co-existent *falciparum* infection or dengue virus infection.

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2. History of chronic HBV and/or HCV infection.
3. Patients with advanced stage of HIV or AIDS.
4. Patient with history of hepatotoxic drug, toxic herbal medicine, alcohol consumption.
5. Chronic liver disease/renal failure patients.

Detailed medical history was taken and a thorough clinical examination was performed on all patients. Light microscopy Giemsa-stained peripheral blood smear examination and malarial antigen based rapid diagnostic testing (RDT) were used to diagnose *P. vivax* malaria. The malaria card lactate dehydrogenase/histidine-rich protein 2 (pLDH/HRP2) combo (Pf/Pv) test was used. A complete blood count, liver function test, kidney function test, random plasma glucose estimation, ABG, CXR PA view, HEPACARD for HBsAg, HCV TRI-DOT, HIV TRI-DOT, Dengue serology (IgG, IgM) were done for all the patients.

Patients were also evaluated for severe malaria as per WHO criteria[17]. The parameters taken into consideration for identifying severe malaria were: Impaired consciousness, severe anaemia (Hb <7 gm/dl), pulmonary oedema, jaundice (serum bilirubin >3 mg/dl), acute renal failure (serum creatinine >3 mg/dl), convulsions, hypoglycaemia (plasma glucose <40 mg/dl), acute respiratory distress syndrome, bleeding manifestations, hypotension (systolic blood pressure <80 mm of Hg), and metabolic acidosis.

Patients were treated as per standard practice, in accordance with the national malarial management protocol. Patients were observed from the time of admission up to discharge from the hospital.

Statistical analysis

Data was collected in a predetermined proforma and entered in Microsoft excel sheet. The data was analysed using the SPSS version 21 operating on windows 10. All the data represented in tables as frequency, percentage, mean, standard deviation and diagrammatic representation using the pie-chart and bar charts as applicable. P value less than 0.05 was considered significant.

Results

Total of 100 consecutive patients attending the medicine department and diagnosed with *P. vivax* malaria were included in the present study after obtaining informed consent from all patients.

Maximum patients belonged to the age group of 20 - 30 years (62%) (Fig. 1). 55% were males, whereas 45% were females (Fig. 2). The most common symptom was fever, found in all patients. Vomiting, abdominal pain and

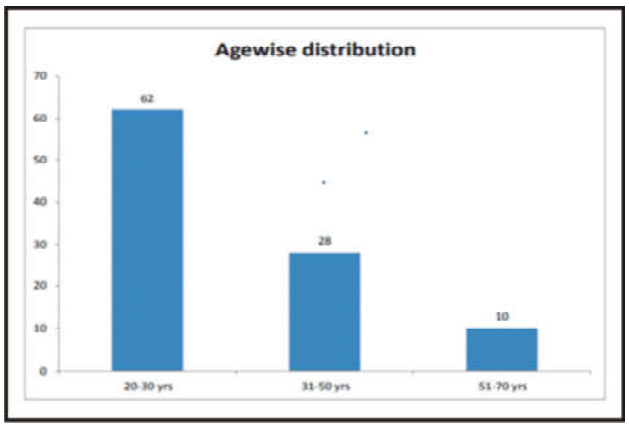

Fig. 1: *Agewise distribution of patients.*

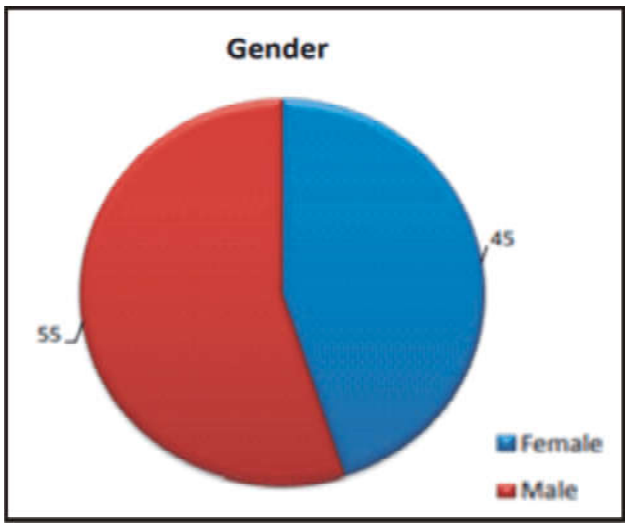

Fig. 2: *Gender distribution of patients.*

headache were next in frequency, being found in 53%, 44% and 44%, respectively. 18% patients had cough, 12% patients had breathlessness, and 1 patient presented with altered sensorium (Fig. 3).

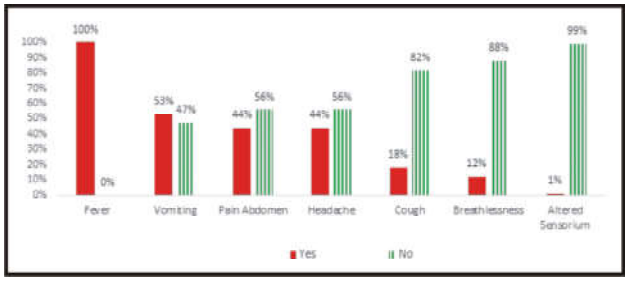

Fig. 3: *Symptoms among patients of vivax malaria.*

On examination, 53% patients had pallor, 32% patients had icterus, 61% patients had splenomegaly, and 36% patients

had hepatomegaly. On respiratory examination, 12% patients had bilateral crepitations and 3% had wheezing. One patient with severe anaemia was found to have a murmur (Fig. 4).

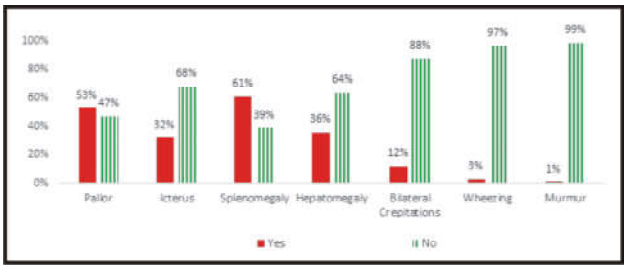

Fig. 4: *Signs among patients of vivax malaria.*

On laboratory investigations, 11 patients had severe anaemia with Hb lower than 7 gm%, 35% were in range of 7.1 - 10 gm%, 34% patients in range of 10.1 - 13 gm% and 20% were in range of 13.1 - 18 gm% (Fig. 5).

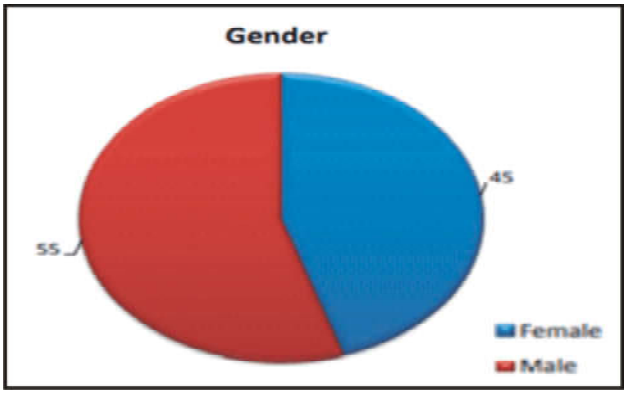

Fig. 5: *Haemoglobin distribution among the patients with malaria.*

38% of patients were found to have thrombocytopenia with platelet counts lower than 50,000/cmm followed by 33% patients in range of 50,000 - 1,00,000/cmm. Only 12% of patients with *P. vivax* were with platelet count more than 1.5 lakh (Fig. 6).

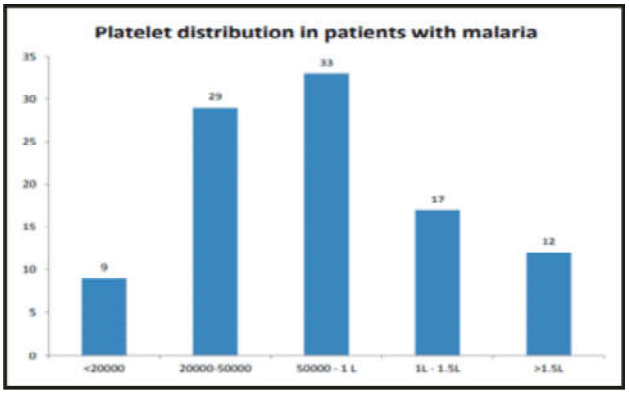

Fig. 6: *Platelet distribution in patients with malaria.*

76% of patients had elevated bilirubin more than 1 mg/dl. Only 24% were with bilirubin lower than 1 mg/dl. Majority of the patients who had elevated bilirubin were in the range group of 1.01 - 3 mg/dl of serum bilirubin (48%), and the remaining 28% patients had bilirubin more than 3 mg/dl. Serum AST level was more than 41 IU/L in 70% of the patients and serum ALT was more than 40 IU/L in 64% of the patients. The mean level of AST and ALT was 62.2 IU/L and 62.5 IU/L in our patients (Fig. 7).

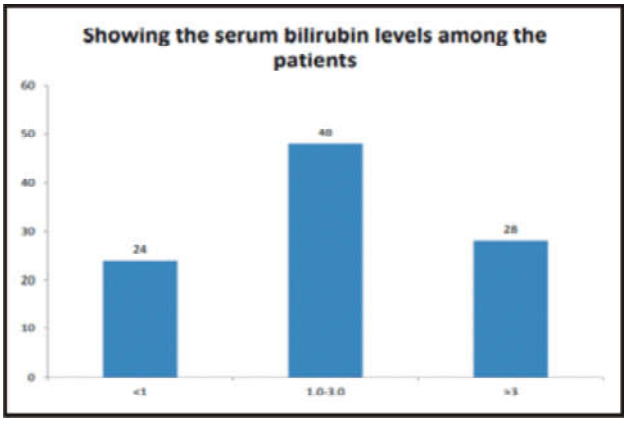

Fig. 7: *Serum billirubin level among patients of malaria.*

The serum creatinine was elevated among the 33% of patients and blood urea was elevated in 29% of patients. 5% of the patients required dialysis support (Fig. 8).

CXR suggestive of ARDS was seen in 2% of cases. Cerebral malaria was seen in 1% of cases. All 100 patients improved and were discharged.

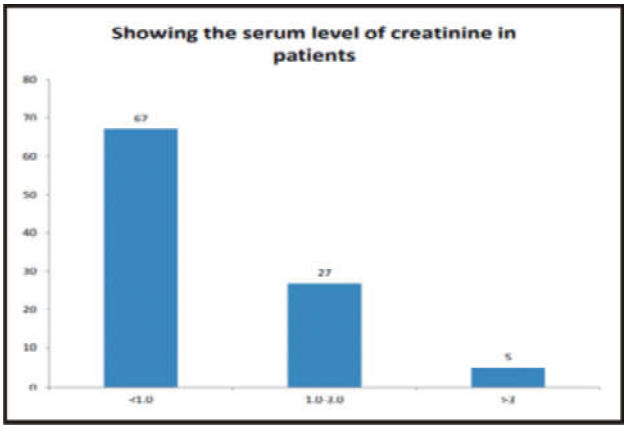

Fig. 8: *Serum creatinine level among patients of malaria.*

Discussion

Total of 100 patients diagnosed with *P. vivax* fulfilling the inclusion and exclusion criteria were included in this study.

Among 100 patients, 55 were males and 45 were female

patients (with male preponderance). Similar results have been reported by various studies conducted across India[18-20]. The mean age of all the participants was 30.59 ± 13.35 years. The majority of the patients in the present study were in the age group of 20 - 30 years (62%) followed with patients in age group of 31 - 50 years (28%) and 51 - 70 years with 10%.

In the present study, all patients presented with fever at presentation. 53% patients also had vomiting, 44% patients presented with pain abdomen, 44% patients with headache, 18% patients with cough, 12% patients with breathlessness, and 1 patient with altered sensorium. No patient in the present study had history of convulsion at presentation. Similar findings have been reported by many others authors such as Mathews *et al* (fever 100%, vomiting 36%)[20], Manikyamba *et al* (fever 80%, headache 40%, vomiting 30%)[21], and Yadav *et al* (fever 98.2%, vomiting 42.8%)[22] (Table I).

Table I:

Clinical features	Present study	Manikyamba *et al*	Mathews *et al*	Yadav *et al*
Fever	100.0%	80.0%	100.0%	98.2%
Vomiting	53.0%	30.0%	36.0%	42.8%
Pain abdomen	44.0%	–	24.7%	28.5%
Cough	18.0%	–	13.3%	12.3%
Headache	44.0%	40.0%	20.0%	24.2%
Breathlessness	12.0%	14.0%	10.0%	–
Altered sensorium	1.0%	–	3.3%	20.2%

Severe anaemia (Hb < 7 gm%) was seen in 11% of patients, moderate anaemia (Hb 7.1 - 10 gm%) was seen in 35% of patients. Many previous studies have also documented severe anaemia in *vivax* malaria[23-25]. The pathophysiology of malarial anaemia is multifactorial. In developing tropical countries, pre-existing anaemia – most commonly due to malnutrition and helminthiasis – compounds the problem. *Vivax*-associated anaemia is an important public health concern that underscores the importance of reducing global transmission of *P. vivax*.

Thrombocytopenia was seen in 88% of our patients (platelet count <1,50,000/mm^3) (Table II). Thrombocytopenia was the most common complication seen in the present study. Naha *et al*[26] reported thrombocytopenia in 86.4%, George *et al*[27] and Singh *et al*[28] reported thrombocytopenia in 93.3% and 96% of patients respectively. Increased splenic sequestration, immune-mediated degradation, and shortened platelet survival are known to cause thrombocytopenia.

Table II:

Lab feature/ Result	Present study	Manikyamba *et al*	Mathews *et al*	Yadav *et al*
Anaemia	11.0%	37.5%	4.0%	17.4%
Thrombocytopenia	85.0%	12.5%	86.7%	81.2%
Raised ESR	47.0%	–	36.0%	–
Deranged KFT	29.0%	6.3%	–	–
Hyperbilirubinaemia	76.0%	10.0%	36.0%	13.5%
Altered liver transaminases	50.0%	4.9%	–	–

Hepatic dysfunction was the second most common complication noted in the present study. Majority of patients had elevated bilirubin ranging from 1.01 - 3 mg/dl (48%) and the remaining 28% patients had bilirubin more than 3 mg/dl. Serum AST and ALT levels were more than 40 IU/L in 70% and 64% of the patients respectively. The mean level of AST and ALT was 62.2 and 62.5 IU/L in our patients respectively. Serum ALP was also elevated in 28% of patients. Kochar *et al*[29] and George *et al*[27] reported hepatic dysfunction in 57.5% and 43.3% of patients respectively. Jaundice in malaria results from haemolysis of both parasitised and non-parasitised red cells as well as malarial hepatitis. Raised liver enzymes occur because of injury to hepatocyte and cholestasis. Hepatic dysfunction is reversible in acute malaria and patients respond favourably to antimalarial therapy without any residual effects.

Renal dysfunction was seen in 33% of patients in the present study. 5% of patients required dialysis support. Kochar *et al*[29], Naha *et al*[26], George *et al*[27], and Singh *et al*[28] reported renal dysfunction in 57.5%, 27.5%, 26.7% and 26% patients of *vivax* malaria. In a retrospective study on 93 patients of malarial ARF, 19 (20.4%) were found due to *P. vivax* infection[30]. Aetiology of renal failure in malaria is multifactorial – volume depletion, intravascular haemolysis, and hyperbilirubinaemia are factors considered responsible for renal injury[31].

Two patients in the present study developed ARDS. Indian studies described above have also observed occurrence of ARDS in their study groups[27]. Mathews *et al*[20], and Yadav *et al*[22], found ARDS IN 12.7% and 2.2% of patients, respectively.

In the present study, cerebral malaria was seen in 1 patient. Cerebral malaria due to *P. vivax* infection has been documented in various case reports[32,33], Hazra *et al*[34], and Singh *et al*[28], found cerebral malaria in 1.3%, and 13%, respectively. Cerebral dysfunction in vivax malaria may occur through the generation of nitric oxide[32].

Atypical presentation encountered in the present study was hyperglycaemia which was found in 3% of our patients, which could be explained as stress-induced. Stress-induced

hyperglycaemia usually occurs in children during serious illness, including in those with previously normal homoeostasis of glucose[35].

In the present study, 5% patients required dialysis support and no patients were put on the ventilator. All the 100 patients recovered and were discharged with no significant adverse events.

Limitations

1. Molecular diagnosis (by PCR assay) which has emerged as the most sensitive method for malaria diagnosis was not used in our study. However, despite the high negative predictive value of molecular testing, the majority of laboratories in non-endemic settings do not use PCR as a first-line diagnosis for all their malaria suspected cases. The major reason for not fully replacing existing conventional methods with this real-time PCR is the longer "time to results" period of the PCR.
2. The number of patients included in our study was not very large.

Conclusion

P. vivax infection has a varying clinical profile. Though previously called as benign tertian malaria, it is actually not a benign disease. It has immense potential to cause life-threatening complications – and even death – if not detected early and treated appropriately. This study highlights that certain clinical characteristics such as vomiting, abdominal pain, headache, altered consciousness, breathlessness, cough, hepatosplenomegaly, and laboratory parameters such as extreme thrombocytopenia, leucopenia, increased total bilirubin, elevated serum creatinine and blood urea may serve as indicators of the onset of severe malaria.

References

1. Remme JH, Binka E, Nabbaro D. Towards a framework and indicators for monitoring roll back malaria. *Am J Trop Med Hyg* 2001; 64: 76-84.
2. Gething PW, Elyazar IR, Moyes CL *et al*. A long – neglected world malaria map: Plasmodium vivax endemicity in 2010. *PLoS Neglected Tropical Dis* 2012; 6: 1814.
3. Battle KE, Gething PW, Elyazar IR *et al*. The global public health significance of Plasmodium vivax. *Advances in Parasitolo* 2012; 80: 1-111.
4. Rosalind EH, Katherine EB, Kamini NM *et al*. Global epidemiology of Plasmodium vivax. Am J Tropic Med Hygiene 2016; 95: 15-34.
5. Hay SI, Guerra CA, Tatem AJ *et al*. The global distribution and population at risk of malaria: Past, present, and future. *Lancet Infect Dis* 2004; 4: 327-36.
6. Genton B, D'Acremont V, Rare L *et al*. Plasmodium vivax and mixed infections are associated with severe malaria in children: a prospective cohort study from Papua New Guinea. *PLoS Med* 2008; 5: e127.
7. Barcus MJ, Basri H, Picarima H *et al*. Demographic risk factors for severe and fatal vivax and falciparum malaria among hospital admissions in Northeastern Indonesian Papua. *Am J Trop Med Hyg* 2007; 77: 984-91.
8. Tjitra E, Anstey NM, Sugiarto P *et al*. Multidrug-resistant Plasmodium vivax associated with severe and fatal malaria: a prospective study in Papua, Indonesia. *PLoS Med* 2008; 5: e128.
9. Rodriguez-Morales AJ, Benitez JA, Arria M. Malaria mortality in Venezuela: focus on deaths due to Plasmodium vivax in children. *J Trop Pediatr* 2008; 54: 94-101.
10. Kochar DK, Das A, Kochar SK *et al*. Severe Plasmodium vivax malaria: a report on serial cases from Bikaner in Northwestern India. *Am J Trop Med Hyg* 2009; 80: 194-8.
11. Beg MA, Sani N, Mehraj V *et al*. Comparative features and outcomes of malaria at a tertiary care hospital in Karachi, Pakistan. *Int J Infect Dis* 2008; 12: 37-42.
12. Kochar DK, Saxena V, Singh N *et al*. Plasmodium vivax malaria. *Emerg Infect Dis* 2005; 11: 132-4.
13. Lawn SD, Krishna S, Jarvis JN *et al*. Case reports: pernicious complications of benign tertian malaria. *Trans R Soc Trop Med Hyg* 2003; 97: 551-3.
14. Price RN, Tjitra E, Guerra CA *et al*. Vivax malaria: neglected and not benign. *Am J Trop Med Hyg* 2007; 77: 79-87.
15. Price RN, Douglas NM, Anstey NM. New developments in Plasmodium vivax malaria: severe disease and the rise of chloroquine resistance. *Curr Opin Infect Dis* 2009, 22: 430-35.
16. Baird JK. Neglect of Plasmodium vivax malaria. *Trends Parasitol* 2007; 23: 533-9.
17. Maguire JD, Baird JK. The 'non-falciparum' malarias: the roles of epidemiology, parasite biology, clinical syndromes, complications and diagnostic rigour in guiding therapeutic strategies. *Ann Trop Med Parasitol* 2010; 104: 283-301.
18. World Health Organization, Severe Malaria. *Trop Med Int Health* 2014; 19: 7-131.
19. Saravu K, Docherla M, Vasudev A *et al*. Thrombocytopenia in vivax and falciparum malaria: an observational study of 131 patients in Karnataka, India. *Ann Trop Med Parasitol* 2011; 105: 593-8.
20. Verma P, Shukla U, Kalraiya A. Retrospective Study on Clinical Profile of Severe Malaria in Children Admitted in a Tertiary Care Centre of Central India. *People's J Sci Res* 2014; 7: 22-7.
21. Mathews S, Bhagwati M, Agnihotri V. Clinical spectrum of Plasmodium vivax infection, from benign to severe malaria: A tertiary care prospective study in adults from Delhi, India. *Trop Parasitol* 2019; 9: 88-92.
22. Manikyamba D, Prasad AK, Satyavani A *et al*. Clinical profile and complications of acute malaria caused by different species of Plasmodium. *Pediatr Rev Int J Pediatr Res* 2016; 3: 13-8.
23. Yadav G, Pardeshi G, Roy N. Clinico-epidemiological profile of patients admitted with Plasmodium vivax malaria in a tertiary care hospital, Delhi. *Int J Community Med Public Heal* 2018; 5: 5420-8.
24. Kochar DK, Saxena V, Singh N *et al*. *Plasmodium vivax* malaria. *Emerg Infect Dis* 2005; 11: 132-4.
25. Naha K, Dasari S, Prabhu M. Spectrum of complications associated with *Plasmodium vivax* infection in a tertiary hospital in South-Western India. *Asian Pac J Trop Med* 2012; 5: 79-82.

26. George P, Alexander LM. A study on the clinical profile of complicated *plasmodium vivax* mono-infections. *Asian Pac J Trop Med* 2010; 3: 560-62.
27. Naha K, Dasari S, Prabhu M. Spectrum of complications associated with *Plasmodium vivax* infection in a tertiary hospital in South-Western India. *Asian Pac J Trop Med* 2012; 5: 79-82.
28. George P, Alexander LM. A study on the clinical profile of complicated *plasmodium vivax* mono-infections. *Asian Pac J Trop Med* 2010; 3: 560-62.
29. Singh H, Parakh A, Basu S *et al*. *Plasmodium vivax* malaria: Is it actually benign? *J Infect Public Health* 2011; 4: 91-5.
30. Kochar DK, Das A, Kochar SK *et al*. Severe *Plasmodium vivax* malaria: a report on serial cases from Bikaner in northwestern India. *Am J Trop Med Hyg* 2009; 80: 194-8.
31. Prakash J, Singh AK, Kumar NS *et al*. Acute renal failure in *Plasmodium vivax* malaria. *J Assoc Physicians India* 2003; 51: 265-7.
32. Maheshwari A, Singh AK, Sinha DK *et al*. Spectrum of renal disease in malaria. *J Indian Med Assoc* 2004; 102: 143.
33. Beg MA, Khan R, Baig SM *et al*. Cerebral involvement in benign tertian malaria. *Am J Trop Med Hyg* 2002; 67: 230-2.
34. Harish R, Gupta S. *Plasmodium vivax* malaria presenting with severe thrombocytopenia, cerebral complications and hydrocephalus. *Indian J Pediatr* 2009; 76: 551-2.
35. Hazra BR, Chowdhury RS, Saha SK *et al*. Changing scenario of malaria: A study at Calcutta. *Indian J Malariol* 1998; 35: 111-6.
36. Srinivasan V, Spinella PC, Drott HR *et al*. Association of timing, duration, and intensity of hyperglycaemia with intensive care unit mortality in critically ill children. *Pediatr Crit Care Med* 2004; 5: 329-36.

ORIGINAL ARTICLE

JIACM 2021; 22 (3-4): 99-103

Comparison of Non-Invasive Scoring Systems with Ultrasound and Liver Elastography in Predicting Non-Alcoholic Fatty Liver Disease in Healthy Population

Kartik Balankhe, Rishabh Ramu Nayak*, Rajesh Kumar Modi**, Pulin Kumar Gupta***, Princi Jain****, AK Varshney***, Kuldeep Singh****, Gurmeet Kaur***, Nitin Sinha****

Abstract

***Introduction:** Non-Alcoholic Fatty Liver Disease (NAFLD) is a common but frequently overlooked entity in the general population. Though liver biopsy is the gold standard, Ultrasound (USG) is the benchmark modality for diagnosing NAFLD. Since it is observer dependent and subjective, hence newer markers or scoring systems are the need of the hour.*

***Methods:** 55 apparently healthy individuals were recruited as cases for the study and subjected to 2-D USG, Transient elastography (TE) and routine laboratory investigations and various scores were calculated. Appropriate statistical methods were applied.*

***Results:** Amongst all 55 cases, the prevalence of NAFLD as per USG and TE was found to be 32.73% and 30.91% respectively. A statistically significant correlation was found between NAFLD and NAFLH- liver fat score (NAFLD-LFS) (P = 0.046), Aspartate aminotransferase to platelet ratio index (APRI) (P = 0.006) and Fibrosis-4 Score (FIB-4) (P = 0.011). Multivariate analysis revealed only NAFLD-LFS to be a significant independent predictor of NAFLD in healthy population. No correlation of Lipid accumulation product (LAP) score, Hepatic Steatosis Index (HSI), Fatty Liver Index (FLI) and Homoeostatic Model Assessment of Insulin Resistance/Beta (HOMA-IR) score was found with the occurrence of NAFLD.*

***Conclusion:** NAFLD-LFS, FIB-4 and APRI scores can be used for the diagnosis of NAFLD. These are cheap, precise, handy tools with good objectivity and hence may be used for monitoring of disease in future.*

Introduction

Non-alcoholic fatty liver disease (NAFLD) has become one of the most important emerging public health issues. NAFLD represents a spectrum of liver disease ranging from simple fatty infiltration (steatosis) to significant inflammation, i.e., steatohepatitis (NASH) leading to fibrosis and cirrhosis, in the absence of excessive alcohol consumption.

Liver biopsy is the conventional gold standard technique to confirm NAFLD, but it is an expensive, invasive procedure that needs hospitalisation and is associated with the rare risk of death. Ultrasonography (USG) of the liver is an optimum, accurate and reliable non-invasive approach for the detection of NAFLD and has become the imaging modality of choice, replacing liver biopsy for the diagnosis of NAFLD[1]. However, USG abdomen is an observer dependent, time consuming qualitative system and is not useful for serial measurements or monitoring of the disease. Another non-invasive technique is Transient Elastography (TE), marketed and commonly known as fibroscan, that measures liver fibrosis by measuring the liver stiffness. This technique takes approximately five minutes to perform, is painless and does not require fasting, sedation, or analgesia[2]. However, it is again expensive and meant primarily for diagnosing fibrosis and not simple steatosis, is not available everywhere and is not ideal for repeated evaluation, especially in a developing country like India.

To overcome these shortcomings, many easily available non-invasive scoring systems such as NAFLD-Liver Fat Score (NAFLD-LFS), Fibrosis-4 Score (FIB-4), Lipid accumulation product (LAP) score, Hepatic Steatosis Index (HSI), Fatty Liver Index (FLI) and aspartate aminotransferase-to-Platelet Ratio Index (APRI) have been proposed and are being used frequently. However, their validation in a normal healthy population is still lacking and very little literature is available regarding the same in this part of the world and hence the present study was executed.

Methods

This was a cross-sectional observational study done among fifty five non-alcoholic, non-obese (BMI < 25kg/1.76 m^2), healthy individuals, less than sixty years of age without any co-morbidities. These subjects were enrolled from the community, i.e., apparently healthy relatives of patients visiting the hospital. All subjects with history of diabetes,

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hypertension, dyslipidaemia, metabolic syndrome, cardiac, kidney or liver disease, or on any medication including vitamin/calcium supplements were excluded. All cases were subjected to a thorough history and examination. 10 ml fasting venous sample was withdrawn for routine laboratory parameters. Ultrasound was done using Samsung-Medison Ultrasound Machine (serial number-SOQQM3HF400117L) and was performed by a single observer using a 2-5 MHz convex transducer and staged as grade 1 (when the echogenicity is just increased), grade-2 (when the echogenic liver obscures the echogenic walls of portal vein branches) and grade-3 (when the echogenicity of liver obscures the diaphragmatic outline). We could not use liver biopsy – the gold standard in diagnosis of NAFLD – as cases subjects did not give consent and also deemed it ethically incorrect to subject healthy individuals to an invasive procedure. Instead we used USG as a surrogate gold standard for the diagnosis as it is also a highly accurate and non-invasive method to pick up steatosis and early fibrosis. Transient elastography/fibroscan was done using fibroscan 402, ECHOSENS by a single observer. It was performed with a curved array ultrasound probe at 4 MHz for B-mode imaging. A normal liver's shear stiffness was taken between 6.5-7 kPa. Ten successful acquisitions were performed in each patient, and the median value was determined and used as a representative measurement of the liver elasticity. We did not use the CAP parameter as it is not measured in our machine.

The following non-invasive scores were calculated for every subject.

1. FIB-4 score was calculated by using age, liver enzymes values (AST/ALT) and platelet counts. A FIB-4 score of < 1.3 indicates the absence of advanced disease and the presence of fibrosis denoted by a score of ≥ 1.3[3].
2. NAFLD liver fat score (NAFLD-LFS) uses the values of AST/ALT and whether T2DM is present or not. Value ≤ 0.640 rules-out and values > 0.640 rules in NAFLD[4].
3. Hepatic steatosis index (HSI) includes AST/ALT, BMI, sex and DM presence or absence in calculations. With values < 30 ruling-out, and values > 36 ruling in steatosis[4].
4. Fatty liver index (FLI) uses BMI, triglycerides, and waist size for calculations. Values < 30 rules-out and values ≤ 60 rules in steatosis[4].
5. Aspartate aminotransferase-to-platelet ratio index (APRI) uses AST and platelet count for determination of liver fibrosis. At a threshold value of ≤ 0.3, it rules-out significant fibrosis; and at a threshold of ≤ 1.5, it rules in significant fibrosis[5].
6. Lipid Accumulation Product (LAP) Index is calculated using triglyceride levels and waist circumference. The cut-off values for LAP in men and women were taken as 30.5 and 23.0 respectively[6].
7. HOMA-IR and HOMA-Beta uses fasting glucose and insulin levels[7].

Statistical Analysis

Categorical variables are presented in number and percentage (%) and continuous variables are presented as mean ± SD and median. P value ≤ 0.05 was considered statistically significant. The data was entered in MS EXCEL spreadsheet and analysis was done using Statistical Package for Social Sciences (SPSS) version 16.0 (IBM, Chicago).

Results

Amongst all 55 subjects (12 males and 43 females), 37 (67%) had ultrasonographically documented normal liver morphology (gpA) and 18 (33%) had NAFLD (gpB) out of which 12 had grade 1, six had grade 2 and none had grade 3 NAFLD. Majority of the cases were in the age group of 30 - 50 years and the mean age was 43.4 ± 12.4 years. The prevalence of NAFLD was found to be 32.73% by USG and 30.91% by liver elastography. The laboratory parameters of all subjects have been depicted in Table I.

Table I: Laboratory parameters amongst all subjects.

Haematological and biochemical parameters	Cases (n = 55)
Haemoglobin (mg/dl)	12.68 ± 1.27
Total leucocyte count (per cubic mm)	6817.45 ± 1836.36
Platelet count (per cubic mm)	270 ± 84
Urea (mg/dl)	37.24 ± 12
Serum creatinine (mg/dl)	0.61 ± 0.27
Uric acid (mg/dl)	4.21 ± 0.95
Total bilirubin (mg/dl)	0.61 ± 0.22
Direct billirubin (mg/dl)	0.24 ± 0.08
Indirect bilirubin (mg/dl)	0.36 ± 0.18
Aspartate transaminase (U/L)	34.11 ± 11.79
Alanine transaminase (U/L)	32.98 ± 10.34
Alkaline phosphatase (U/L)	70.71 ± 21.28
Total protein (gm/dl)	7.38 ± 0.8
Albumin (gm/dl)	4.23 ± 0.63
Globulin (gm/dl)	3.16 ± 0.57
Total cholesterol (mg/dl)	173.33 ± 33.93
HDL (mg/dl)	45.71 ± 11.69
LDL (mg/dl)	92.33 ± 31.98
Triglyceride (mg/dl)	168.36 ± 51.33
Gamma-glutamyl transferase (U/L)	33.65 ± 24.65

The mean waist circumference was found to be 85.97 ±

9.26 cm and 89.5 ± 10.34 cm amongst cases in gpA and gpB, respectively (p = 0.207). Similarly, no difference was found in mean body mass index between gpA and gpB [24.7 ± 2.7 kg/m^2 vs 24.9 ± 2.6 kg/m^2 respectively (p = 0.775)].

The mean shear pressure value in liver elastography (kPa) in Group B was 7.86 ± 0.87, which was significantly higher as compared to 4.75 ± 1.25 in Group A (p < 0.001).

Table II: Comparison of mean shear pressure between gp A and gp B.

Fibroscan (kPa)	Group A (n = 37)	Group B (n = 18)	p value
< 7	36 (97.30%)	2 (11.11%)	< .0001
7 - 8.6	1 (2.70%)	13 (72.22%)	
8.7 - 10.2	0 (0%)	3 (16.67%)	
Mean ± Stdev	4.75 ± 1.25	7.86 ± 0.87	< .0001
Median (IQR)	4.6 (4.1 - 5.4)	7.85 (7.375 - 8.1)	
Range	1.9 - 7.3	5.8 - 9.5	

The mean shear pressure was found to be significantly higher in cases with grade 2 fatty liver (8.68 ± 0.68) as compared to 7.44 ± 0.64 in cases with grade 1 fatty liver (p = 0.001) implying that not only liver elastography accurately picks up NAFLD, it can even significantly stage its severity.

Table III: Association of non invasive scoring systems with ultrasonographically proven NAFLD.

Non-invasive scores of NAFLD	Group A (n = 37)	Group B (n = 18)	P value
HIS			
< 30	4 (10.81%)	2 (11.11%)	0.512
30 - 36	26 (70.27%)	10 (55.56%)	
> 36	7 (18.92%)	6 (33.33%)	
Mean ± Stdev	33.47 ± 2.88	35.14 ± 4.88	0.116
FLI			
< 30	13 (35.14%)	5 (27.78%)	0.748
30 - 59	18 (48.65%)	9 (50%)	
> = 60	6 (16.22%)	4 (22.22%)	
Mean ± Stdev	38.86 ± 16.71	42.94 ± 22.81	0.455
NAFLD-LFS			
< = .64	37 (100%)	18 (100%)	
Mean ± Stdev	-3.1 ± 1.33	-2.28 ± 1.28	0.046
APRI			
< = .3	30 (81.08%)	8 (44.44%)	0.006
.4 - 1.4	7 (18.92%)	10 (55.56%)	
Mean ± Stdev	0.28 ± 0.13	0.48 ± 0.29	0.01
FIB-4			
< 1.3	32 (86.49%)	10 (55.56%)	0.011
> = 1.3	5 (13.51%)	8 (44.44%)	
Mean ± Stdev	0.88 ± 0.39	1.53 ± 0.72	0.001
HOMA-IR			
< 2	26 (70.27%)	15 (83.33%)	0.346
> = 2	11 (29.73%)	3 (16.67%)	
Mean ± Stdev	1.87 ± 1.11	1.51 ± 0.58	0.197
HOMA-Beta			
< = 86.2	24 (64.86%)	14 (77.78%)	0.372
> 86.2	13 (35.14%)	4 (22.22%)	
Mean ± Stdev	67.97 ± 90.26	56.94 ± 69.82	0.65
LAP for women			
< 23	2 (10%)	1 (10%)	1
> 23	18 (90%)	9 (90%)	
LAP for men			
< 30.5	3 (17.65%)	1 (12.50%)	1
> 30.5	14 (82.35%)	7 (87.50%)	
LAP			
Mean ± Stdev	45.43 ± 19.2	53.67 ± 21.26	0.155

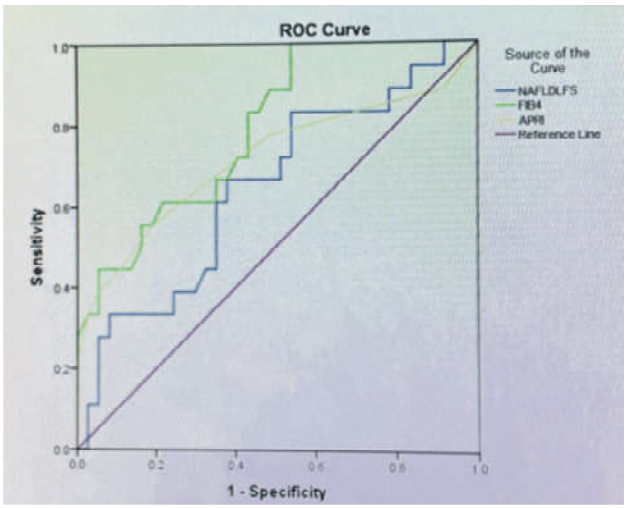

Fig. 1: *Comparison of receiver operating characteristic curve of NAFLD-LFS, APRI, FIB-4 in predicting NAFLD in study subjects.*

The AUROC for NAFLD-LFS to diagnose NAFLD was found to be 0.643 (CI = 0.481 - 0.801). At a value of ≥ - 3.37, the sensitivity and specificity to diagnose NAFLD was found to be 77.2% and 51% respectively. The AUROC for FIB - 4 to diagnose NAFLD was found to be 0.789 (CI = 0.661 - 0.911). At a value of ≥ 0.895, the sensitivity and specificity to diagnose NAFLD was found to be 72% and 60% respectively.

Similarly the AUROC for APRI to diagnose NAFLD was found to be 0.715 (CI = 0.55 - 0.87). At a value of ≥ 0.250, the sensitivity and specificity to diagnose NAFLD was found to be 78% and 52% respectively.

A statistically significant higher value of NAFLD-LFS score (p = 0.046) was found in gp B as compared to gp A. Higher values of APRI (p = 0.006) and FIB-4 score (p = 0.011) were observed in gp B indicating that healthy population with NAFLD not only can have just simple fatty liver but concomitant liver fibrosis also. Hence, these three scores, i.e., NAFLD-LFS, APRI and FIB-4, needing just a platelet count, liver function tests and basic lipid profile, can accurately pick up most cases with NAFLD in normal healthy population. There is always a trade-off between sensitivity and specificity so while choosing that variable as best in which a combination of sensitivity and specificity gives the maximum predictive value, i.e., maximum diagnostic accuracy and hence overall NAFLD-LFS, FIB-4 and APRI were found to be the best predictors of NAFLD and all three markers together can satisfactorily pick up most cases of NAFLD and also rule-out false positives. Since these three scoring systems depend upon three basic routine cheap tests, i.e., LFT, lipid profile and platelets counts, hence not only these novel scoring systems are affordable, repeatable but may also be used for monitoring of the disease. HSI, FLI, and LAP were not found to be predictive of NAFLD. The traditional markers of central obesity and NAFLD like HOMA-IR and HOMA-α were not found to be helpful in predicting NAFLD in our healthy non obese population. On performing multivariate logistic regression analysis, only NAFLD-LFS was found to be a significant independent predictor of NAFLD in healthy population (p < 0.01).

Discussion

Various risk factors like central obesity, diabetes, dyslipidaemia and metabolic syndrome have long been regarded as predictors of NAFLD[8]. Age, gender, and ethnicity also influence the prevalence of NAFLD as mentioned in a study by Chalasaini N *et al* in 2012[2]. During the selection of cases for this study, we excluded all people with obesity, diabetes, dyslipidaemia and metabolic syndrome; hence the biasing by these risk factors was automatically removed. USG and TE were found to be suggestive of NAFLD in 32.7% and 30.9% of subjects respectively in our study, which correlates well with the data available in literature suggesting 30 - 35% NAFLD in healthy Indian population. In our study, NAFLD-LFS, FIB-4 and APRI were also found to be significantly predictive of NAFLD in our healthy subjects.

Kotronen A *et al* in 2009 stated that NAFLD-LFS score can be helpful in identification of NAFLD using easily available variables[9]. On the contrary Kahl S *et al* in 2004 did not find NAFLD-LFS suitable for the prediction of hepato-cellular lipid content (quantified by magnetic resonance spectroscopy) at least in the non-obese and non-diabetic individuals[4]. In our cases, the mean NAFLD-LFS in group B (-2.28 ± 1.28) was significantly higher than that in group A (-3.1 ± 1.33) (p = 0.046) and the most significant to suggest NAFLD amongst all scoring systems. This was similar to the study by Chueng *et al* in 2014, in which NAFLD-LFS was found to be the best non-invasive prediction score for NAFLD out of FLI, HIS, LAP and NAFLD-LFS[10].

FIB-4 marker was initially derived in patients in hepatitis C and HIV co-infection by Dyson J *et al* in 2014 and they found FIB-4 to be one of the most useful non-invasive tests for diagnosing advanced fibrosis in NAFLD[11]. In our study also, 86.49% of subjects without NAFLD had FIB-4 score < 1.3 while 44.44% of subjects with NAFLD had FIB-4 score >/=1.3, both of which were significant. (p = 0.011). Further, the mean of FIB-4 in cases with NAFLD was 1.53 which was significantly higher as compared to mean 0.88 in those without NAFLD (p = 0.001).

Kruger FC *et al* in 2010 found that APRI compared favourably to NAFLD Liver Fibrosis Score (which was already a validated marker) and was superior to AST/ALT for the prediction of advanced fibrosis[12]. Even in our study, 81% of cases in gpA had APRI of </= 0.3 and 55% of cases with NAFLD, i.e., gpB had APRI in the range of 0.4 - 1.4, both of which were significant (p = 0.006). The median of APRI in cases with NAFLD was 0.48 which was significantly higher as compared to 0.28 amongst cases without NAFLD (p = 0.01).

Bedogni G *et al* in 2006 found FLI to be an accurate and simple predictor of hepatic steatosis in the general population[13]. Huang X *et al* in 2015 also demonstrated that FLI could detect NAFLD accurately in the middle-aged and elderly Chinese population[14]. We could not find any significant association between FLI with ultrasonographically documented NAFLD (p = 0.455). Similarly in our study, no significant association was seen between HIS and NAFLD (p = 0.116). This was contrary to the results of Lee H *et al* in 2010 who concluded HIS to be a simple and efficient screening tool for NAFLD in the general population[15]. The reason behind no correlation of FLI and HIS with NAFLD in our study may be that BMI is an important component of HIS and FLI and hence, having already chosen only those people who were non-obese with mean BMI 24.7 kg/m^2, the absence of correlation between FLI and HIS and NAFLD in our study can thus be explained.

Fujii H *et al* in 2019 concluded that HOMA-IR is an independent predictor of advanced fibrosis in non-diabetic patients with NAFLD[16]. In a study conducted by Salgado AL *et al* in 2010, the universal concurrence of insulin resistance (IR) was suggestive of it being a parameter for the diagnosis

of NAFLD[17]. However, no correlation of HOMA-IR was found with NAFLD in our subjects. In fact, mean HOMA-IR was numerically lower (1.51 +/- 0.58 vs 1.87 +/- 1.11) in cases without NAFLD (p = 0.19). The reason for this is that HOMA-IR is mostly useful in comparing IR between or within the groups. It has little relevance in individual patients due to several factors including non-standardised insulin assays and the pulsatile insulin secretion occurring normally in every individual.

In a study conducted by Siddiqui MS *et al* in 2005, the subjects with NAFLD had higher HOMA-α as compared to both lean and obese controls[8]. In our study, no significant association between HOMA- α and NAFLD was found to be present in cases (p = 0.746). Most of our cases with NAFLD had grade-1 steatosis and very few cases with NAFLD in our study had evidence of high-grade inflammation in the liver, (i.e., grade 2 and 3 fatty liver) and hence, pancreatic damage might be absent or only minimally present in these cases. Also, since all the cases were already healthy (without any obvious evidence of inflammation anywhere), the pancreatic beta-cell functions were expected to be relatively preserved hence with no correlation found between NAFLD and HOMA-α.

Bedogni G *et al* in 2010 found LAP to be a reasonably accurate approach to identify individuals with ultrasonographic liver steatosis[18]. In our study, no significant association was seen between LAP and NAFLD (p = 0.98). Waist circumference is a surrogate measure of central obesity and it is a component of LAP score as well and hence because of the lack of significant central obesity in our cases (mean waist circumference = 87.13 cm) no correlation was found between LAP and NAFLD. The limitation of our study was that we did not use the gold standard technique of liver biopsy in diagnosing NAFLD. Also, the controlled attenuation parameter (CAP) was not available in our hospital machine, hence may be we could have missed out on very mild cases.

Conclusion

NAFLD is rampant even in non obese healthy population of India. USG abdomen has been the gold standard investigative modality of choice but it is expensive, observer dependant, not meant for repetitive monitoring and in this COVID-19 era may not be advisable (because of the need to visit diagnostic centre and the risk of transmission of COVID-19 via bed linen). NAFLD-LFS, FIB-4 and APRI score can be a useful tool in the diagnosis and monitoring of the disease. Not only are these novel scoring systems cheaper, but are also reproducible, repetitive, bedside, and can be used for monitoring of the disease. However, long-term prospective studies with large number of cases are required for further validation.

References

1. Hernaez R, Lazo M, Bonekamp S *et al*. Diagnostic accuracy and reliability of ultrasonography for the detection of fatty liver: a meta-analysis. *Hepatology* 2011; 54 (3): 1082-90.
2. Chalasani N, Younossi Z, Lavine JE *et al*. The diagnosis and management of non-alcoholic fatty liver disease: practice guideline by the American Gastroenterological Association, American Association for the Study of Liver Diseases, and American College of Gastroenterology. *Gastroenterology* 2012; 142 (7): 1592-609.
3. Shah AG, Lydecker A, Murray K *et al*. use of the FIB4 index for non-invasive evaluation of fibrosis in nonalcoholic fatty liver disease. *Clin Gastroenterol Hepatol* 2009; 7 (10): 1104-12.
4. Kahl S, Straburger K, Nowotny B *et al*. Comparison of Liver Fat Indices for the Diagnosis of Hepatic Steatosis and Insulin Resistance. *PLoS One* 2014; 9 (4): 940-59.
5. Loaeza-del-Castillo A, Paz-Pineda F, Oviedo-Cárdenas E *et al*. AST to platelet ratio index (APRI) for the noninvasive evaluation of liver fibrosis. *Ann Hepatol* 2008; 7 (4): 350-*57*.
6. Dai H, Wang W, Chen R *et al*. Lipid accumulation product is a powerful tool to predict non-alcoholic fatty liver disease in Chinese adults. *NutrMetab* 2017; 14 (1): 49-54.
7. Singh Y, Garg MK, Tandon N *et al*. A Study of insulin resistance by HOMA-IR and its cut-off value to identify metabolic syndrome in urban Indian adolescents. *J Clin Res Pediatr Endocrinol* 2013; 5 (4): 245-51.
8. Siddiqui MS, Cheang KL, Luketic VA *et al*. Non alcoholic steato-hepatitis (NASH) is associated with a decline in pancreatic Beta cell (β-Cell) function. *Dig Dis Sci* 2015; 60 (8): 2529-37.
9. Kotronen A, Peltonen M, Hakkarainen A *et al*. Prediction of Non-Alcoholic Fatty Liver Disease and Liver Fat Using Metabolic and Genetic Factors. *Gastroenterology* 2009; 137 (3): 865-72.
10. Cheung CL, Lam KSL, Wong ICK *et al*. Non-invasive score identifies ultrasonography diagnosed non-alcoholic fatty liver disease and predicts mortality in the USA. *BMC Med* 2014; 12 (1): 154-9.
11. Dyson JK, Anstee QM, McPherson S. Non-alcoholic fatty liver disease: a practical approach to diagnosis and staging. *Frontline Gastroenterol* 2014; 5 (3): 211-8.
12. Kruger FC, Daniels CR, Kidd M *et al*. APRI:A simple bedside marker for advanced fibrosis that can avoid liver biopsy in patients with NAFLD/NASH. *South African Med J* 2011; 101 (7): 477-80.
13. Bedogni G, Bellentani S, Miglioli L *et al*. The fatty liver index: A simple and accurate predictor of hepatic steatosis in the general population. *BMC Gastroenterol* 2006; 6 (1): 1-7.
14. Huang X, Xu M, Chen Y *et al*. Validation of the fatty liver index for nonalcoholic fatty liver disease in middle-aged and elderly Chinese. *Med (United States)* 2015; 94 (40): 1-7.
15. Lee JH, Kim D, Kim HJ *et al*. Hepatic steatosis index: A simple screening tool reflecting nonalcoholic fatty liver disease. *Dig Liver Dis* 2010; 42 (7): 503-08.
16. Fujii H, Imajo K, Yoneda M *et al*. HOMA-IR: An independent predictor of advanced liver fibrosis in non-diabetic non- alcoholic fatty liver disease. *J Gastroenterol Hepatol* 2019; 34 (8): 1390-95.
17. Salgado ALFDA, De Carvalho L, Oliveira AC *et al*. Insulin resistance index (HOMA-IR) in the differentiation of patients with non-alcoholic fatty liver disease and healthy individuals. *Arq Gastroenterol* 2010; 47 (2): 165-9.
18. Bedogni G, Kahn HS, Bellentani S *et al*. A simple index of lipid over accumulation is a good marker of liver steatosis. *BMC Gastroenterol* 2010; 10 (1): 98-102.

ORIGINAL ARTICLE

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Professional Challenges Encountered by Healthcare Professionals and its Impact on their Well-Being during the COVID-19 Pandemic

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Abstract

Background: The unprecedented public health concern of COVID-19 pandemic poses challenges at structural and psychosocial levels, increases the risk of psychological morbidity and affects the well-being of healthcare professionals (HCPs).

Objective: The study aimed to understand the professional challenges faced by HCPs during the COVID-19 pandemic in India. It also aimed to understand the psycho-social impact of these challenges on the subjective well-being of HCPs.

Method: A cross-sectional web-based survey was designed, consisting of twenty-eight multiple choice or Likert-type questions. The survey had four mental health domains to be explored.

It was an anonymous survey with online informed consent and was circulated on social media groups of doctors and nurses in Delhi, India using snow-ball method.

Multivariate analysis of variance (MANOVA) was performed to study demographic and COVID-19 related factors on psychological and occupational variables. Further, thematic analysis and Latent Dirichlet Allocation (LDA) were used for short answer qualitative questions.

Results: Results of 253 HCPs showed that those dealing with COVID-19 patients, scored lower on perceived occupational preparedness ($F_{1,251} = 6.266$, $p < 0.0001$), work satisfaction ($F_{1,251=}$ 40.998, $p < 0.0001$) and well-being ($F_{1,251} = 53.529$, $p < 0.0001$) as compared to non-COVID-19 duty HCPs; although they displayed less challenges in adaptation and protection ($F_{1,251}=17.413$, $p < 0.0001$). Based on LDA and thematic analysis, seven clusters were identified each- to understand the greatest struggle and subsequent support needed by HCPs during the pandemic.

Conclusions: The study highlights the immediate need to protect the mental health of HCPs by establishing comprehensive psychological interventions and services.

Key words: Covid-19, Well-being, Healthcare professionals, Latent Dirichlet Allocation.

Introduction

The COVID-19 pandemic reached the Indian subcontinent in January 2020[4], and impacted the healthcare, lifestyles, and economy of the country. In India, the first case of COVID-19 was reported on January 30, 2020 and by 17th May, 2020 the number of cases stood at an enormous 90,927 (approx.) infected individuals and 2,872 deaths; and these numbers have only been surging higher since then[5].

In facing and fighting COVID-19, the role of health care professionals (HCPs) is of prime importance[6]. However, emerging literature around COVID-19 in India shows that healthcare workers face unparalleled challenges. With the increasing number of cases, overwhelming workload, lack of PPE kits, wide media coverage and difficulties in implementing health policies, serious concerns about the safety of frontline medical staff have been raised[7,8]. Previous research focusing on SARS or H1N1 disease has also highlighted, how these circumstances result in adversities for HCPs[9]. High levels of psychological distress, extreme uncertainty, vulnerability, threat to life, anxiety, and increased depressive symptoms among healthcare workers, across different countries were reported[10-12]. Recent studies, centering specifically on the mental health of frontline healthcare workers, during COVID-19, have also established similar symptoms[6].

Risks to healthcare workers, can be intensified by pre-existing gender roles as well. For instance, it was observed in a study, that most healthcare workers in emergency departments were female nurses, and they experienced amplified distress due to the gendered expectations[6, 13]. Distress and disengagement are further elevated in

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healthcare workers during such pandemics due to the additional fear of risk of infection to family, relatives, and colleagues[9].

Working in difficult conditions often leads to dissatisfaction and negative organisational performance[14]. Further, psychosocial and work environment factors have a direct impact on job satisfaction[15]. However, in India, in addition to fighting the deadly disease, healthcare workers are also facing out-turns of stigma[16]. Cases of physical violence and assault of health workers have been reported at several places[8].

Persistent challenges during the COVID-19 pandemic in India have thus heightened the psychological burden experienced by HCPs, posing a threat to their well-being. Studies pertaining to HCPs mental health outcomes and interventions during COVID-19 outbreak are the need of the hour, but are relatively scarce in India. To address this gap, the present study had two aims. The primary aim was to explore the professional challenges encountered by HCPs during COVID-19 pandemic. Secondarily, the study aimed to understand the psycho-social impact of these challenges on the subjective well-being of HCPs. Through this, it aims at providing insights into the mental health of Indian HCPs and pave the way for timely and comprehensive actions to be taken to protect their mental health.

Material and Methods

Study design

The current study was conducted from the first phase till the second phase of lockdown during the COVID-19 pandemic in India, i.e., from 25th March 2020 - 17th May 2020. For this purpose, a cross-sectional web-based survey was designed to explore the nature of professional challenges encountered by HCPs and its impact on their well-being.

***Development of the Survey*:** The survey questions were based on previous pandemic researches and available literature on COVID-19. Furthermore, brief informal interviews were conducted with five experienced physicians, to identify the key challenges faced by HCPs and its bearing on their mental health, relationship with family, and occupational engagement and satisfaction. Utilizing these anchor points, questions were formulated, which were further subjected to face validity by a working group of senior researchers and practicing physicians and were accordingly reviewed and revised. These questions were thereafter pilot tested on a subset of intended participants. Based on the validation process, some questions were discarded from the item pool and subsequently, final survey consisted of 28 questions apportioned into three sections.

1. Socio-Demographics: This section focused on the demographic details and professional characteristics such as age, sex, region, occupational designation (doctors, nurses, or paramedics) and workplace details.
2. Changing dynamics and negotiating professional challenges explored the psycho-social impact of the pandemic on the HCPs professional and personal roles and responsibilities. A total of 21 questions were distributed among three domains, i.e., Perceived Occupational Preparedness, Work Satisfaction, and lastly Adaptation and Protection.

   These sub-domains were chosen, based on researches highlighting the impact of a pandemic on HCPs, bringing to attention factors like-overwhelming workload, insufficient knowledge, depleted safety resources, inadequate support, and fear of contagion to one's family, as sources of psycho-emotional stress[17,18]. Additionally, with the confinement norms, increased teleconsulting, managing multiple COVID-19 related queries, and indirect patient care, further manifested as dissatisfaction and discontent among the HCPs[9,19].
3. Well-Being and coping was directed at investigating the HCPs overall well-being status and the coping mechanisms adopted by them. This becomes critical, since occupational and emotional distress, directly impinge on one's subjective well-being[6]. The well-being profile assessed physical, behavioural, cognitive and emotional parameters, as well as the sense of autonomy experienced. These domains were borrowed from the well-being models of Diener[20] and Ryff[21]. Furthermore, inspiration from various scales of subjective well-being was drawn, to identify the key aspects, while ensuring a short well-being profile.

Questions were then transferred in a Google form. For multiple-choice questions, response options were organised on a 5-point Likert scale, ranging from lowest to highest, in the form of "not at all to extremely" respectively. Two short answer questions were also incorporated to gain insights about the biggest struggle confronted and the consequent aid required by the HCPs. Lastly, a brief question aimed at understanding the multiple coping mechanisms adopted, was included. Some of the questions were negatively directed to warrant valid responses amongst participants. The survey was designed to be comprehensive yet short and user friendly, requiring 8 - 10 minutes for completion. The form was then shared with the target population using a URL link.

Participants

Participants were recruited through purposive and snowball sampling. The inclusion criteria for participants was: to be a practicing medical professional, i.e., doctor, nurse, and paramedic. Those without an encounter with COVID-19 patients were also eligible. Those without completion of their medical degrees, were excluded.

Procedure

The link along with a brief rationale of the study was circulated among professional networks of practicing medical professionals, which was further shared with their professional colleagues and friends. The reach of the study was widened by spreading it on social media platforms such as Facebook groups of doctors' and nurses' bodies, groups of working residents across government and private hospitals, WhatsApp groups and the like. Gentle reminders were consistently sent once a week to ensure maximum participation. After explaining the study, informed consent was obtained using an electronic agreement, and the survey was available for completion for a period of 54 days.

Statistical methods

Data gathered was analysed using descriptive and inferential statistics, by employing SPSS for windows version 23[22]. Multivariate analysis of variance (MANOVA) was used to see effect of all socio-demographic and COVID-19 related factors on four main domains,i.e., Perceived Occupational Preparedness, Work Satisfaction, Adaptation and Protection, and Well- being Parameters.

The data gathered was transported onto a spreadsheet and coded, to ensure confidentiality of all the participants. For analysis, all tests were conceived to be 2-tailed and the significance level was set at $\alpha = 0.05$.

Short answer questions were analysed using thematic analysis and run through an unsupervised Machine Learning model, known as Latent Dirichlet Allocation (LDA), to ensure rigour in analysis.

Results

Demographic and Psychological Profile of the sample (Table 1): A total of 264 participants responded to the survey. Out of these 11 were from abroad and were excluded. Sample (N = 253) was equally represented by both sexes, and also among those who were posted for COVID-19 duty (hereafter referred as CoD) and those not involved in COVID duty (Non-COVID-19, hereafter referred as N-CoD). CoD participants were significantly younger than N-CoD participants ($F_{1,251}$ =27.08 ($p < 0.0001$). Sample included 176 doctors, 66 nurses and only 11 paramedics. 31% of doctors were on CoD while 48% nurses were performing COVID-19 related duties. Significantly more government employees took the survey among CoD persons while among N-CoD participants significantly more were private practitioners ($F_{1,251} = 39.39$, $p < 0.0001$). Majority of CoD participants were from outside Delhi NCR ($F_{1,251} = 19.31$, $p < 0.0001$).

In case of psychological profile, N-CoD participants perceived better occupational preparedness ($F_{1,251} = 28.606$, $p < 0.0001$), more work satisfaction ($F_{1,251} = 77.205$, $p < 0.0001$); more difficulty in adaptation and protection ($F_{1,251} = 36.031$, $p < 0.0001$) and better score on wellbeing parameters ($F_{1,251}$ 98.135, $p < 0.0001$), than CoD participants (Table I).

Table I: Demographic and psychological profile of the sample.

	COVID-19 duty (CoD)	No COVID-19 duty (N-CoD)	Chi square/ F-value	p-value
Gender Male/Female	45/45	81/82	0.002	0.534
Age in years	35.28 ± 8.842	42.92 ± 12.282	27.077	< 0.0001
Occupation Doctor/Nurse/Paramedic	54/32/4	122/34/7	6.57	0.038
Type of working place Government/private	68/22	56/107	39.39	< 0.0001
Type of duty 1/2/3/4*	43/29/10/8	2/20/50/91	124.57	< 0.0001
Region Delhi NCR/Outside Delhi	40/118	50/45	19.31	< 0.0001
Perceived occupational Preparedness	9.74 ± 3.33	12.05 ± 3.20	28.606	< 0.0001
Work satisfaction	34.56 ± 6.78	42.55 ± 7.01	77.205	< 0.0001
Adaptation and protection	8.33 ± 2.97	10.95 ± 3.50	36.031	< 0.0001
Well- being parameters	61.60 ± 9.68	74.14 ± 9.61	98.135	< 0.0001

**COVID-19 ward/ Emergency ward/General ward/General OPD*

Changing dynamics and negotiating professional challenges: MANOVA was performed to see the effect of various demographic and COVID-19 related factors on all outcome variables (Table II). Analysis suggested that male ($F_{1,246} = 3.89$, $p = 0.041$); participants working in private facilities ($F_{1,246} = 19.163$, $p < 0.0001$) and participants from Delhi ($F_{1,246} = 4.207$, $p = 0.049$) had higher Occupational Preparedness scores than their counterparts. Those on CoD perceived lower Occupational Preparedness than N-CoD participants ($F_{1,246} = 6.266$, $p = 0.013$).

Delhi/NCR participants had better work satisfaction than outside Delhi participants ($F_{1,246} = 4.135$, $p = 0.043$) and CoD participants had less work satisfaction ($F_{1,246} = 40.998$,

$p < 0.0001$). There was no difference between males and females, and doctors and nurses.

Older participants had higher scores, indicating greater difficulty in adaptation and protection ($F_{1,246} = 9.358$, $p = 0.002$), and participants on CoD had better ability to adapt and protect themselves and their family ($F_{1,246} = 17.413$, $p < 0.0001$).

Further, participants with higher age had better well-being than younger age groups ($F_{1,246} = 19.426$, $p < 0.0001$. Similarly, those on CoD perceived less well-being than N-CoD participants ($F_{1,246} = 53.529$, $p < 0.0001$).

Table II: Effect of demographic parameters in CoD on perceived occupational preparedness, work satisfaction, adaptation and protection, and well-being parameters (only significant factors).

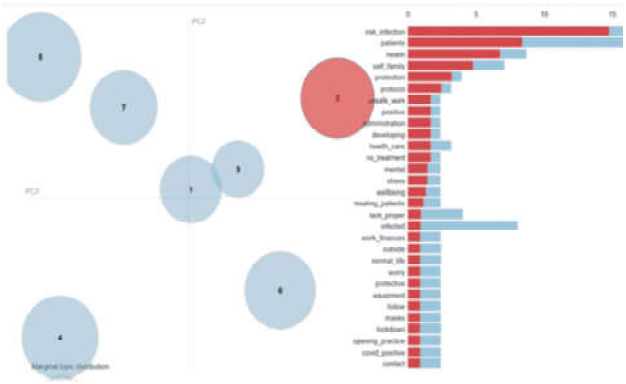
Outcome variable	Correlates	Mean sum of squares	F (1,246)	p-value
Perceived	Gender	36.610	3.897	.049
Occupational	Type of working place	180.000	19.163	.000
Preparedness	Region	39.521	4.207	.041
	COVID-19 duty	58.857	6.266	.013
Work satisfaction	Age	245.986	5.219	.023
	Region	194.905	4.135	.043
	COVID-19 duty	1932.211	40.998	.000
Changing dynamics and	Age	99.005	9.358	.002
perceived risk to family	COVID-19 duty	184.227	17.413	.000
Well- being	Age	1666.476	19.426	.000
Parameters	COVID-19 duty	4592.102	53.529	.000

Well-Being parameters of Health Professionals: MANOVA was computed to see the effect of socio-demographic and COVID-19 participation on the subdomains of wellbeing (Table III). Contrary to expectations, older persons had better physical health ($F_{1,246} = 8.16$, $p = 0.005$) while CoD participants had deterioration in physical health ($F_{1,246} = 26.097$, $p < 0.0001$). Younger professionals felt more behavioural disturbances ($F_{1,246} = 30.82$, $p < 0.0001$). In case of cognitive operations, older HCPs had better cognition than younger ($F_{1,246} = 10.343$, $p = 0.001$); persons with CoD had more cognitive disturbances than N-CoD ($F_{1,246} = 25.41$, $p < 0.0001$); and females experienced greater struggle in maintaining their cognitive operations than males ($F_{1,246} = 4.46$, $p = 0.036$). Males also showed fewer negative emotions ($F_{1,246} = 12.12$, $p = 0.001$) and more positive emotions ($F_{1,246} = 3.92$, $p = 0.049$). Higher age ($F_{1,246} = 9.12$, $p = 0.003$) and N-CoD professionals ($F_{1,246} = 39.09$, $p < 0.0001$) showed fewer negative emotions, while nurses expressed more negative emotions than doctors ($F_{1,246} = 5.37$, 0.021). Better sense of autonomy was related to higher age ($F_{1,246} = 19.52$, $p < 0.0001$), professionals working in Delhi NCR ($F_{1,246} = 5.73$, $p = 0.017$), and N-CoD ($F_{1,246} = 53.026$, $p < 0.0001$).

Table III: Effect of demographic parameters on various well-being parameters (only significant factors).

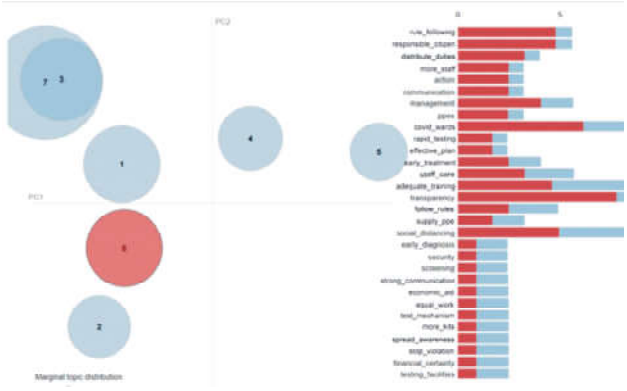
Outcome variable	Correlates	Mean sum of squares	F (1,246)	p-value
Physical health	Age	15.284	8.156	0.005
	COVID	48.903	26.097	0.000
Behavioral disturbances	Age	89.718	30.819	0.000
	COVID	132.564	45.537	0.000
Cognitive operations	Gender	8.992	4.463	0.036
	Age	20.840	10.343	0.001
	COVID	51.197	25.410	0.000
Negative emotions	Gender	273.075	12.121	0.001
	Age	205.475	9.120	.003
	Occupation	121.051	5.373	.021
	COVID	880.724	39.091	.000
Sense of autonomy	Age	49.603	19.518	.000
	Region	14.562	5.730	.017
	COVID	162.981	64.131	.000
Positive emotion	Gender	37.925	3.916	.049
Well being	Age	1902.205	21.348	.000
	COVID	4724.952	53.026	.000

Lastly, triangulation analysis, by employing thematic analysis



[1] *Other themes, as represented by different circular clusters are:- 1: On-field obstacles impacting professional role and duties, 3: Concerns over callous public behaviour, 4: Challenged work-life balance, 5: Struggling with financial insecurity, 6: Maintaining positive outlook and productivity, and 7: Bearing with constraints of lockdown in everyday life.*

Fig. 1: *Theme 2, titled- "Risk of Infection to self and family" with its salient codes, deduced by using statistical model- LDA (unsupervised machine learning technique).*



[2] *Other themes as represented by different circular clusters are:- 1: Improved availability of safety measures, 2: Responsible citizen behaviour, 3: Transparent communication and transmission of reliable information, 4: Regulated teamwork, 5: Financial assistance, and 7: Stern measures for implementation of safety norms.*

Fig. 2: *Theme 6, titled- "Keen surveillance and Organised planning" with its salient codes, deduced by using statistical model- LDA (unsupervised machine learning technique).*

and LDA, led to seven prominent clusters each, for both the questions, aimed at understanding the biggest struggle and subsequent support mechanisms needed by HCPs respectively (Fig. 1 and 2).

Coping strategies adopted

Question 27 required participants to choose coping mechanisms, from a list of coping strategies which they believed were helpful for them. Family support (81.4%) and receiving sufficient knowledge about the pandemic (76.3%) were two chief coping strategies adopted by all the HCPs. Further, those professionals engaged in N-CoD also employed other coping mechanisms like maintaining adequate connectivity, relying on the solidarity of their co-workers,

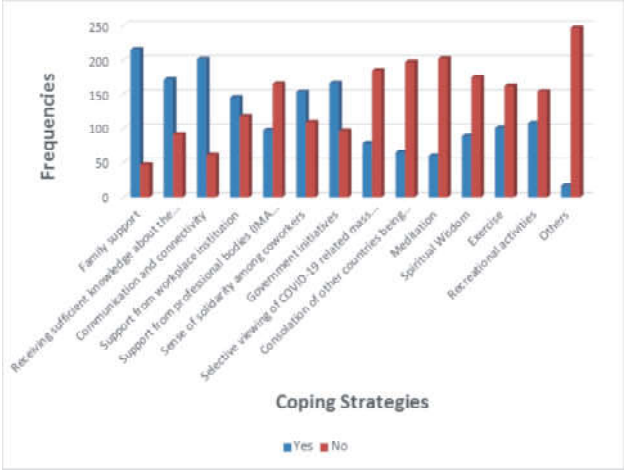


Fig. 3: *Comparison of coping strategies adopted by COVID-19 and Non-COVID-19 healthcare professionals.*

and exercising, etc. The CoD professionals, however, could not employ much of these strategies. Fig. 3 illustrates the perceived coping strategies of all participants.

Discussion

This cross-sectional survey revealed a severe impact on HCPs occupational and psychological well-being during COVID-19 pandemic in India. It revealed that HCPs on CoD had significantly lesser perception of occupational preparedness, due to close encounter with the practical challenges of adhering to protocols for infection control measures[7]. Though N-CoD professionals received similar training for new barrier precautions and hygiene recommendations[23], but being physically distant from the field, their appraisal of various shortcomings like lack of protective gear, and intensive workload, could have been relatively diminished[17]. Males were found to have significantly better occupational preparedness than females. This could be because relatively more males are often found in high-level positions at workplace, making it easier for them to be a part of a quick information loop, and avail resources easily, thus feeling more prepared, as found in previous studies[25]. Also, women have more household and caretaking roles in our society in general, making it more difficult to maintain work-life balance[26]. Professionals working in private settings, perceived better occupational preparedness, since the load to deal with Covid-19 patients has been majorly borne by government institutions[27]. Further, professionals working in Delhi/NCR experienced higher occupational preparedness, which could be because of frequent training measures, greater sense of awareness and improving infrastructure.

HCPs positioned at the COVID care units, experienced much lower work satisfaction. This may be due to exponential increase in workload and numerous challenges thrust upon them, like-fluctuating work shifts, wearing PPE for long hours, high-risk environments, and facing misconducts[29 30]. Furthermore, the absence of well-established management guidelines, inefficient communication, depleted workforce, and working with scarce resources also held implications for their mental well-being and work satisfaction[7]. This was seen more in younger HCPs because they constituted the bulk of frontline workers. Although unfamiliarity of disease was persistent among all HCPs, these younger professionals lacked experience, adequate knowledge, attitude and appropriate preparedness towards the pandemic, as found in corresponding research[31]. Since elderly are more vulnerable to fatal infection of COVID-19, the private institutions in India avoided engaging HCPs of >60 years, unless absolutely necessary, further resulting in their higher work satisfaction[32, 33]. Work satisfaction was also found to be higher among HCPs working in Delhi, since they felt occupationally better prepared at their work environment.

Additionally, Delhi being the capital, the medical associations have been more influential in warranting better safeguarding measures against the stigmatisation, violence and harassment faced by the HCPs[28].

N-CoD professionals struggled more with the challenges of adaptation and protection. In congruence with former studies, the current study revealed that individuals working with COVID patients for some time now, experienced less anxiety, because of having adjusted to their roles and new protocols[6]. Age is another influencing variable primarily for two reasons. Firstly, for the older HCPs, newer challenges like adapting to rapid digitalisation with virtual/telemedicine generated frustration being available for their patients at uncertain hours and longer periods of time posed additional challenges of work life balance and burnout[34,35]. Secondly, the presence of comorbidities with increasing age, and thereby greater risk from COVID, added to the fear of infection and difficulty in adapting to the changed healthcare dynamics[36].

Lastly, the overall well-being of all the HCPs was compromised. Feelings of uncertainty, fear of contagion, psycho-social impact of confinement and the challenge of disrupted healthcare delivery systems were common among all medical staff[17,37,38]. These factors can act as distressing agents, inducing a sense of helplessness and powerlessness[39]. Well-being of CoD professionals was found to be significantly lower in comparison with the N-CoD professionals, in congruence with other researches, highlighting the numerous challenges confronted by them, like, irregular working hours, heightened workload, anxiety, apprehension of unfamiliar clinical roles and critical decisions about patient care[40,41]. Additionally, while on one end persistent exposure to the virus posed the grave risk of infection to self and family, on the other end was the challenge to provide physical and emotional support to one's family[42]. This was more pronounced among the female HCPs, who showed more negative emotions, especially the nursing staff, frequently involved in direct COVID patient care. Similar trends of gendered nature of occupational distress and amplified challenge of work life balance have been observed in former studies[43,13]. These stressors, undoubtedly, exhaust physically, strain emotionally and lead to psychological burdening of frontline workers, severely impacting their well-being. However, the older HCPs showed higher well-being. This could be, because most of the younger professionals were incorporated in COVID-related responsibilities. Also, it is the tendency of older people to perceive hassles as less stressful and wisdom helps them remain emotionally stable in times of distress[44].

Hence, uncertainties like fear of risk of infection, ensuring performativity during confinement and adhering to professional and personal responsibilities were the most prominent stressors for the HCPs, as observed through LDA and thematic analysis. To deal with these stressors various coping strategies were used by HCPs though, more prominently by the N-CoD professionals. The CoD professionals had lesser opportunity to employ many of the coping strategies due to limited flexibility in working hours and lack of adequate mental health intervention at workplaces. Further, majority of participants emphasized the need for improved availability of safety measures, stern measures for implementing safety norms/principles, and keen surveillance and organised planning. Therefore, intensive intervention at the psychological and administrative level is critical for improving well-being of HCPs.

Conclusion and Implications

The present study on HCPs has highlighted the various struggles being faced by them at structural and psycho-social level, especially those placed in CoD. The study further showed that their overall mental health and well-being was negatively impacted. Mental health of these professionals is important for ensuring quality of care, keeping the healthcare system running and for their personal health and quality of life. Hence, services of psychological assistance and counselling, targeted at HCPs should be deployed immediately at workplaces. In addition, coping strategies like "containment" and "buddying-up" can further improve their well-being.

Limitations

The study has certain limitations. First, the sample size is small and taken from different regions of India, without equal distribution. Therefore, representation of the sample is skewed, limiting the generalisation of our findings. Second, the study was carried out during the first two months of the pandemic in India; thereafter, the number of cases has overwhelmingly risen. Consequently the study cannot account for the changing dynamics of HCPs well-being. Lastly, since there was no prior assessment of the mental health of these HCPs, effectively distinguishing between the pre-existing symptoms and newer symptoms of declined well-being is not feasible.

References

1. Chan JF, Yuan S, Kok KH *et al*. A familial cluster of pneumonia associated with the 2019 novel coronavirus indicating person-to-person transmission: a study of a family cluster. Lancet 2020; 395: 514-23.
2. WHO. Virtual press conference on COVID-19 – 11 March 2020. In: Director W, (ed.). 2020.
3. Johncox C. Global COVID-19: Tracking countries with the most

cases, deaths on June 30. 2020, June 30.

4. Unnithan, Kerala PS. Reports first confirmed coronavirus case in India. *India Today* 2020, January 31.
5. India COVID-19 Updates May 17. Maharashtra cases cross 30,000-mark; some states seek lockdown extension. *The Indian Express*, 2020, May 17.
6. Lai J, Ma S, Wang Y *et al*. Factors Associated With Mental Health Outcomes Among Healthcare Workers Exposed to Coronavirus Disease 2019. *JAMA Netw Open* 2020; 3: e203976.
7. Ornell F, Halpern SC, Kessler FHP *et al*. The impact of the COVID-19 pandemic on the mental health of healthcare professionals. *Cad Saude Publica* 2020; 36: e00063520.
8. Sharma N. Stigma: the other enemy India's overworked doctors face in the battle against Covid-19. *Quartz India*, 2020, March 24.
9. Tsamakis K, Rizos E, Manolis AJ *et al*. COVID-19 pandemic and its impact on mental health of healthcare professionals. *Exp Ther Med* 2020; 19: 3451-3.
10. Chong MY, Wang WC, Hsieh WC *et al*. Psychological impact of severe acute respiratory syndrome on health workers in a tertiary hospital. *Br J Psychiatry* 2004; 185: 127-33.
11. Lee AM, Wong JG, McAlonan GM *et al*. Stress and psychological distress among SARS survivors 1 year after the outbreak. *Can J Psychiatry* 2007; 52: 233-40.
12. Goulia P, Mantas C, Dimitroula D *et al*. General hospital staff worries, perceived sufficiency of information and associated psychological distress during the A/H1N1 influenza pandemic. *BMC Infect Dis* 2010; 10: 322.
13. Pearce and L. How COVID-19 is affecting nurses' mental health, and what to do about it. *Nursing Standard* 2020, April 8.
14. Bakotic D, Babic TB. Relationship between Working Conditions and Job Satisfaction: The Case of Croatian Shipbuilding Company. *Inter J Business and Social Science* 2013; 4: 206-13.
15. Sell L and Bryan C. Job Satisfaction, Work Environment, and Rewards: Motivational Theory Revisitedlabr. *Labour* 2011; 25: 1-23.
16. Soni and P. Indian doctors have an unexpected problem while battling Coronavirus. . *Business Insider*, 2020.
17. Liu CY, Yang YZ, Zhang XM *et al*. The prevalence and influencing factors in anxiety in medical workers fighting COVID-19 in China: a cross-sectional survey. *Epidemiol Infect* 2020; 148: e98.
18. Karampelias V, Karonis D, Psaroudi V. The psycho-emotional impact of COVID-19 on surgical staff working in emergency departments. *Eur J Trauma Emerg Surg* 2020, DOI: 0.1007/s00068-020-01411-3.
19. Pfefferbaum B, North CS. Mental Health and the Covid-19 Pandemic. *N Engl J Med* 2020; DOI: 10.1056/NEJMp2008017.
20. Diener E. Subjective well-being. *Psychol Bull* 1984; 95: 542-75.
21. Ryff CD. Happiness is everything, or is it? Explorations on the meaning of psychological well-being. *J Personality and Social Psychology* 1989; 57: 1069-81.
22. IBM Corp. IBM SPSS Statistics for Windows, Version 23.0. Armonk, NY: IBM Corp. , 2015.
23. Modi PD, Nair G, Uppe A *et al*. COVID-19 Awareness Among Healthcare Students and Professionals in Mumbai Metropolitan Region: A Questionnaire-Based Survey. *Cureus* 2020; 12: e7514.
24. Hausmann R, LD T and SZ Gender Gap Report. 2011.
25. Waller N. How Men and Women See the Workplace Differently. *The Wall Street Journal* 2016, September 27.
26. Kumari KT, Devi VR. A Study on Work–Life Balance of Women Employees in Selected Service Sectors. *Pacific Business Review International* 2015; 7: 17-23.
27. India Today. How Corona crisis exposes private healthcare system, calls for an overhaul. *India Today*, 2020, May 8.
28. Aravind I. Covid-19: How Healthcare workers are paying a heavy price in this battle. *Economic Times*, 2020, April 12.
29. Loibner M, Hagauer S, Schwantzer G *et al*. Limiting factors for wearing personal protective equipment (PPE) in a health care environment evaluated in a randomised study. *PLoS One* 2019; 14: e0210775.
30. Sasangohar F, Jones SL, Masud FN *et al*. Provider Burnout and Fatigue During the COVID-19 Pandemic: Lessons Learned From a High-Volume Intensive Care Unit. *Anesth Analg* 2020; 131: 106-11.
31. Agarwal V, Gupta L *et al*. Undergraduate medical students in India are underprepared to be the young-taskforce against Covid-19 amid prevalent fears. *BMJ Yale* 2020. DOI: https://doi.org/10.1101/2020.04.11.20061333.
32. Verity R, Okell LC, Dorigatti I *et al*. Estimates of the severity of coronavirus disease 2019: a model-based analysis. *Lancet Infect Dis* 2020; 20: 669-77.
33. Bhaumik S and Babu RG. Why healthcare workers above 60 should be 'benched'. *The Hindu*, 2020.
34. Bashshur RL, Howell JD, Krupinski EA *et al*. The Empirical Foundations of Telemedicine Interventions in Primary Care. *Telemed J E Health* 2016; 22: 342-75.
35. Greenberg N, Docherty M, Gnanapragasam S *et al*. Managing mental health challenges faced by healthcare workers during covid-19 pandemic. *BMJ* 2020; 368: m1211.
36. Barnett K, Mercer SW, Norbury M *et al*. Epidemiology of multimorbidity and implications for health care, research, and medical education: a cross-sectional study. *Lancet* 2012; 380: 37-43.
37. Shanafelt TD, Boone S, Tan L *et al*. Burnout and satisfaction with work-life balance among US physicians relative to the general US population. *Arch Intern Med* 2012; 172: 1377-85.
38. Varshney M, Parel JT, Raizada N *et al*. Initial psychological impact of COVID-19 and its correlates in Indian Community: An online (FEEL-COVID) survey. *PLoS One* 2020; 15: e0233874.
39. Elovainio M, Virtanen M, Oksanen T. Physicians' working conditions, health and working capacity. *Duodecim* 2017; 133: 647-52.
40. Ayanian JZ. Editor's Comment: Mental Health Needs of Healthcare Workers Providing Frontline COVID-19 Care. *JAMANetwork*, 2020.
41. Shyrock T. COVID-19 Raises Ethical Dilemmas for Many Physicians. 2020.
42. Blake H, Bermingham F, Johnson G *et al*. Mitigating the Psychological Impact of COVID-19 on Healthcare Workers: A Digital Learning Package. *Int J Environ Res Public Health* 2020; 17 DOI: 10.3390/ijerph17092997.
43. Bird. Women and COVID-19: Studying the Impact of Sex and Gender. 2020, April 13.
44. Oaklander M. Old People Are Happier Than People *Time*, 2016, August 24.

ORIGINAL ARTICLE

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Relationship of Age and Viral Load with Clinical and Laboratory Profile in COVID-19 Patients at Presentation

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Abstract

***Background:** The COVID-19 pandemic caused by novel SARS-COV-2 has caused a sudden and substantial increase in hospitalisations. The incidence and severity are higher among elderly patients, and those with co-morbidities. Its severity has been linked with the viral load of infection. We aim to investigate the link of age and viral load with patients' symptoms and their laboratory findings at the time of presentation to hospital.*

***Methods:** Cross-sectional, observational study of randomly selected COVID-19 RT PCR positive indoor patients.*

***Results:** Patients above 50 years of age were more in number than those less than 50 years of age. Shortness of breath, cough (dry/sputum), body ache and fever were the main presenting complaints in both age groups. In patients below 50 years of age, those with a high viral load were reported to have a lower SpO2 (p = 0.07) than patients with a low viral load. In patients above 50 years, those with a high viral load were found to have a lower SpO2 (p = 0.009), higher HRCT scores (CT Severity Score) (p = 0.0002) and higher Neutrophil Lymphocyte Ratio (NLR) (p = 0.09), as compared to patients with a low viral load. In patients with a low viral load, the NLR was found to be higher (p = 0.007) in those above 50 years. Finally, in patients with a high viral load, higher HRCT scores (p = 0.08) were found in those above 50 years.*

***Conclusion:** Our results indicate that a higher age can adversely affect some biomarkers and disease outcome, irrespective of the viral load of the patients. At the same time, we found that a higher viral load can also adversely affect the severity of disease, irrespective of the age of patients.*

***Key words:** SARS CoV-2, COVID-19, cycle threshold, CT severity score, NLR, age.*

Introduction

The world was hit by the first large pandemic, The Spanish flu, in 1918. It was caused by the H_1N_1 influenza A virus, infected 500 million people (about a third of the world's population at that time) and lasted for more than 2 years.

The COVID-19 pandemic (as declared by WHO on March 11, 2020), hit the world in Dec 2019. As of Jan 16, 2021, more than 94.4 million cases and more than 2.02 million deaths have been attributed to COVID-19. The world has seen the waves of other Corona virus infections like SARS CoV-1 and MERS in the past. The SARS-CoV-2 is a novel Corona virus, thought to have originated from either bats or the animal market of Wuhan, China.

The disease caused by SARS-CoV-2 was named COVID-19. The clinical presentation of the disease is highly variable, ranging from asymptomatic to severe illness involving multiple organs. The virus spreads through the air, when two or more people are at a close distance to each other, in the form of droplets through cough, sneeze, breath, or through contact via contaminated surfaces. Therefore, the preventive measures are wearing masks, washing hands properly, and physical distancing. As the virus is a novel virus, the understanding about its transmissibility, clinical presentation, pathogenicity, and treatment gradually evolved and changed with experience.

The main concerns associated with the virus are asymptomatic carriers and vulnerable elderly population with or without comorbidities. As time passed, it was realised that the disease was affecting elderly patients with comorbidities more severely, who were being hospitalised for prolonged durations. The younger population, who were either affected less severely, or remained asymptomatic, probably acted as carriers.

As this virus spreads through air and contact, another question which was raised was, "How the viral load of infection is associated with infectivity and patient outcome?" High viral load was earlier reported to be associated with severe disease and poor outcome.

Aims and objectives

The severity of COVID-19 has been linked with higher age and viral load in patients. We aim to investigate the link of

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age and viral load with patients' symptoms and their laboratory findings at the time of presentation to hospital.

Data source

This study was planned and executed at the Mahatma Gandhi Medical College and Hospital, Jaipur. Of all the COVID-19 RT PCR positive patients who were admitted in COVID-19 ward at the hospital between October 2020 to December 2020, 106 were randomly selected for this study, after the approval and consent of Institutional ethics committee.

Material and Methods

COVID-19 RT PCR was confirmed by RNA amplification by ABI 7,500 Fast Dx Real-time Polymerase Chain Reaction. We also measured cycle threshold or Ct value; i.e., the number of cycles after which fluorescence of the PCR product is detectable over and above the background signal. Theoretically, the Ct value is inversely proportional to the amount of genetic material (RNA) in the starting sample and lower Ct values generally correlate with high viral load.

At presentation, each patient's HRCT chest, baseline investigations, and inflammatory markers were recorded. Along with these, a detailed history was also recorded, and treatment was started as per prevailing protocol of the Ministry of Health and Family Welfare (MOHFW), Government of India.

Data was captured on Google forms and was sent to the investigator. Google sheets were prepared from Google forms and the data was then transferred to Microsoft Excel. Finally, this data was imported into STATA, a statistical analysis software, where it was interpreted and analysed.

Inclusion and exclusion criteria

A patient was included in the study only if:-

- His/her age was more than 18 years
- He/she tested positive for COVID-19 by RT PCR
- Gave informed consent to be a part of the study

Patients who did not meet the inclusion criteria were excluded.

Statistical analysis

All data was analysed using the statistical analysis software, STATA. The analysis started with a calculation of the descriptive statistics of the data: number of patients with co-morbidities, number of patients displaying various symptoms of COVID-19, and a simple calculation of the percentage of patients in various categories (gender, symptoms, co-morbidities and oxygen requirement) according to their age (below or above 50 years of age) and viral load (cycle threshold value of less than and more than 24). The results of this analysis are presented in Figs. 1 and 2, as well as in Table I.

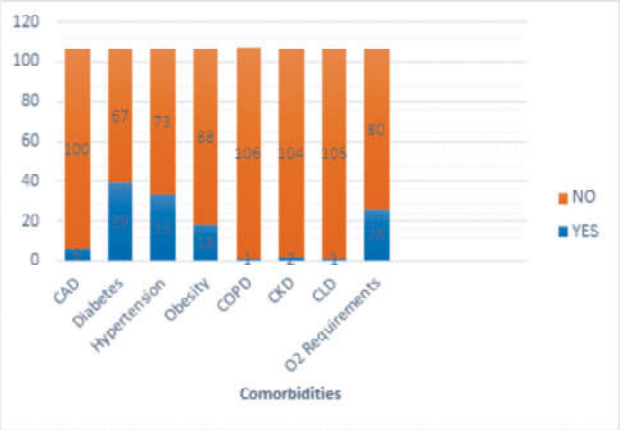
Following this, two analyses were undertaken. In the first, patients were divided into two categories, according to their age, and within each category, were divided according to their viral load. Following this, the mean value of each sub-category was calculated, and independent t-tests for difference in means were undertaken to infer if, given the age category of a patient, his viral load had an effect on the value of his/her investigation variables. The results of this analysis are presented in Table II.

In the second analysis, patients were divided into two categories according to their viral load and then, within each category, were divided according to their age. Following this, the mean value of each sub-category was calculated, and independent t-tests for difference in means were undertaken to infer, if given the viral load of a patient, the age had an effect on the value of his/her investigation variables. The results of this analysis are presented in Table III.

Results

Out of the 106 patients, 29 (27.3%) were females and 77 (72.6%) were males. Patients less than 50 years of age were 30 (6 females, 24 males), and patients more than 50 years of age were 76 (23 female and 53 males).

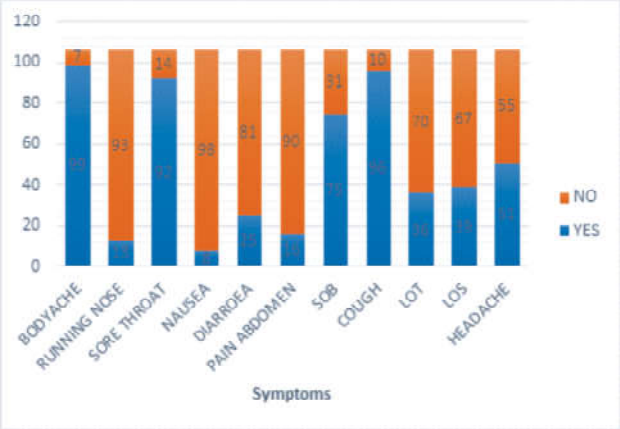
2 patients had coronary artery disease (1.8%), 39 had diabetes mellitus (36.7%), 33 had hypertension (31.1%), 18 had obesity (16.9%), 1 had chronic obstructive pulmonary disease (0.94%), 2 had chronic kidney disease (1.8%), and 1 had chronic liver disease (0.94%) (Fig. 1).



(CAD = Coronary artery disease, COPD = Chronic obstructive pulmonary disease, CKD = Chronic kidney disease, CLD = Chronic liver disease).

Fig. 1: *Number of patients with different comorbidities.*

99 patients presented with bodyache (93.3%), 13 with running nose (12.2%), 92 with sore throat (86.2%), 8 with nausea/vomiting (7.54%), 25 with diarrhoea (23.58%), 16 with pain in abdomen (15%), 75 with shortness of breath (70.7%), 46 with dry cough (43.3%), 50 with cough with sputum (47.1%), 36 with loss of taste (33.9%), 39 with loss of smell (36.7%), 51 with headache (48.1%), and 7 presented with no symptoms (6.6%) (Fig. 2). Oxygen was required in 25 patients (23.5%). 1 patient required ventilation, while 80 (75.47%) did not require oxygen (Fig. 2). We divided symptoms according to age below 50 years and above 50 years and further divided according to low and high viral load in Table I.



(SOB = Shortness of breath, LOT = Loss of taste, LOS = Loss of smell).

Fig. 2: *Number of patients with different symptoms.*

In patients below 50 years of age, those with a high viral load (Cycle threshold value less than 24) were reported to have a lower SpO2 (92.5%) (p = 0.07) than the patients with a low viral load (SpO2 - 94.3%). In patients above 50 years, those with a high viral load (Cycle threshold value less than 24) were found to have a lower SpO2, (91.1% versus 94.4% in the low viral load group; p = 0.009), higher HRCT scores (CTSS) (13.4 versus 9.4 in the low viral load group; p = 0.0002) and higher Neutrophil Lymphocyte Ratio (9.1 versus 5.7 in the low viral load group; p = 0.09) (Table II).

In patients with a low viral load, the Neutrophil Lymphocyte ratio (NLR) was found to be higher in those above 50 years (5.7 versus 3.6; p = 0.007). Finally, in patients with a high viral load, higher HRCT scores were found in those above 50 years (13.4 versus 10.4 out of 25; p = 0.08) (Table III).

We did not find any significant difference in Neutrophil to Monocyte Ratio (NMR), C-Reactive Protein, SGOT, SGPT, D-Dimer, LDH, Ferritin, and Procalcitonin between the two age groups and there was no difference in these markers between high and low viral load groups, too.

Table I: Percentage of patients in various variable categories.

According to their age and Cycle threshold (Ct) value						
Variable	**Age below 50 yrs**		**N = 30**	**Age above 50 yrs**		**N = 76**
	Low viral load (Ct: > 24)	**High viral load (Ct: < 24)**		**Low viral load (Ct: > 24)**	**High viral load (Ct: < 24)**	
Gender			30			76
Female	67%	33%	6	78%	22%	23
Male	63%	37%	24	58%	42%	53
Shortness of breath			30			75
No	100%	0%	7	70%	30%	23
Yes	52%	48%	23	62%	38%	52
Loss of taste			30			76
No	58%	42%	19	63%	37%	51
Yes	73%	27%	11	68%	32%	25
Loss of smell			30			76
No	60%	40%	20	64%	36%	47
Yes	70%	30%	10	66%	34%	29
Cough			28			76
No	67%	33%	3	60%	40%	5
Dry	50%	50%	14	53%	47%	32
Sputum	73%	27%	11	74%	26%	39
Headache			30			76
No	77%	23%	13	74%	26%	42
Yes	53%	47%	17	53%	47%	34
Bodyache			30			76
No	100%	0%	1	50%	50%	6
Yes	62%	38%	29	66%	34%	70
Hypertension			12			46
No	20%	80%	5	33%	67%	15
Yes	57%	43%	7	68%	32%	31
Diabetes			11			49
No	17%	83%	6	33%	67%	15
Yes	60%	40%	5	62%	38%	34
Temperature			30			75
No	83%	17%	6	67%	33%	15
Yes	58%	42%	24	63%	37%	60
Oxygen/ventilator			10			16
Oxygen	37%	63%	8	36%	64%	14
HFNC	0%	100%	1	0%	100%	2
Ventilation	100%	0%	1	-	-	0

(N = Number, Ct = Cycle threshold, HFNC = High flow nasal cannula).

Table II: Mean values of investigations by age and Cycle threshold (Ct) value and associated t-test results.

Variable	Age below 50 yrs N=30			p-value	Age above 50 yrs N=76			p-value
	Low viral load (Ct: >24)	High viral load (Ct: < 24)			Low viral load (Ct: >24)	High viral load (Ct: < 24)		
SpO2	94.3	92.5*	30	0.0674	94.4	91.9***	76	0.0087
HRCT	10.3	10.4	28	0.9308	9.4	13.4****	73	0.0002
NLR	3.6	9.3	29	0.2380	5.7	9.1**	72	0.0861
NMR	79	12.6	3	NA[1]	24.4	27.9	11	0.8139
CRP	33.9	40.8	30	0.7333	47.8	64.1	76	0.2979
SGOT	42.8	53.7	30	0.3187	50.4	43.1	76	0.4104
SGPT	52.9	67.8	30	0.5054	41.1	41.6	76	0.9373
D-Dimer	385.9	557.2	30	0.3280	610	862.1	76	0.4138
LDH	210.1	286.6	30	0.2086	257.9	284.2	76	0.4036
HBA1c	5.8	7	14	0.5463	6.8	6.6	57	0.4547
Ferritin	274	313.8	30	0.6726	303.2	328.2	76	0.7271
PCT	0.108	0.154	16	0.4665	0.207	0.229	46	0.8970

*Notes: 1. *Difference in means (two tailed independent t-test) was significant at 10% significance level; **difference in means was significant at 5% significance level; ***difference in means was significant at 1% significance level; ****difference in means was significant at 5% significance level.*

2. There was only one observation with low viral load so the software was unable to calculate standard deviation.

3. Prior to conducting the t-test for each difference in means, tests of equality of variance were undertaken. Wherever the hypothesis of equal variance was rejected, the t-test was undertaken assuming unequal variance.

(N = Number, Ct = Cycle threshold, HRCT = High resolution computed tomography, SpO2 = Oxygen saturation, NLR = Neutrophil to lymphocyte ratio, NMR = Neutrophil to monocyte ratio, CRP = C reactive protein, SGOT = Serum glutamic oxaloacetic transaminase, SGPT = Serum glutamic pyruvic transaminase, LDH = Lactate dehydrogenase, PCT = Procalcitonin).

Table III: Mean values of investigations by Cycle threshold (Ct) value and age and associated t-test results.

Variable	Age below 50 yrs N = 68			p-value	Age above 50 yrs N = 38			p-value
	Low viral load (Ct: >24)	High viral load (Ct: < 24)			Low viral load (Ct: >24)	High viral load (Ct: < 24)		
SpO2	94.3	94.4	68	0.9376	92.5	91.9	38	0.5367
HRCT	10.3	9.4	68	0.5683	10.4	13.4*	33	0.0800
NLR	3.6	5.7***	66	0.0070	9.3	9.1	35	0.9735
NMR	79	24.4	5	NA[1]	12.6	27.9	9	0.4611
CRP	33.9	47.8	68	0.3835	40.8	64.1	38	0.3346
SGOT	42.8	50.4	68	0.4095	53.7	43.1	38	0.3200
SGPT	52.9	41.1	68	0.1696	67.8	41.6	38	0.2415
D-Dimer	385.9	610	68	0.2581	557.2	862.1	38	0.3356
LDH	210.1	257.9	68	0.1960	286.6	284.2	38	0.9636
HBA1c	5.8	6.8***	49	0.0028	7	6.6	22	0.7945
Ferritin	274	303.2	68	0.7480	313.8	328.2	38	0.8692
PCT	0.108	0.207	37	0.4713	0.154	0.229	25	0.4988

*Notes: 1. * Difference in means (two tailed independent t-test) was significant at 10% significance level; **difference in means was significant at 5% significance level; ***difference in means was significant at 1% significance level; ****difference in means was significant at 5% significance level.*

2. There was only one observation with low viral load so the software was unable to calculate the standard deviation.

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(N = Number, Ct = Cycle threshold, HRCT = High resolution Computed tomography, SpO2 = Oxygen saturation, NLR = Neutrophil to lymphocyte ratio, NMR = Neutrophil to monocyte ratio, CRP = C reactive protein, SGOT = Serum glutamic oxaloacetic transaminase, SGPT = Serum glutamic pyruvic transaminase, LDH = Lactate dehydrogenase, PCT = Procalcitonin).

Discussion

Starting as a new disease, the SARS-CoV-2 outbreak soon engulfed the world and turned into a pandemic. As this was a novel virus with little understanding about how it presents and spreads, it posed many challenges, including difficulty in early recognition of infection and the difficulty in diagnosing patients who were asymptomatic. Gradually, an understanding of pathogenesis, investigations, and treatment protocol developed, along with some repurposed drugs.

COVID-19 was deemed to be more severe with a higher risk mortality in elderly patients and those with co-morbidities, compared to younger patients and those without co-morbidities. On the other hand, children and younger patients have been less severely affected by the disease. Men have been found to be at higher risk of mortality due to COVID-19[4] and different age groups were found to have different outcomes.

In this study, we compared patients' symptoms, severity and laboratory investigations according to their age group

(below and above 50 years of age) and according to their viral load (cycle threshold value of less than and more than 24). It was found that patients above 50 years of age were more in number in the randomly selected sample. Main presenting complaints in both the age groups were shortness of breath, cough (dry/sputum), body ache, and fever.

In patients above 50, a high viral load was found to be associated with a lower SpO2, higher HRCT scores (CT Severity Score) and higher Neutrophil Lymphocyte Ratio (NLR). Additionally, in patients with low viral load, those above 50 years were reported to have a higher NLR. These two observations show that patients above 50 years had higher biomarkers and an advanced disease, irrespective of their viral load.

In a study, done in 16 countries, persons age 65 years or older had significantly higher COVID-19 mortality rates compared to younger individuals; men had a higher risk of COVID-19 death than women[1].

Pence *et al* found age related poor outcome and related it to pathological monocyte responses in COVID-19 as monocytes and pulmonary monocyte derived macrophages play an early and key role in the progression to severe COVID-19 by promoting cytokine storm, ARDS and other tissue damage[2].

Our results are corroborated by the hypothesis advanced by Chen *et al* that age-related decline and dysregulation of immunological functions, immunosenescence and inflammation play a major role to increase vulnerability to severe COVID-19 infections and poor outcome[3]. Our results are also in line with Moreno *et al*, who discussed the role of damaged/aged mitochondria responsible for oxy-inflammaging, immunosenescence, chronic inflammation, deficient antiviral response mainly responsible for severe disease in the aged population[4].

While patients with a high viral load had higher HRCT scores in above 50 years age group, even patients who were below 50 years were found to have lower SpO2 if they had a high viral load. These observations show that patients with a higher viral load are likely to have advanced disease, irrespective of their age group.

Our results are supported by the findings of Rao *et al*, who undertook a systematic review of the clinical utility of cycle threshold value in the context of COVID-19 and found that lower cycle threshold values are potentially associated with worse outcomes, severe disease and increased morbidity and morality[5]. However, Singanayagam *et al* showed no significant difference in cycle threshold values or culture positivity for upper respiratory tract samples across different age groups[6.] Karahasan *et al* showed that viral load was not a critical factor for hospitalisation and mortality – outdoor patients also had considerable viral load in their nasopharynx and were contagious[7]. Yang *et al* concluded that elevated age and NLR can be considered independent biomarkers of poor clinical outcomes[8]. Liu *et al* concluded that NLR is an independent risk factor of the in-hospital mortality for COVID-19 patients, specially for males[9].

There was no significant difference in other markers like, Neutrophil to Monocyte Ratio (NMR), C-Reactive Protein, SGOT, SGPT, D-Dimer, LDH, Ferritin, Procalcitonin between the two age groups, and high and low viral load groups.

Limitations

Our study was undertaken at a single centre and the patients and doctors were not blinded. A large multi-centre study with a higher number of subjects may be helpful in determining the role of age, gender, viral load and different biomarkers in the severity and outcome of COVID-19 disease.

Conclusion

Our results indicate that a higher age can adversely affect serum biomarkers and disease outcome, irrespective of the viral load of COVID-19 patients. At the same time, we have found that a higher viral load can also adversely affect the severity of disease, irrespective of the age of patients.

While our results are corroborated by and support many existing findings in the literature, a multi-centre study with a higher number of patients would be able to shed more light on the relationships we have sought to explore by the means of this study.

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References

1. Yanez ND, Weiss NS, Romand JA *et al.* COVID-19 mortality risk for older men and women. *BMC Public Health* 2020; 20: 1742.
2. Pence BD. Severe COVID-19 and aging: are monocytes the key? *Geroscience* 2020; 42 (4): 1051-61.
3. Chen Y, Klein SL, Garibaldi BT *et al*. Aging in COVID-19: Vulnerability, immunity and intervention. *Ageing Res Rev* 2021; 65: 101205.
4. Moreno Fernández-Ayala DJ, Navas P, López-Lluch G. Age-related mitochondrial dysfunction as a key factor in COVID-19 disease. *Exp Gerontol* 2020; 142: 111147.
5. Rao SN, Manissero D, Steele VR *et al*. A Systematic Review of the Clinical Utility of Cycle Threshold Values in the Context of COVID-19. *Infect Dis Ther* 2020; 9 (3): 573-86.

6. Singanayagam A, Patel M, Charlett A *et al*. Duration of infectiousness and correlation with RT-PCR cycle threshold values in cases of COVID-19, England, January to May 2020. *Euro Surveill* 2020; 25 (32): 2001483.
7. Karahasan Yagci A, Sarinoglu RC, Bilgin H *et al*. Relationship of the cycle threshold values of SARS-CoV-2 polymerase chain reaction and total severity score of computerised tomography in patients with COVID 19. *Int J Infect Dis* 2020; 101: 160-66.
8. Yang AP, Liu JP, Tao WQ *et al*. The diagnostic and predictive role of NLR, d-NLR and PLR in COVID-19 patients. *Int Immunopharmacol* 2020; 84: 106504.
9. Liu Y, Du X, Chen J *et al*. Neutrophil-to-lymphocyte ratio as an independent risk factor for mortality in hospitalised patients with COVID-19. *J Infect* 2020; 81 (1): e6-e12.

REVIEW ARTICLE

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Diagnosis and Management of Invasive Fungal Infections in Critical Care Setting

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Introduction

The incidence of invasive fungal infections is rising worldwide due to an increase in the numbers of susceptible individuals, increase in the usage of broad-spectrum antimicrobials, immunosuppressive therapies and central vascular devices. Improvement in diagnostics as well as therapeutics has led to improved survival of patients with neoplasms, transplant recipients, HIV/AIDS, in post-trauma and age extremes. The challenge is being dealt with the advent of newer antifungals with lesser toxicity and broader activity spectrum. Also, better diagnostic strategies such as improved radiological imaging and rapid serology tests have provided the caregivers with better tools to detect invasive fungal infections earlier, thus improving outcomes. Newer molecular techniques have been devised which can facilitate specific identification of fungal species thus aiding rapid diagnosis. Despite these improvements, results of therapy remain abysmal and resistance levels to existing antifungal agents are on the rise. The mortality rates related to invasive mycoses have been estimated to be 50% to 60% among ICU patients and increases to 75% to 90% among patients with shock. Invasive fungal infections lead to significant stress on healthcare facilities due to prolonged duration of hospitalisation, use of expensive antifungals and increased utilisation of healthcare resources. Recognition of the interaction between the fungal pathogens and host factors is still a major element in the diagnosis and management of fungal infections. Some fungal diseases have typical presentations but many of these occur so infrequently that clinicians may not initially consider them in their differential diagnoses. In the presence of immunocompromised state, invasive fungal infections may manifest unusual signs and symptoms, making their diagnosis a big challenge. Early diagnosis and prompt therapy is the basis to improving disease outcomes in life-threatening invasive mycoses, especially among immunosuppressed patients. Knowledge of the important risk factors and various clinical manifestations of invasive fungal infections may enable both internists and intensivists to develop an inclusive approach towards timely diagnosis of these infections and guiding appropriate therapeutic response.

Epidemiology

Candida species

Candida spp. is the usual flora associated with mucosal surfaces in humans. In the event of mucosal barrier breakdown or immunosuppression, these organisms may become clinically significant pathogens and can cause fulminant infections leading to increased morbidity and mortality. Candidiasis ranges from infections involving mucosal surfaces to more widespread disease (e.g., pyelonephritis, meningitis/encephalitis, ocular, pneumonitis, endocarditis, intra-abdominal infections/abscesses, sepsis). Invasive candidiasis is a deep-seated mycosis among critically ill patients and is associated with a mortality rate exceeding 50 - 60% in some ICU settings[1]. An estimation of the exact prevalence of these infections is difficult because of variations in study methodologies, number of healthcare institutes involved and type of patients included. However, some studies cite a prevalence of approximately 7 cases per 1,000 ICU patients. The definitive diagnosis of invasive candidiasis is established when candida is isolated in tissue specimens from normally sterile body sites or if cultured from a normally sterile body fluid. Among critically ill individuals, invasive diagnostic methods are often not feasible, and delays in awaiting culture results can deny patients timely institution of specific antifungal therapy. Delay in specific therapy is commonly associated with adverse clinical outcomes in invasive candidiasis[2].

Candida albicans is the most commonly isolated species but accounts for only about 50% of the *Candida spp.* isolated among both ward and ICU patients. Incidence of non-*albicans* species has recently increased with *C. glabrata* being the second most common species isolated. *C. parapsilosis* is commonly seen in patients with chronic indwelling vascular catheters for delivering total parenteral nutrition. The infection rates of *C. tropicalis*, *C. krusei*, and *C. lusitaniae* have somewhat stabilised but are still considered

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as important pathogens. This epidemiological shift has major therapeutic implications as non-*albicans* species often carry either reduced susceptibility or absolute resistance to fluconazole[3]. *C. auris* has been identified in various regions worldwide as a major cause of drug resistant candidaemia in the ICUs. The haploid genome of *C. auris* is approximately 12.5 Mb with a guanine-cytosine content of almost 45%[4,5]. Genomic studies suggest that there are almost 6,500 to 8,500 nucleotide sequences, with a number of these nucleotides coding for proteins considered as virulence factors in other candida species, such as biofilm formation. Many transporter genes as well as protein kinases, contributing to acquisition of drug resistance, have also been identified[5].

Mould Pathogens

Invasive mould infections caused by *Aspergillus spp.* are not uncommon among critically ill patients. Invasive aspergillosis (IA) was considered to be a disease seen mainly in neutropenic patients and hematopoietic stem cell transplant recipients. It's now understood that IA is also an important pathogen in critically sick patients without neutropenia, such as those receiving immunosuppressant therapy and those suffering from any chronic organ system failure[6].

Aspergillus spp. are commonly implicated in sinonasal diseases or chronic pulmonary diseases and also in the dermatologic or CNS infections. Outbreaks of *Aspergillus* have been usually associated with faulty air filtration, construction work material and even infected medical equipment as the infection usually starts from inhalation of the conidia. Diagnosis of IA is particularly challenging in critical patients because typical radiological features (halo- or air crescent-sign) are not usually seen in non-neutropenic individuals. Such patients do not progress rapidly to angioinvasive disease frequently. IA is associated with significantly higher mortality to the extent of 65% to 85%. Such high mortality figures are not usually driven by severity of underlying disease as study findings depict mortality rates to be similar between individuals with intact immune responses and hematopoietic stem cell transplant recipients with IA[7].

The moulds causing disease among immunocompromised patients are *Cryptococcus spp.*, *Fusarium spp.*, *Scedosporium spp.*, and *Mucormycoses spp*. These infections are relatively less common in ICU settings but are seen more frequently among individuals on long-term immunotherapy for various rheumatological and other chronic diseases[8].

Invasive aspergillosis and mucormycosis have been more frequently seen recently among patients who were infected with SARS-CoV-2 virus and treated with high dose steroids or were diabetics.

High-risk individuals

It is well known that invasive mycoses are not restricted to individuals with immunocompromised health status. Critically sick ICU patients have monocytes and macrophages with impaired activity along with dysfunctional neutrophils that put them at higher risk of such opportunistic microorganisms.

Various risk factors for invasive fungal infections in the ICU setting

Broad-Spectrum Antimicrobial Therapy
Candida spp. colonisation
Indwelling Central Vascular or Urinary Tract Catheters
Steroid use
Uncontrolled Diabetes Mellitus
Hematopoietic Stem Cell Transplant
Solid Organ Transplant
Graft-versus-Host Disease
Immunosuppressive chemotherapy
Necrotising pancreatitis
Structural pulmonary disease
Hepatic failure
Renal failure
Haemodialysis
Major surgery (esp. abdominal)
Malignancy
Major burns injury
Mucosal damage
Neutropenia
Higher illness severity (APACHE II > 20)
Prolonged mechanical ventilation
Prolonged duration of ICU stay
Total parenteral nutrition

The presence of the aforementioned risk factors among ICU patients makes the clinical decision of when to use antifungal therapy pre-emptively more difficult. Various clinical decision tools and risk prediction models which were developed, have not been adequately tested in prospective multi-center trials. These algorithms have inadequate diagnostic applicability due to their poor positive predictive values and tendency towards over-prescription of antifungals.

There is a strong correlation between colonisation of candida and it's disease manifestation. Rate of colonisation increases linearly with the presence of various risk factors as enumerated already. Most patients who develop invasive mycoses are already colonised to some extent, but only

about 10% to 35% of the colonised individuals develop clinical manifestations. The Candida Colonisation Index has been formulated in surgical ICU patients to evaluate the risk of developing IC in colonised subjects. Ratio of the number of colonised anatomical sites to the number of cultured sites, if greater than 0.5 is associated with an increased risk of invasive candidiasis. Utilising this threshold to initiate empiric antifungal therapy substantially decreases the incidence of infection as compared with historical controls. The shortcomings with the usage of this index are its poor positive predictive value (8 - 10%) and the greater use of antifungal agents[9]. Some of the other clinical prediction tools incorporating several of the risk factors into a scoring system have been assessed for their capacity to predict invasive candidiasis. Their positive and negative predictive values have been depicted in Table I.

Advances in diagnosis of invasive fungal infections

Limitations of fungal culture and radiographic methods

Clinical symptomatology/signs, radiological studies, tissue/blood cultures and histopathology are the usual methods for diagnosing invasive fungal infections. These approaches have many shortcomings and cause much delays in the initiation of appropriate therapy. Invasive mycoses often have a protracted clinical course with nonspecific clinical features. Typical radiological signs (halo sign or macronodules) are not always visualised, particularly in immunocompromised individuals, and thus may not be detectable earlier on. These radiological features are too non-specific, resulting in

Table I: Clinical prediction scores for invasive candidiasis.

Score (years)	Patient population	Model risk factor	Cut-off value	Sensitivity/specificity (%)	PPV (%)	NPV (%)
Dupont score (1994)	Surgical ICU peritonitis	Female upper GI tract origin of peritonitis, perioperative cardiovascular failure, antimicrobial therapy at least 48 hours before penritonitis onset	Grade C= at least three risk factors	84/50	67	72
Candida score (2006)	Medical/surgical ICUs for ≥ 7 days	Severe sepsis (2 points), major surgery (1 point), total parenteral nutrition (1 point), multi-focal *candida* colonisation (1 point)	Score ≥ 3	81/74	16	98
Ostrosky rule (2007, 2011)	Medical/surgical ICUs for ≥ 4 days	Major criteria: systemic antibiotic use days 1 - 3, central venous catheter	Major factors	89/38	4	99
			Two major + at factor	66/69	6	98
		Minor criteria surgery, immunosuppressants, corticosteroids, pancreatitis, dailysis, total parenteral mulrition	One major + at least two minor factors	34/90	10	97
		Modified to add mechanical ventilation for at least 48 hours as an additional major criteria	Three major factors + at least one minor factor	50/83	10	97
Nebraska medical Center rule (2011)	Medical/Surgical ICUs for ≥ 4 days	Broad spectrum antibiotics (1.5 points), central venous catheter (0.9 points), and total parenteral nutrition days 1 - 3 (0.9 points), steroid use in the 7 days before ICU admission up to day 3 (0.4 points), adbominal surgery (0.9 points), and pre-ICU length of stay x 0.0039	Score ≥ 2.45	84.1/60.2	4.7	99.4
Candidaemia rule (2015)	All hospitalised patients with culture positive severe sepsis or septic shock	Antibiotics with 30 days, central venous catheter, admitted from nursing home, or total parenteral nutrition (2 points each), transferred from outside hospital or receving mechanical ventilation (1 point each), lung as presumed source of sepsis (subtract 6 points)	Score ≥ 3	87.6/55.9	18.5	97.5

NPV = Negative predicative value, PPV = Positive predictive value.

Information from: Dupont H. Can yeast isolation in peritoneal fluid be predicted in intensive care unit patients with peritonitis ? *Crit Care Med* 2003, 31: 752-6. Leon C. A bedside scoring system ("*Candida score*") for every antifungal treatment in non-neutropenic critically ill patients with *Candida* colonisation. *Crit Care Med* 2006; 34: 730-7. Ostrosky-Zeichner L. Multicenter retrospective development and validation of a clinical prediction rule for nosocomial invasive candidiasis in the intensive care setting. *Eur J Clin Microbiol Infect Dis* 2007; 26: 271-6. Ostrosky-Zeichner L. Improvement of a clinical prediction rule for clinical trials on prophylaxis fof invasive candidiasis in the intensive care unit. *Mycoses* 2011; 54: 46-51. Hermsen ED, Zapapas MK, Maiefski M *et al*. Validation and comparison of clinical prediction rules for invasive candidiasis in intensive care unit patients: a matched case-control study. *Crit Care* 2011, 15: R198; and Vasques Gullamet C, Vazquez R, Micek ST *et al*. Development and validation of a clinical prediction rule for candidiaemia in hospitalised patients with severe sepsis and septic shock. *J Crit Care* 2015; 30: 715-20.

inappropriate clinical decisions. Fungal blood culture is the gold standard for the diagnosis of invasive candidiasis but has only 50% sensitivity for candida detection and rarely grows any moulds. Blood cultures do not detect deep seated infections and have significant time lag till results are obtained.

Rapid diagnostic tests

Rapid diagnostic tests may aid in the diagnosis of invasive fungal infections before the signs of infection develop. These modalities have reasonable sensitivity and specificity over usual methods and can be utilised in combination with the various risk assessment models to help guide empirical antifungal therapy to target a particular organism. These tests are explained in detail in Table II.

The β-D-glucan diagnostic test is an assay detecting activation of the coagulation cascade by β-D-glucan. It has a good negative predictive value of about 80%, thus making it a valuable tool to avoid inappropriate antifungal use[10].

The positive predictive value of this test is reported to be 30%, when a cut-off of two consecutive tests greater than 80 pg/ml was used[11]. The recommended cut-off value in a single test result is greater than 80 pg/ml and in two consecutive test results, greater than 60 pg/ml, if serial monitoring is done. Values greater than 150 pg/ml for a single test and greater than 80 pg/ml for two consecutive testing have been suggested for critically ill patients. Two consecutive results (twice within a week) above this value are recommended to enhance the diagnostic accuracy of the test.

Mannan is specific to *Candida spp.* and is a polysaccharide component of the fungal cell wall. Latex agglutination and enzyme immunoassay methods exist for both mannan antigen (Mn) and anti-mannan antibodies (Anti-Mn). These tests are more specific than the β-D-glucan test, but not as sensitive and do not become positive until later in the course of the disease. It is seen that the sensitivity of these tests vary based on the *Candida spp.*, with the highest sensitivity reported for *C. albicans* and the lowest for *C. parapsilosis* and *C. krusei*[12].

Galactomannan is a specific assay for *Aspergillus*. The positive predictive value of this assay is relatively weak in non-neutropenic ICU patients and solid organ transplant recipients. The optimal cut-off value is 0.5 depending upon test optical density. Non-neutropenic patients may show false negative test result because of the slow progression to angioinvasive disease[6,7,13]. False-positive results usually occur when administering β-lactams (piperacillin/tazobactam) or Plasma-Lyte[14]. The test can be performed from bronchoalveolar lavage specimens, which tends to increase both the sensitivity and specificity over serum values.

Detection of fungal nucleic acids by polymerase chain reaction is another method to diagnose invasive mycoses. The test allows for the rapid diagnosis of candidaemia and is better than fungal culture in isolating nonviable organisms. It is reported to have a high sensitivity (96%) and specificity (97%) among ICU patients[15].

A major shortcoming of fungal cultures is the long time lag

Table II: Rapid diagnostic tests for invasive fungal infections.

Test	Application	Sensitivity %	Specificity %	Limitations
β-D-glucan	*Candida* spp. and Aspergillus	57 - 97	56 - 93	False-positive: glucan-contaminated tubes/gauze, cellulose-containing dialysis membranes/filters, contaminated albumin/IVIG with fungal elements, gram-positive infections, gut inflammation, some antibiotics (amoxicillin-clavulanic acid) Controversy surrounding optimal cut-off value
Mannan antigen/ Anti-Mannan antibody	*Candida* spp. only	Mannan: 58 Anti-Mannan: 59 Combination: 83	Mannan: 93 Anti-Mannan: 83 Combination: 86	Positive results occur later in disease course Sensitivity varies depending on species Best results when used together Cut-off value unclear
Nucleic-acid PCR	All Species, but only available currently for *Candida* spp.	96	97	Using test too early may decrease sensitivity Unavailable for many organisms
Galactomannan	Aspergillus and some other molds	Serum: 71 BAL: 76 - 88	Serum: 89 BAL: 87 - 100	False-positive: b-lactams, Plasma-Lyte Not as sensitive in non-neutropenic patients

IVIG = Intravenous immunoglobulin, PCR = Polymerase chain reaction.

Information from: Leon C. What's new in the clinical and diagnostic management of invasive candidiasis in critically ill patients. Int Care Med 2014; 40: 808-19; and Perfect JR. Fungal diagnosis. how do we do it and can we do better ? *Curr Med Res Opin* 2013; 29: 3-11.

to positivity. Post-isolation, it takes many days for speciation and susceptibility testing. Molecular-based identification methods like peptic nucleic acid fluorescence in situ hybridisation (PNA-FISH) differentiate between common candida species within a few hours only[16]. Matrix-assisted laser desorption/ionisation time of flight (MALDI-TOF) detects candida directly from whole blood specimens, aiding in rapid diagnostics[17].

Antifungal susceptibility testing

Antifungal susceptibility testing (AST) is of utmost importance in defining resistance patterns and in aiding appropriate drug selection and timely de-escalation of antifungal therapy. Clinical breakpoints for *Candida spp.* and selected azoles are described as susceptible, susceptible-dose dependent, and resistant (Table III). Suggested breakpoints are based on pharmacokinetic-pharmacodynamic (PK-PD) relationships and show close correlation with disease outcomes.

Table III: Antifungal susceptibility breakpoints for *Candida* spp.

Antifungal agent	Secies	Susceptible (mcg/ml)	Susceptible-dose dependent (mcg/dl)	Resistant (mcg/ml)
Fluconazole	*C. albicans* *C.parapsilosis* *C. tropicalis*	≤ 2	4	≥ 8
	C. glabrata	n/a	≤ 32	≥ 64
	C. krusei	n/a	n/a	n/a
Posaconazole	All *candida* spp.	n/a	n/a	n/a
Voriconazole	*C. albicans* *C. parapsilosis* *C. tropicalis*	≤ 0.12	0.25 - 0.5	≥ 1
	C. glabrata	n/a	n/a	n/a
	C. krusei	≤ 0.5	1	≥ 2
Antifungal Agent	**Species**	**Susceptible (mcg/ml)**	**Intermediate (mcg/ml)**	**Resistant (mcg/ml)**
Anidulafungin	*C. albicans* *C. tropicalis* *C. krusei*	≤ 0.25	0.5	≥ 1
	C. parapsilosis *C. guilliermondii*	≤ 2	4	≥ 8
	C. glabrata	≤ 0.12	0.25	≥ 0.5
Caspofungin	*C. albicans* *C. tropicalis* *C. krusei*	≤ 0.25	0.5	≥ 1
	C. parapsilosis *C. guilliermondii*	≤ 2	4	≥ 8
	C. globrata	≤ 0.12	0.25	≥ 0.5
Micafungin	*C. albicans* *C. tropicalis* *C. krusei*	≤ 0.25	0.5	≥ 1
	C. parapsilosis *C. guilliermondil*	≤ 2	4	≥ 8
	C. glabrata	≤ 0.06	0.12	≥ 0.25

n/a = not applicable. Information from: Clinical and laboratory standards institute M27-S4.

A definitive dose: MIC relationship for azole therapy has not been established from research data till date. Absence of correlation studies makes it impossible to evaluate appropriate therapeutic options for drugs with susceptible-dose dependent activity, which require higher than usual doses. Susceptibility testing and clinical outcome has not been established for voriconazole to *C. glabrata* and posaconazole to any *Candida spp*. Clinical breakpoints do not exist for *C. krusei* to fluconazole because of intrinsic resistance. The newer breakpoints are now described as susceptible, intermediate and resistant (Table III). These drug breakpoints have been derived primarily from trials in non-neutropenic patients[18].

Antifungal resistance

Detection of resistance between identifiable species is now possible with the availability and increased use of antifungal susceptibility testing. The resistance rates for most fungal species are increasing gradually. Drug resistance in treatment-naive patients is even more disconcerting. This change is a result of selective pressure from increased antifungals usage in the prophylaxis of immunocompromised individuals; increased pre-emptive and empiric use, particularly in ICU patients because of poor diagnostics; inappropriate use of antifungals in the community for treating even minor fungal infections and rampant use of agricultural fungicides.

C. albicans is only rarely resistant to fluconazole therapy (less than 5% of isolates). Antifungal resistance to other *Candida spp.* is rising, with prevalence rates around 10% for several species. Intrinsic drug resistance of some *Candida spp.* (e.g., *C. krusei*, *C. auris*) to fluconazole is well recognised. Approximately 25% to 30% of candidaemia cases involve intrinsically resistant species, and prior use of antifungals is the most common risk factor for selecting these pathogens.

Resistance acquired during treatment is more difficult to predict and remains to be well defined. Acquired resistance has been studied during treatment of *C. glabrata*, particularly with fluconazole. These species are often cross-resistant to other azoles and may even display multi-drug resistant phenotypes. Acquired resistance to echinocandins is also described in individuals receiving antifungals for longer durations[19].

Mechanisms of drug resistance seen in various fungal microbes include genetic mutations, induction of efflux pumps and increased expression of genes encoding for these mechanisms. Biofilms are a major cause of

resistance in *Candida spp.* because of poor penetration of azoles into these complex cellular matrices. *Aspergillus* forms biofilms in pulmonary parenchyma as well as cavities that contribute to the difficulty in eradicating these infections. The usual resistance mechanisms of each of the drug classes are described in the following Table IV[20].

increases the incidence of drug resistance and leads to fluconazole-resistant species selection, resulting in breakthrough drug resistant colonisation and clinical infections. Prophylactic therapy should not be substituted for proper infection control practices, particularly with indwelling vascular as well as urinary catheters.

A retrospective study was conducted in a surgical ICU in France to assess colonisation trends over an 8-year duration and found a substantial increase in the acquired *C. glabrata* colonisation and a decline in *C. parapsilosis* colony clearance in a cohort where 13% of the subjects received prophylactic fluconazole therapy for significant candidal colonisation[21]. The IDSA guidelines (2016) recommend instituting prophylactic therapy with fluconazole only in those individuals who have a 10% or higher risk of infection on risk prediction score assessment. Prophylactic antifungal therapy has been shown to decrease the incidence of intra-abdominal candidiasis in a particular high-risk group including those who are undergoing intra-abdominal surgery with recurrent anastomotic leaks.

Table IV: Common antifungal resistance mechanisms.

Drug class	Site of action	Resistance mechanism	Implications
Azoles	Inhibit lanosterol-14a-demethylase ERG11 Candida CYP51 Aspergillus	Up-regulation of efflux pump ABC transporters/CDR1, CDR2 genes TAC1 transcription factors Up-regulation of efflux pump MFS transporters/MDR1 gene MRR1 transcription factors ERG11 and CYP51 mutations ERG11 and CYP51 overexpression ERG3 inactivation Biofilm formation Increase in cell wall chitin content	Decrease drug entry into cell (all azoles) Decrease drug entry into cell (fluconazole) Decrease binding affinity, increase MIC Counteract drug effects Ergosterol replaced by another sterol (cross-resistance all azoles) Inhibit drug pentration Increase tolerance to drug
Echinocandins	Inhibit Fksp catalytic subunit of (1,3)-β-D-glucan synthase	FKS1 and FKS2 mutation Increase in cell wall chitin content	Alter catalytic capacity, increase MIC (cross-resistance to entire class) Increase tolerance to drug, paradoxical growth May correlate better with response to therapy than actual MIC
Polyenes	Bind ergosterol Induce oxidative stress	ERG2, ERG3, ERG5, ERG6, ERG1 mutations Increase in anti-oxidative enzymes Alteration in production of free radicals	Decrease ergosterol biosynthesis Decrease oxidative stress

ABC = ATP-binding cassetle; MFS = Major facilitator superfamily..

Information from Spampinato *C. Candida* infections, causes, targets, and resistance mechanisms. Traditional and alternative antifungal agents. *Biomed Res Int* 2013; 204237; Cuenca-Estrella M. Antifungal drug resistance mechanisms in pathogenic fungi: from bench to bedside. *Clin Microbiol Infect* 2014; 20 (Suppl 6): 54-9; and Maubon D. Resistance of *Candida* spp. to antifungal drugs in the ICU: where are we now ? *Int Care Med* 2014; 40: 1241-55.

Approach to invasive candidiasis treatment

Prophylactic therapy

Guidelines from Infectious Disease Society of America (IDSA) for the management of invasive candidiasis support a prophylactic approach to prevent disease in high-risk individuals. Many single-centre studies have suggested that use of prophylactic fluconazole therapy in ICU patients reduces the occurence of invasive candidal infections by about 40 - 50%, however, the approach had doubtful mortality benefit due to inconsistent results and the wide variations in the study cohort. Initiating prophylactic therapy with fluconazole for a large number of ICU patients

Empirical antifungal therapy

Empirical antifungal therapy must be considered in critical ICU patients with risk factors for invasive mycoses and unidentified cause of pyrexia. The decision should be based on evaluation of risk factors, surrogate clinical markers for fungaemia and culture isolates from nonsterile anatomic sites *(strong recommendation; moderate-quality evidence)*. Empirical therapy must be initiated at the earliest in patients with risk factors and having features of septic shock *(strong recommendation; moderate-quality evidence)*. Preferable empirical therapy for suspected candidiasis in non-neutropenic critical patients in ICU is an echinocandin (caspofungin: loading dose of 70 mg, then 50 mg daily; micafungin: 100 mg daily; anidulafungin: loading dose of 200 mg, then 100 mg daily) *(strong recommendation; moderate-quality evidence)*. An acceptable alternative for patients with no prior exposure to azoles and not colonised with azole-resistant Candida species is fluconazole in 12 mg/kg loading dose, followed by 6 mg/kg maintenance dose *(strong recommendation; moderate-quality evidence)*. Lipid formulation Amphotericin B is an option if there is intolerance to the preferred antifungal agents *(strong recommendation; low-quality evidence)*. Recommended duration of empirical therapy for suspected invasive candidiasis in patients who show clinical improvement is 2 weeks, similar to documented candidaemia *(weak recommendation; low-quality evidence)*. For patients who show no improvement to empirical therapy after 4 - 5 days and with no documented evidence of invasive candidiasis after starting empirical therapy, should be considered for discontinuation of antifungal therapy *(strong recommendation; low-quality evidence)*[22].

Pre-emptive antifungal therapy

Screening of high-risk patients before or as soon as the symptoms appear by using diagnostic markers is the basis of this strategy. Screening limits the unwarranted exposure to antifungals but detects patients earlier in the disease course. Similar to prophylactic therapy, the difficulty lies in selecting target patients. In the INTENSE study[23] micafungin was compared to placebo for pre-emptive therapy in high-risk surgical patients with intra-abdominal source of infections. It failed to depict any variation in the incidence of invasive mycoses. Also, there was no difference in the mortality rates or any improvement in organ dysfunction. A study assessing the incidence of resistant *Candida spp.* in subjects with intra-abdominal candidiasis with recent exposure to echinocandins, found the abdomen to be a reservoir of resistant *Candida spp.*[24]. This study found FKS mutant *Candida spp.* in around 25% of individuals with an overall echinocandin failure rate of almost 50%, which explains the lack of utility with micafungin in the INTENSE study.

An approach for empiric/pre-emptive antifungal therapy in suspected invasive candidiasis is outlined below in Fig 1.

Therapeutic strategies for patients with invasive fungal infections

Candida infections

Treatment for candidaemia in non-neutropenic patients[22] recommendations:

1. An echinocandin (caspofungin: 70 mg loading dose followed by 50 mg daily maintenance; micafungin: 100 mg per day; anidulafungin: 200 mg loading dose followed by 100 mg daily maintenance) should be initiated as starting therapy *(strong recommendation; high-quality evidence)*.
2. Fluconazole, 12 mg/kg loading dose followed by 6 mg/kg daily maintenance, is an alternative to echinocandins as starting therapy in patients who are not critically ill and who are not likely to have fluconazole-resistant Candida isolates *(strong recommendation; high-quality evidence)*.
3. Azole sensitivity testing is recommended for all bloodborne and clinically important Candida isolates. Echinocandin sensitivity testing should be done in all patients who have received prior therapy with echinocandins and among those who have been infected with *C. glabrata* or *C. parapsilosis (strong recommendation; low-quality evidence)*.
4. Switchover from echinocandins to fluconazole (mostly within 5 to 7 days) is recommended for patients who have been stable and have specimen isolates sensitive to fluconazole (e.g., *C. albicans*), or are sterile on repeat cultures following administration of antifungal therapy *(strong recommendation; moderate-quality evidence)*.
5. In infections due to *C. glabrata*, switchover to high-dose fluconazole, 12 mg/kg daily; or voriconazole, 3 - 4 mg/kg twice daily; should be considered in patients with fluconazole-sensitive or voriconazole-sensitive isolates *(strong recommendation; low-quality evidence)*.
6. Lipid formulation amphotericin B (3 - 5 mg/kg daily) is an alternative if there is intolerance or resistance to other antifungal agents *(strong recommendation; high-quality evidence)*.
7. Switchover from Amphotericin B to fluconazole is suggested after 5 - 7 days in clinically stable patients who have isolates susceptible to fluconazole and in whom repeat cultures on antifungal therapy have been sterile *(strong recommendation; high-quality evidence)*.

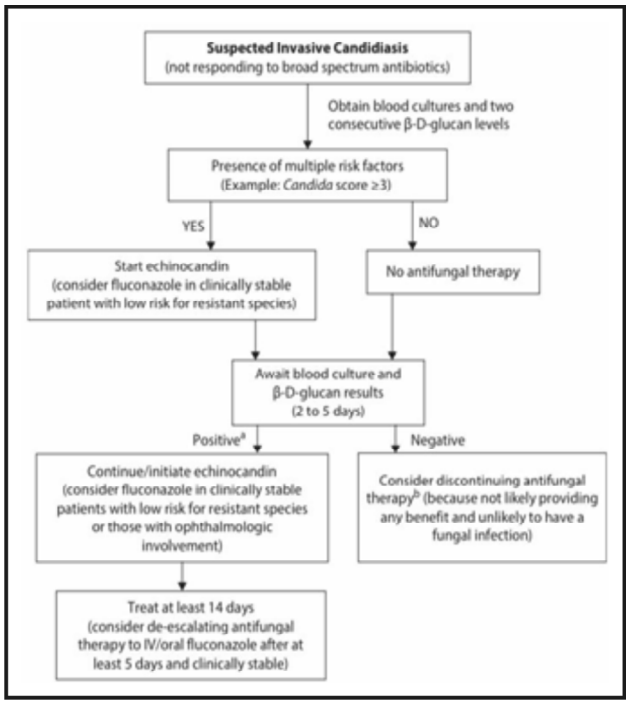

Fig. 1: *General approach to preemptive/empiric antifungal therapy.*

[a]Positive b-D-glucan diagnostic test result is two consecutive tests > 80 pg/ml.

[b]If clinically improving on antifungal therapy, then consider a short course of therapy for no more than 7 days.

Information from: Blot S, Charles PE. Fungal sepsis in the ICU: are we doing better? Trends in incidence, diagnosis, and outcome. *Minerva Anestesiol* 2013; 79: 1396-405.

8. In patients with suspected azole- and echinocandin-resistant Candida infections, lipid formulation amphotericin B is recommended *(strong recommendation; low-quality evidence)*.
9. Voriconazole is recommended as step-down oral therapy for selected cases of candidemia due to *C. krusei (strong recommendation; low-quality evidence)*.
10. All non-neutropenic patients with invasive candidiasis should have a dilated ophthalmological examination by an expert ophthalmologist, within first week of diagnosis *(strong recommendation; low-quality evidence)*.
11. Follow-up blood cultures should be done daily or on alternate days to define the reference point at which candidaemia has cleared *(strong recommendation; low quality evidence)*.
12. Recommended duration of therapy for invasive candidiasis without metastatic complications is 2 weeks after documented clearance of organisms from the bloodstream and resolution of clinical features related to infection *(strong recommendation; moderate-quality evidence)*.
13. Central vascular catheters (CVCs) must be removed at the earliest in the course of infection when the source is presumed to be the CVC; the decision must be customised for each individual *(strong recommendation; moderate-quality evidence)*.

Treatment for candidaemia in neutropenic patients[22] recommendations:

1. An echinocandin (caspofungin: 70 mg loading dose followed by 50 mg daily maintenance; micafungin: 100 mg per day; anidulafungin: 200 mg loading dose followed by 100 mg daily maintenance) should be initiated as starting therapy (strong recommendation; moderate-quality evidence).
2. Lipid formulation amphotericin B (3 - 5 mg/kg daily) is an alternative if there is intolerance or resistance to other antifungal agents (strong recommendation; moderate-quality evidence).
3. Fluconazole, 12 mg/kg loading dose followed by 6 mg/kg daily maintenance, is an alternative to echinocandins as starting therapy in patients who are not critically ill and who are not likely to have fluconazole-resistant Candida isolates *(weak recommendation; low-quality evidence)*.
4. Fluconazole, 6 mg/kg daily, is a useful option for stepdown oral therapy in clinically stable patients with persistent neutropenia, who have sensitive isolates and definitive bloodstream clearance *(weak recommendation; low-quality evidence)*.
5. Voriconazole, 6 mg/kg twice daily for 2 loading doses followed by 3 - 4 mg/kg twice daily maintenance therapy, can be utilised where additional mould coverage is required *(weak recommendation; low-quality evidence)*. Voriconazole can also be used as an oral step-down option in clinically stable patients with neutropenia, who have definitive bloodstream clearance and established voriconazole sensitivity *(weak recommendation; low-quality evidence)*.
6. In *C. krusei infection*, echinocandins, lipid formulation Amphotericin B or voriconazole is recommended *(strong recommendation; low-quality evidence)*.
7. The minimum duration of therapy recommended for invasive candidiasis without metastatic complications is 2 weeks after established Candida clearance from the bloodstream, alongwith resolution of neutropenia and clinical features related to invasive candidiasis *(strong recommendation; low-quality evidence)*.
8. In neutropenic patients, sources of infection other than

vascular access devices (e.g., gastrointestinal tract) predominate. Removal of such infected invasive devices must be done promptly on an individual basis *(strong recommendation; low-quality evidence)*.

9. Granulocyte colony-stimulating factor (G-CSF) transfusions can be considered in cases of persistent candidaemia with predictable prolonged neutropenia *(weak recommendation; low-quality evidence)*.

Treatment for chronic disseminated candidiasis[22] recommendations:

1. Initiation of therapy with lipid formulation Amphotericin B or an echinocandin for several weeks, is recommended. It is followed by oral fluconazole, 6 mg/kg daily, for patients not likely to grow fluconazole resistant isolate on culture *(strong recommendation; low-quality evidence)*.
2. Treatment must be continued till lesions resolve on repeated imaging, can take several months. Premature termination of antifungal therapy commonly leads to relapse *(strong recommendation; low-quality evidence)*.
3. If hematopoietic cell transplantation or anticancer chemotherapy is warranted, it should not be delayed due to the presence of disseminated candidiasis, and antifungal therapy should be continued throughout the period of high-risk to prevent recurrence *(strong recommendation; low-quality evidence)*.
4. In patients with incapacitating persistent fever, brief duration (1 - 2 weeks) therapy with nonsteroidal anti-inflammatory drugs and/or steroids must be considered *(weak recommendation; low-quality evidence)*.

Treatment for intra-abdominal candidiasis[22] recommendations

1. Source control must be achieved in all cases with intra-abdominal candidiasis, with appropriate drainage and/or debridement *(strong recommendation; moderate-quality evidence)*.
2. Choice of antifungal agent is the same as for the treatment of invasive candidiasis or empiric therapy for non-neutropenic patients in the ICU *(strong recommendation; moderate-quality evidence)*.
3. Duration of therapy has to be defined by adequacy of source control and clinical resolution of clinical features *(strong recommendation; low-quality evidence)*.

Treatment of Candida isolates from the tespiratory tract[22] recommendations

Candida growth from respiratory secretions mostly denotes colonisation and seldom requires therapy with antifungal agents *(strong recommendation; moderate-quality evidence)*. Isolation of candida from the respiratory tract of critically ill patients is usual but the occurrence of pneumonia from these organisms is uncommon due to innate mechanisms of defence within the lungs. Clinical decision to treat must be based on evidence of invasive disease or host factors indicating an increased risk of infection with a different source. Definite host factors include neutropenia, hematopoietic stem cell transplant, immunosuppressive therapy, steroids and severe immunodeficiency. Most of the available antifungals penetrate the lung well and are viable options.

Treatment for Candida intravascular infections, including endocarditis and infections of implantable Cardiac devices[22] recommendations

1. Lipid formulation Amphotericin B with or without flucytosine, 25 mg/kg 4 times daily, OR high-dose echinocandins (caspofungin 150 mg daily, micafungin 150 mg daily, or anidulafungin 200 mg daily) are recommended as initial therapy for native valve endocarditis *(strong recommendation; low-quality evidence)*.
2. Fluconazole, 6 - 12 mg/kg daily, is recommended as step down therapy for patients who have sensitive candida isolates with clinically stable condition and have candida clearance from the bloodstream *(strong recommendation; low-quality evidence)*.
3. Oral voriconazole, 3 - 4 mg/kg twice daily, or posaconazole, 300 mg daily, are considered as step-down therapy for isolates that are susceptible to these agents but resistant to fluconazole *(weak recommendation; very low-quality evidence)*.
4. Replacement of affected valve is recommended and drug therapy must be continued for at least 6 weeks after surgery and for an even longer duration in subjects with perivalvular abscesses and other related complications *(strong recommendation; low-quality evidence)*.
5. In patients who are not suitable candidates for valve replacement, long-term suppressive therapy with fluconazole is recommended in drug sensitive cases *(strong recommendation; low-quality evidence)*.
6. In prosthetic valve endocarditis, similar regimens to native valve endocarditis are recommended (strong recommendation; low-quality evidence). Chronic suppressive antifungal therapy with fluconazole is recommended to prevent relapse *(strong recommendation; low-quality evidence)*.

7. In case of pacemaker and implantable cardiac defibrillator infections, whole of the device must be removed at the earliest *(strong recommendation; moderate-quality evidence)*. Antifungal therapy is similar as that recommended for native valve endocarditis *(strong recommendation; low quality evidence)*.
8. Four weeks of antifungal therapy is recommended for infections limited to generator pockets after device removal *(strong recommendation; low-quality evidence)*.
9. Six weeks of antifungal therapy is recommended for infections involving the wires after wire removal *(strong recommendation; low-quality evidence)*.
10. Antifungal regimen is the same as that recommended for native valve endocarditis for ventricular assist devices that cannot be removed *(strong recommendation; low-quality evidence)*. Chronic suppressive therapy with fluconazole is recommended if the isolate is susceptible, for as long as the device remains in place *(strong recommendation; low-quality evidence)*.

Treatment for central nervous system Candidiasis and other yeasts[22] recommendations

1. Liposomal Amphotericin B, 5 mg/kg daily, with or without oral flucytosine, 25 mg/kg 4 times daily is recommended for initial treatment *(strong recommendation; low-quality evidence)*.
2. Fluconazole 6 - 12 mg/kg daily is recommended for step-down therapy after the patient has responded to initial therapy *(strong recommendation; low-quality evidence)*.
3. Treatment must be continued until all the clinical features alongwith CSF and radiological abnormalities have resolved completely *(strong recommendation; low-quality evidence)*.
4. Infected CNS devices, including ventriculostomy drains, shunts, stimulators, prosthetic reconstructive devices, and biopolymer wafers that deliver chemotherapy must be removed whenever possible *(strong recommendation; low-quality evidence)*.
5. In patients where ventricular device cannot be removed, Amphotericin B deoxycholate must be given through the device into the ventricle at doses ranging from 0.1 mg to 0.5 mg in 2 mL of 5% dextrose *(weak recommendation; low-quality evidence)*.

Treatment for urinary tract infections due to Candida species[22] recommendations

1. Treatment of asymptomatic candiduria with antifungals is NOT warranted unless the subject belongs to a category at high risk for dissemination; patients at high risk include neutropenic patients and patients undergoing urological instrumentation *(strong recommendation; low-quality evidence)*.
2. Patients undergoing urological instrumentation must be treated with oral fluconazole, 6 mg/kg daily, OR Amhotericin B deoxycholate, 0.3 - 0.6 mg/kg daily, both before and after the instrumentation *(strong recommendation; low-quality evidence)*.
3. In symptomatic ascending candida pyelonephritis, oral fluconazole 3 - 6 mg/kg daily for 2 weeks is recommended for fluconazole susceptible isolates *(strong recommendation; low-quality evidence)*.
4. In fluconazole-resistant *C. glabrata*, Amphotericin B deoxycholate, 0.3 - 0.6 mg/kg daily for 7 days with or without oral flucytosine, 25 mg/kg 4 times daily, is recommended *(strong recommendation; low-quality evidence)*.
5. For *C. krusei*, Amphotericin B deoxycholate, 0.3 - 0.6 mg/kg daily, for 1 - 7 days is recommended *(strong recommendation; low quality evidence)*.
6. Removal of urinary tract obstruction is strongly recommended *(strong recommendation; low-quality evidence)*. Patients with nephrostomy tubes or DJ stents in situ should be considered for device removal or replacement, if possible *(weak recommendation; low-quality evidence)*.

Invasive mould infections

Prophylactic and empirical therapy

The guidelines for management of infections due to invasive moulds in critical patients are largely derived from clinical studies involving treatment of haematological malignancies. Lipid formulations of Amphotericin B remain the most widely considered for empiric therapy in the setting of an unidentified invasive mould and in subjects with history of recent azole therapy. Voriconazole is suggested as first-line therapy in infections with *Aspergillus*. The echinocandins are also active against *Aspergillus*. But only caspofungin has been approved for this indication. Prophylactic treatment in immunocompetent or non-neutropenic patients cannot be strongly recommended based on current clinical studies. Empirical therapy must be instituted even in those without traditional risk factors, at the earliest clinical suspicion for invasive aspergillosis (IA). There is no well-defined duration of therapy, although a prolonged course is usually required.

Combination antifungal therapy

Combination therapy for invasive aspergillosis is an alternative for rescue therapy in individuals unresponsive to a single agent or with breakthrough symptoms while on therapy. Approximately 25 - 35% of critical patients in ICUs are suspected to have resistant mycotic infections, and meta-analysis of various studies has shown that around 45 - 55% of patients are usually administered combination antifungal therapy. Usual regimens include two agents with dissimilar action mechanisms, like an echinocandin (acting on fungal cell wall) with either amphotericin B (acting on fungal cell membrane) or an azole. Because of the high probability for antagonism, both amphotericin B and azole together are not used in combination therapy. A randomized clinical study that compared the combination of voriconazole with anidulafungin versus voriconazole alone established a tendency towards decreased mortality in haematopoietic stem cell transplant recipients with combination antifungal therapy[25].

COVID-19 associated pulmonary aspergillosis (CAPA)

COVID-19 associated pulmonary aspergillosis (CAPA) has been reported to worsen the disease course of COVID-19, resulting in increased mortality. Usual risk factors are corticosteroid therapy or anti-interleukin 6 therapy usually given for cytokine release syndrome. Also, the initially reported few cases have been found to be azole-resistant. The first line recommended therapy is either voriconazole or isavuconazole. But in azole-resistant CAPA, liposomal amphotericin B is the drug of choice. CAPA is defined as possible, probable or proven, based on sample validity and thus diagnostic certainty. Radiological findings are not sufficient to define patients with CAPA, but multiple pulmonary nodules or lung cavitation should prompt thorough investigation for CAPA in COVID-19 patients. Frequently observed radiological features of invasive pulmonary aspergillosis, such as the halo sign, are not sufficient to define CAPA without mycological evidence. This feature is insufficient because the halo sign suggests local infarction, and can be seen in severe COVID-19 due to endothelialitis and thrombosis. Fungal biomarkers have poor predictive value in diagnosing CAPA as compared to invasive pulmonary aspergillosis. Histopathological or direct microscopic detection of fungal hyphae, showing invasive growth with associated tissue damage; or aspergillus recovered by culture or microscopy or histology or PCR obtained by a sterile aspiration or biopsy from a pulmonary site, showing an infectious disease process are the only ways to conclusively prove CAPA.

Voriconazole treatment (loading dose 6 mg/kg twice a day for two doses, followed by 4 mg/kg twice a day) has a better outcome than does treatment with amphotericin B deoxycholate. However, liposomal amphotericin B can be considered for initial therapy if, epidemiologically, drug-resistant patterns support this treatment, before the results of susceptibility testing for voriconazoles are available.

Isavuconazole (loading dose 200 mg three times a day for six doses, followed by 200 mg once a day, 12 - 24 h after the last loading dose) has similar clinical efficacy to voriconazole but less hepatotoxicity and neurotoxicity and decreased risk of corrected QT-interval prolongation. Posaconazole has excellent *in-vitro aspergillus* activity and has been successfully used as salvage treatment in patients without COVID-19. Duration of therapy recommended is upto 6 to 12 weeks depending upon the clinical response[26].

COVID-19 associated mucormycosis (CAM)

Mucormycosis is a medical emergency even when clinically suspected. During COVID-19 pandemic, there have been various reports of mucormycosis among patients with COVID-19, especially in those who are diabetic or those who have received steroids. Also, patients who received empirical voriconazole therapy were found to be at an increased risk for getting mucormycosis. COVID-19 associated mucormycosis (CAM) has resulted in high morbidity and mortality, exorbitant treatment costs and shortage of antifungal drugs. There are two usual clinical presentations: rhino-orbito-cerebral mucormycosis (ROCM) and pulmonary mucormycosis.

ROCM may manifest as nasal blockade or congestion, nasal discharge (bloody or brown/black), local pain, facial pain or numbness or swelling, headache, orbital pain, toothache, loosening of maxillary teeth, jaw involvement, blurred or double vision with pain; paresthesia, fever, skin lesions, thrombosis or necrosis (eschar).

Pulmonary mucormycosis may manifest as fever, cough, chest pain, pleural effusion, haemoptysis, worsening of respiratory symptoms. Lung CT may be confused with COVID-related shadows; suspect mucormycosis in patients with thick-walled lung cavity (need to differentiate from covid-associated pulmonary aspergillosis), reverse halo sign, multiple nodules, pleural effusion (not usually seen in aspergillosis). Also, there are repeated negative galactomannan and beta-glucan tests in CAM.

Suspected patients should undergo appropriate radio-imaging study: MRI paranasal sinuses with brain contrast study for ROCM, CT thorax for pulmonary mucormycosis. Histopathological examination of biopsy (endoscopic or CT-guided) specimen from affected tissues/sites, and staining with hematoxylin-eosin, periodic acid-Schiff stain or Grocott-Gomori's methanamine-silver stain reveals aseptate or pauci-septate hyphae (ribbon like) which are irregularly branching at 90^0 angle, indicates mucorales.

Surgical debridement and antifungal therapy are the mainstay of treatment and usually require a multi-disciplinary approach. Liposomal amphotericin B (5 mg/kg/day), diluted in 5% dextrose may be given over 2 - 3 hours infusion. Slow escalation should be avoided and higher doses upto 10 mg/kg/day may be given with brain involvement. In patients who are intolerant to amphotericin B, alternative agents like posaconazole or isavuconazole may be used. Posaconazole: 300 mg twice a day on first day, followed by 300 mg once a day. Check posaconazole trough level after 7 days of therapy and avoid interacting drugs. Isavuconazole: 200 mg three time a day for two days, followed by 200 mg once a day. Isavuconazole fared better than posaconazole in mucormycosis in various clinical studies. After 3 to 6 weeks of amphotericin B therapy, consolidation therapy with either posaconazole or isavuconazole should be given for atleast 3 to 6 months or as per clinical response[27].

Pharmacological aspects of antifungal agents

Amphotericin B

Amphotericin B is the most important therapeutic option for treatment of invasive fungal infections among critically ill patients. It has wide spectrum anti-fungal activity against a variety of fungal microbes causing infections in ICU patients. Amphotericin B is recommended for disseminated infections in immunocompromised patients, in CNS involvement, or when resistance to other antifungal agents is seen. Amphotericin B deoxycholate used to be the main formulation available before the advent of three newer adjuvant products (i.e., amphotericin B lipid complex, liposomal amphotericin B and amphotericin B colloidal dispersion). All three formulations have similar efficacy but significantly favourable toxicity profile than the parent compound. The liposomal product has lesser nephrotoxicity compared to the other two lipid-based products. Nephrotoxicity can be mitigated with all the amphotericin B formulations by appropriate hydration of the patient with a normal saline loading (250 - 500 ml) prior to each administered dose and by avoiding concomitant use of other nephrotoxic drugs, particularly diuretics. Nephrotoxicity can also be prevented by using continuous drug infusions but should be avoided because the concentration-dependent pharmacodynamics of amphotericin B must not be compromised. Lipid formulations of amphotericin B, except amphotericin B colloidal dispersion, have more than 50% reduced rate of infusion-related reactions than deoxycholate formulation. Infusion-related adverse events can be reduced further with the use of diphenhydramine and acetaminophen 30 minutes before starting the infusion. Other noteworthy reactions defined with the liposomal products are flushing, retrosternal chest heaviness, hypoxia, flank pain and urticarial rashes. Amphotericin B usage is commonly associated with hypokalaemia and hypomagnesaemia. Amphotericin B binds to the fungal cell wall ergosterol, thereby altering its permeability; tissue binding of the drug may also occur in mammalian renal cells and cause potassium loss. Regular electrolyte monitoring and correction are recommended.

Echinocandins

The first available echinocandin was approved by the FDA in 2001 and changed the entire approach to management of disseminated fungal infections. Echinocandins have a unique mechanism of action specific to the fungal cell wall. They have fungicidal activity against *Candida* species and fungistatic activity against moulds, and are also active against the biofilms. They are not recommended for treatment of fungal urinary tract infections as they are not well excreted in urine, thus having poor drug concentration there. Adverse effects associated with the echinocandins are usually benign, with few reports of hepatotoxicity and infusion related reactions. The infusion reactions are mostly histamine-mediated and are similar to the red-man syndrome usually seen with vancomycin. The echinocandins do not have any significant interactions with the hepatic CYP enzymes; thus, drug interactions are negligible. Caspofungin and micafungin are reported to increase serum levels of tacrolimus and cyclosporine but dose reductions are not usually required; monitoring of serum drug concentrations is useful.

Extended-spectrum triazoles

These agents offer increased activity against many *candida* and other yeasts, alongwith a variety of moulds. Availability of these agents has enhanced the therapeutic options for management of invasive mycoses by providing an effective oral alternative. Oral absorption of triazoles is modified depending upon how they are administered. Voriconazole should be given before meals as presence of food reduces absorption by almost 25%. On the contrary, posaconazole must be given with high-fat meals. Gastric acid also increases the absorption of posaconazole. Therefore, concomitant gastric acid suppression therapy must not be used alongwith posaconazole therapy. Proton pump inhibitors should also be avoided. There is lack of recommendations for dose adjustment with these agents; thus, therapeutic drug monitoring (TDM) may be required for ensuring clinical efficacy.

These agents may cause liver toxicity, adrenal suppression, and QT_c prolongation. Also, significant visual disturbances occur with use of voriconazole. Both oral as well as intravenous therapy cause visual impairments but are usually transient, with patients adjusting to them within 1 - 2 weeks

of therapy. These vision issues are typically-related to the initiation and temporal administration of the drug and have been described as bright flashing lights or hallucinations.

The intravenous formulation of voriconazole contains a second-generation cyclodextrin-solubilising agent. First generation cyclodextrins were reported to cause nephrotoxicity and accumulate in renal failure. It is recommended to use oral voriconazole in patients with creatinine clearance below 50 ml/min.

Clinical interaction with CYP hepatic enzymes is seen with all azoles, with voriconazole and posaconazole being strong inhibitors of CYP3A4. It can lead to significant increase in the serum levels of tacrolimus, sirolimus and cyclosporine. The interaction with sirolimus is totally unpredictable and warrants frequent concentration monitoring, if the combination cannot be avoided. Drug interactions may occur in critical patients receiving triazoles with excessive exposure to midazolam, fentanyl, phenytoin, steroids, warfarin and quetiapine.

Voriconazole is a moderate inhibitor of CYP2C19. It must be used cautiously in combination with strong inhibitors or inducers of CYP2C19 enzyme, like rifampin, and with drugs metabolised by CYP2C19, like clopidogrel. Non-linear pharmacokinetics make voriconazole dose adjustments difficult in the setting of complex critical care regimen. Concomitant administration of azoles with other QT_c -prolonging drugs must be monitored strictly or altogether avoided.

The FDA approved isavuconazonium sulfate for the treatment of patients with invasive aspergillosis and mucormycosis in 2015. Isavuconazonium is a prodrug which is hydrolysed to isavuconazole, the active form which can be dosed once a day. Isavuconazole is reported to be noninferior to voriconazole in the treatment of invasive aspergillosis and mucormycosis resistant to other antifungal agents. The spectrum of activity includes *C. glabrata* (including fluconazole-resistant strains) and *C. krusei*, alongwith Cryptococcus, Coccidioides, Blastomycoses and Histoplasmosis. Isavuconazole is available in an intravenous formulation which does not contain the cyclodextrin excipient, but it does require an in-line filter. Oral formulation is also available, but these capsules cannot be opened and administered via nasogastric tube. Isavuconazole is a moderate inhibitor of CYP3A4 enzyme. The drug interactions are clinically not relevant with isavuconazole as compared to other azoles. Adverse effects are mostly gastrointestinal in the form of nausea, vomiting, diarrhoea, constipation. Hypokalaemia, hepatotoxicity, shortened QT_c interval and infusion reactions are reported in clinical trials. Longer half-life makes management of drug interactions and adverse effects difficult.

An inhibitor of β-(1,3)-glucan synthase is biafungin which is a novel long-acting echinocandin under development. It's long half-life should allow for a once a week dosing and can be used against drug resistant fungal isolates.

Dosing considerations

Inappropriate dosing of antimicrobials is not uncommon in the ICU due to pharmacokinetic-pharmacodynamic variations in critical patients (e.g., increment in distribution volume and increased drug filtration). It commonly leads to increased resistance, treatment failure and poor clinical outcomes. Fluconazole is commonly under-dosed by omission of loading doses and by prescribing fixed (400 mg) versus weight based (6 mg/kg) dosing.

Another important factor in the alteration of kinetics of antifungal agents is obesity. Obese patients have larger variations in the distribution volume of drugs. Both fat as well as lean body mass are more in obese patients, and the blood flow is decreased to the adipose tissue. Such individuals have reduced hepatic/splanchnic blood flow and metabolism due to the hepatic fat infiltration. Also, metabolism of CYP3A4 is reduced in obese patients leading to disturbances in therapeutic drug concentration. Renal clearance increases with increased lean body mass, leading to greater renal clearance of these drugs. As amphotericin B formulations do not get distributed into the adipose tissue, they should be dosed as per lean body weight. On the other hand, fluconazole should be dosed at the higher end of the dose range, based on total body weight. Dosing of voriconazole and posaconazole should be based on lean body weight, whereas dosing should be increased by upto 25% to 50% for echinocandins.

Therapeutic drug monitoring

Therapeutic drug monitoring (TDM) helps in guiding antifungal therapy in a more effective manner. Although routine monitoring is not recommended in all cases, there may be numerous clinical scenarios where TDM is very important, especially in the critical patients.

When to consider therapeutic drug level monitoring[28]

Extremes of age
Questionable compliance
Multiple drug interactions
Concomitant use of acid suppressing therapy
Malabsorption
Morbid obesity
Extensive or disseminated disease (e.g., CNS, Mediastinal)
Multiple organ dysfunction
Renal replacement therapy
Prophylaxis in high-risk patients
Use of ECMO or cardiopulmonary bypass

Monitoring of therapeutic concentration levels of antifungal drugs is mainly restricted to the triazole class which covers moulds (i.e., itraconazole, voriconazole, and posaconazole) and flucytosine. The target trough levels, serum sampling timings and recommendations for dose modifications for these drugs are listed in Table V. Monitoring of concentrations of amphotericin B or the echinocandins is not warranted. Monitoring is also not routinely suggested for fluconazole; although, it is considered when the MIC of the pathogen is increased, in CNS disease, or in patients requiring renal replacement therapy.

Itraconazole demonstrates non-linear pharmacokinetics with wide variations in the oral absorption because of changes in the formulations (30% higher AUC with the oral solution vs. capsules), food, and gastric pH. Therefore, monitoring is warranted in most patients receiving this agent to ensure adequate absorption.

Non-linear pharmacokinetic variability with voriconazole is mainly due to genetic polymorphisms of the CYP2C19 hepatic enzyme and saturable hepatic metabolism. There is also evidence suggesting that the FDA-approved non–weight-based fixed dose oral formulations of voriconazole could be insufficient to achieve effective serum concentrations. It is of prime concern in obese patients and those with active disease, in whom the maximal recommended dose may need to be exceeded, thus warranting monitoring.

Posaconazole TDM is recommended in most patients receiving the suspension because of poor bioavailability from saturable absorption and reduced absorption in the setting of mucositis, graft-versus-host disease, the concomitant administration with acid-suppressing therapies, or administration without a high-fat meal. Drug levels are reported to be suboptimal in 50% of subjects receiving posaconazole suspension for fungal prophylaxis in few studies.

Flucytosine is a pyrimidine analogue used in combination with amphotericin B to treat cryptococcal meningitis. TDM is needed due to its toxicity profile, significant intra- and inter-patient pharmacokinetic variability, dependence on renal elimination where nephrotoxicity is common, and high-risk of developing resistance. Evidence from pharmacodynamics studies indicates a significant exposure-toxicity correlation with a higher incidence of bone marrow suppression and hepatotoxicity seen at peak concentrations exceeding 100 mg/l. Maintaining concentrations above the MIC for at least 50% of the dosage interval is associated with improved clinical outcomes and may prevent the emergence of resistance. It is recommended that peak concentrations be performed to prevent toxicity and minimise the risk of resistance.

Antifungal stewardship

Antifungal stewardship consists of coordinated efforts for surveillance and guidance of appropriate usage of antifungal agents (in terms of both the choice of the selected agent as well as the correct dosage) in order to achieve the best possible clinical results and reduce the incidence of

Table V: Recommendations for therapeutic drug monitoring.

Drug	Minimum target Concentrations[a]	Timing of Concentrations[b]	Concentrations Associated with toxicity	Strategies to increase low concentrations
Itraconazole	P: 0.5 mg/l T: 0.6 - 1 mg/l	7 - 14 days	≥ 17 mg/l[c]	Change to solution Avoid acid suppressants with capsules Take solution in fasting state Increase dose from 200 mg twice daily to 300 mg twice daily
Voriconazole	P: > 1 mg/l T: > 1 mg/l Trough: MIC of 2 - 5	Within 7 days[d]	> 5.5 mg/l	Increase dose: IV: up to 6 mg/kg twice daily PO: up to 300 mg twice daily
Posaconazole[e]	P: 0.35 mg/l P: > 0.7 mg/l	At 48 hours Within 7 days	Unknown	Increase total daily dose to 800 mg Administer total daily dose divided four times daily Switch to the delayed-release tablets Avoid acid suppressants Take with food or high-fat supplement
Flucytosine	T: Peak 20-40 mg/l	Within 72 hours	Peak >100 mg/l	Increase dose by 50%, use caution due to toxicity

[a]Trough concentrations measured using high performance liquid chromatography (HPLC)/mass spectrometry unless otherwise specified. [b]Time listed is the number of days after the initiation of the therapy. [c]Concentration measured with bioassay, would expect 5-fold lower concentration with HPLC/mass spectrometry. [d]Repeat level may be necessary because of fluctuations in concentrations due to Michaelis-Menten kinetics. [e]Recommendations are for oral solution only.

IV = Intravenous; P = Prophylaxis; PO = Oral; T = Treatment.

Information from Ashbee HR. Therapeutic drug monitoring of antifungal agents: guidelines from the British Society of Medical Mycology. *J Antimicrob Chemother* 2014; 69: 1162-76.

selective errors and adverse events. Antifungal utilisation has gradually increased over time in conjunction with an increase in the number of immunosuppressed individuals at high-risk for invasive mycoses. Difficulty in diagnosis of invasive fungal infections leads to delays in institution of specific therapy, and subsequently worse clinical outcomes. There is also emerging data correlating prior antifungal exposure and suboptimal dosing to emergence of antifungal resistance. Antimicrobial stewardship programs must constitute a multi-disciplinary bundle based approach to ensure appropriate utilisation of antifungals via post-prescription review, feedback and prior authorisation from the infectious disease specialist. Institutional guidelines should also be formulated to guide diagnostic testing in high-risk individuals; appropriate selection, dosing, and duration of the antifungal agent; therapeutic drug monitoring, if warranted; and opportunities for de-escalation and stepping down of therapy.

Conclusion

Critical components in the management of invasive fungal infections that are clinician mitigated include (1) prompt antifungal therapy, (2) risk factor analysis to identify patients at greater risk than the usual ICU population for IFI and therefore in need of prophylactic or pre-emptive therapy given the current lack of prompt accurate diagnostics, (3) choice of the appropriate antifungal agent and dosing regimen, and (4) source control. Currently, until a species diagnosis or susceptibility is known, an echinocandin is the recommended first-line therapy for most of the patients with IFI. PK/PD studies suggest that the currently recommended regimens would be useful for most infections. Once the fungal speciation is done for *C. albicans*, *C. parapsilosis* or *C. tropicalis* and if the patient is responding to initial therapy, the appropriate therapy would be to step down to fluconazole. For other species, the therapy should be directed based on susceptibility profile.

Key words: *Fungal Sepsis, Invasive Mycoses, Candidaemia, Aspergillosis, Mucormycosis, Antifungal Stewardship.*

References

1. Lortholary O, Renaudat C, Sitbon K *et al*. French Mycosis Study Group. Worrisome trends in incidence and mortality of candidaemia in intensive care units (Paris area, 2002-2010). *Intensive Care Med* 2014; 40: 1303-12.
2. Morrell M, Fraser VJ, Kollef MH. Delaying the empiric treatment of candida bloodstream infection until positive blood culture results are obtained: a potential risk factor for hospital mortality. *Antimicrob Agents Chemother* 2005; 49: 3640-5.
3. Pfaller MA, Diekema DJ. Epidemiology of invasive candidiasis: a persistent public health problem. *Clin Microbiol Rev* 2007; 20 (1): 133-63.
4. Sharma C, Kumar N, Meis JF *et al*. Draft genome sequence of a fluconazole-resistant Candida auris strain from a candidaemia patient in India. *Genome Announc* 2015; 3: e00722-15. https://doi.org/10.1128/genomeA.00722-15.
5. Chatterjee S, Alampalli SV, Nageshan RK *et al*. Draft genome of a commonly misdiagnosed multidrug resistant pathogen Candida auris. *BMC Genomics* 2015; 16: 686. https://doi.org/10.1186/s12864-015-1863-z.
6. Maschmeyer G, Haas A, Cornely OA. Invasive aspergillosis: epidemiology, diagnosis and management in immunocompromised patients. *Drugs* 2007; 67 (11): 1567-601.
7. Meersseman W, Lagrou K, Maertens J *et al*. Invasive aspergillosis in the intensive care unit. *Clin Infect Dis* 2007; 45 (2): 205-16.
8. Husain S, Alexander BD, Munoz P *et al*. Opportunistic mycelial fungal infections in organ transplant recipients: emerging importance of non-Aspergillus mycelial fungi. *Clin Infect Dis* 2003; 37 (2): 221-9.
9. Piarroux R, Grenouillet, R, Balvay P *et al*. Assessment of preemptive treatment to prevent severe candidiasis in critically ill surgical patients. *Crit Care Med* 2004; 32: 2443-9.
10. Karageorgopoulos DE, Vouloumanou EK, Ntziora F *et al*. b-DGlucan assay for the diagnosis of invasive fungal infections: a meta-analysis. *Clin Infect Dis* 2011; 52: 750-70.
11. Hanson KE, Pfeiffer CD, Lease E *et al*. β-D-glucan Surveillance with preemptive anidulafungin for invasive candidiasis in intensive care unit patients: A randomised pilot study. *PLoS ONE* 2012; 7: e42282.
12. Mikulska M, Calandra T, Sanguinetti M *et al*. Third European Conference on Infections in Leukemia Group. The use of mannan antigen and anti-mannan antibodies in the diagnosis of invasive candidiasis: recommendations from the Third European Conference on Infections in Leukaemia. *Crit Care* 2010; 14: R222.
13. Boluk G, Kazak E, Ozkalemkas F *et al*. Comparison of galactomannan, b-D-glucan, and Aspergillus DNA in sera of high-risk adult patients with haematological malignancies for the diagnosis of invasive aspergillosis. *Turk J Med Sci* 2016; 46: 335-42.
14. Racil Z, Kocmanova I, Lengerova M *et al*. Intravenous PLASMA-LYTE as a major cause of false-positive results of Platelia Aspergillus test for galactomannan detection in serum. *J Clin Microbiol* 2007; 45: 3141-2.
15. Kourkoumpetis TK, Fuchs BB, Coleman JJ *et al*. Polymerase chain reaction-based assays for the diagnosis of invasive fungal infections. *Clin Infect Dis* 2012; 54: 1322-31.
16. Forrest GN *et al*. Peptide nucleic acid fluorescence in situ hybridisation-based identification of Candida albicans and its impact on mortality and antifungal therapy costs. *J Clin Microbiol* 2006; 44: 3381-3.
17. Bellanger A-P, Gbaguidi Haore H, Liapis E *et al*. Rapid identification of Candida sp. by MALDI TOF mass spectrometry subsequent to short term incubation on a solid medium. *APMIS* 2019; 127: 217-21.
18. Sinnollareddy MG, Roberts JA, Lipman J *et al*. Pharmacokinetic variability and exposures of fluconazole, anidulafungin and caspofungin in intensive care unit patients: Data from multinational Defining Antibiotic Levels in Intensive care unit (DALI) patients Study. *Crit Care* 2015; 19: 33.
19. Maubon D. Resistance of *Candida spp.* to antifungal drugs in the ICU: where are we now? *Int Care Med* 2014; 40: 1241-55.
20. Cuenca-Estrella M. Antifungal drug resistance mechanisms in pathogenic fungi: from bench to bedside. *Clin Microbiol Infect*

2014; 20 (suppl 6): 54-9.

21. Ferreira D, Grenouillett F, Blasco G *et al*. Outcomes associated with routine systemic antifungal therapy in critically ill patients with Candida colonisation. *Int Care Med* 2015; 41: 1077-88.
22. Pappas PG, Kauffman CA, Andes DR *et al*. Clinical Practice Guideline for the Management of Candidiasis: 2016 Update by the Infectious Diseases Society of America. *Clinical Infectious Diseases* 2016; 62 (4): e1–e50. https://doi.org/10.1093/cid/civ933.
23. Knitsch W, Vincent JL, Utzolino S *et al*. A randomised, placebo-controlled trial of preemptive antifungal therapy for the prevention of invasive candidiasis following gastrointestinal surgery for intra-abdominal infections. *Clin Infect Dis* 2015; 61 (11): 1671-8. doi:10.1093/cid/civ707.
24. Shields RK, Nguyen MH, Press EG *et al*. Abdominal Candidiasis is a Hidden Reservoir of Echinocandin Resistance. *Antimicrob Agents Chemother* 2014; 58: 7601-5.
25. Marr KA, Schlamm HT, Herbrecht R *et al*. Combination antifungal therapy for invasive aspergillosis, a randomised trial. *Ann Int Med* 2015; 162: 81-9.
26. Koehler P, Bassetti M, Chakrabarti A *et al*. Defining and managing COVID-19-associated pulmonary aspergillosis: the 2020 ECMM/ISHAM consensus criteria for research and clinical guidance. *Lancet Infect Dis* 2021; 21: e149-62. https://doi.org/10.1016/S1473-3099(20)30847-1.
27. Cornely OA, Alastruey-Izquierdo A, Arenz D *et al*. Global guideline for the diagnosis and management of mucormycosis. *Lancet Infect Dis* 2019; 19 (12): e405-e421. doi: 10.1016/S1473-3099(19)30312-3.
28. Ashbee HR. Therapeutic drug monitoring of antifungal agents: guidelines from the British Society of Medical Mycology. *J Antimicrob Chemother* 2014; 69: 1162-76.

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CASE REPORT

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Disseminated Cysticercosis: An Uncommon Case with Unusual Presentation

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Abstract

Cysticercosis is caused by the larval form of the tape worm Taenia solium. Disseminated cysticercosis (DCC) is an uncommon manifestation of this common disease with widespread dissemination throughout the human body. Dissemination of the cysticerci can result in involvement of almost any organ in the body and can have variable presentation. Though this form of disease was reported as early as 1912 by British Army medical officers stationed in India, less than 50 cases of DCC have been reported worldwide. Here we report an uncommon case of disseminated cysticercosis with an unusual presentation.

Key words: Cysticercosis, disseminated cysticercosis, Taenia solium, polyneuritis, myositis.

Case report

A 46-year-old female presented with complaints of pain and weakness of all 4 limbs for a few days. Pain was diffuse, more over the muscles and was progressive. She became incapacitated in the next few days and was brought to the hospital. Patient also had a history of headache for last few months which was diffuse, more or less continuous, not associated with visual disturbances or vomiting. There was no history of fever, loss of consciousness, seizures, bladder or bowel involvement, or any sensory loss. On examination, the patient was conscious and oriented, afebrile, and vitals were stable. CNS examination did not reveal meningeal signs or any cranial nerve involvement. DTRs were decreased and planter reflex was flexor; power could not be assessed due to pain and tenderness in the limbs. There was no sensory or autonomic involvement. Investigations revealed the patient to be a diabetic and hypothyroid for which treatment was started. Lab work-up was normal for blood counts, LFT, KFT, electrolytes, ANA and muscle enzymes. NCCT head and CSF examination were normal. Nerve conduction study showed decreased conduction velocity in bilateral median, ulnar, peroneal, and tibial nerves suggestive of demyelinating motor neuropathy. The patient was treated with IVIG on the suspicion of Guillain Barre' Syndrome (GBS). The patient improved and was discharged on analgesics, insulin, and thyroxine.

The patient presented again after a month with complaints of severe headache, blurring of vision, and radicular pain over buttocks and thighs. Her diabetes and hypothyroidism were under control. X-ray of thoracolumbar spine was normal. NCCT head was repeated which showed right occipital calcified granuloma. Ophthalmologic examination revealed papilloedema in both eyes with a cyst in the left medial rectus muscle. MRI scan of brain showed ocular cysticercosis, further confirmed by MRI (Fig. 1A). MRI lumbar spine showed myocysticercosis (Fig. 1B). Hence, an MRI whole body was done, which revealed disseminated cysticercosis involving brain, orbit, spinal cord and muscles (Fig. 2). Repeat nerve conduction study showed low amplitude and absent F-wave in right peroneal nerve and low NCV in right ulnar nerve with normal distal latencies suggesting motor involvement of upper and lower limbs.

The patient was treated with antiepileptic (levetiracetam) and systemic steroid (prednisolone) followed by Albendazole (15 mg/kg/day) and Praziquantel (20 mg/kg/day) for 3 weeks under supervision for any adverse event. Patient improved symptomatically; there was no limb pain while her headache and blurring of vision were reduced. She was discharged on Levetiracetam and tapering dose of Prednisolone, to be followed-up from OPD.

Discussion

Dissemination of human cysticercosis occurs when the embryo of *Taenia solium* enters the hepatoportal system from the intestine, from where it spreads to various tissues and organs of the body[1,2]. Clinical manifestations of cysticercosis depend upon the location of the cyst, cyst burden and host reaction.[4] Simultaneous and extensive involvement of the brain, spinal cord, eyes, muscles, and subcutaneous tissues is extremely rare. Disseminated

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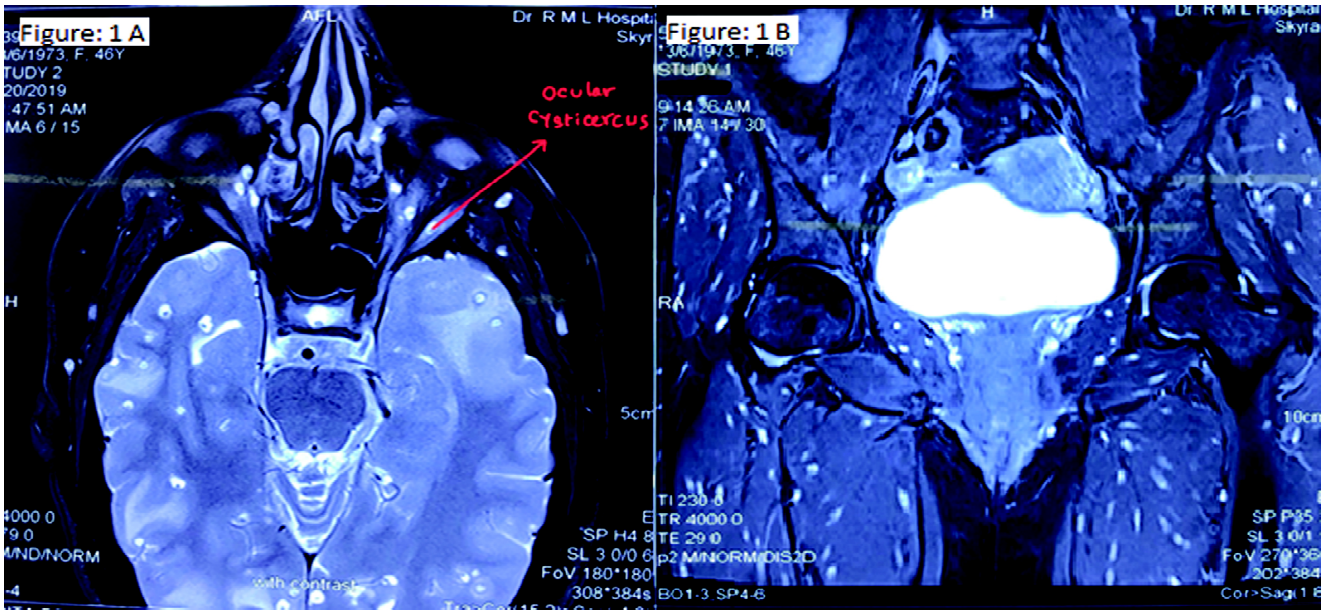

Fig. 1a and 1b:

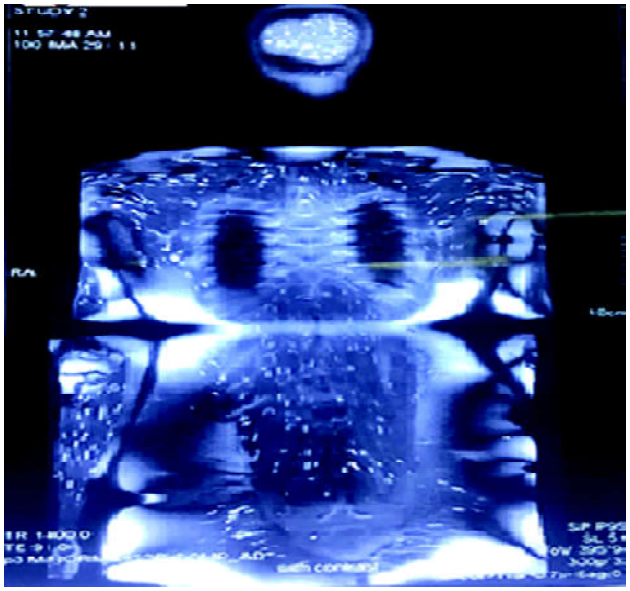

Fig. 2:

cysticercosis (DCC) is an uncommon manifestation of the disease, and less than 50 cases have been reported worldwide[1,3]. Neurocysticercosis (NCC) is considered to be the most common parasitic infestation of the central nervous system (CNS); though most of the NCC cases present with headache and seizures a few may be asymptomatic and detected incidentally on imaging. Unusual presentations of cysticercosis has been reported in literature and include CNS demyelination[5], arthritis[6], myositis[7], and lumbosacral radiculopathy[8].

Anatomical localisation of the cysts can be done using computed tomography scans and magnetic resonance imaging (MRI), though MRI is more sensitive than a CT. MRI is helpful in identifying the scolex and live cysts in cisternal spaces and ventricles[9]. MRI can also be used to identify the response to treatment[3].

Treatment with the cysticidal drugs praziquantel and albendazole is indicated as they help by reducing the parasite burden[10]. However, the pharmacological treatment may be associated with severe reactions, which may result from massive release of antigens causing local tissue swelling and generalised seizure activity. Corticosteroids and antiepileptics decrease the incidence of such complications when used before starting the cysticidal drug[10].

In our case, disseminated cysticercosis had a very unusual presentation simulating polymyositis or polyneuritis. There are rare reports of myositis in cysticercosis[7], However, nerve conduction study and normal muscle enzymes ruled this out in our case. Initial nerve conduction study was suggestive of demyelinating neuropathy involving upper and lower limbs and our patient improved with IVIG treatment, at least temporarily, suggesting a diagnosis of GBS, though CSF did not show any albumino-cytological dissociation. Despite extensive review of literature, we could not find any association between GBS and cysticercosis.

The repeat nerve conduction study suggesting motor neuropathy and radicular pain experienced by the patient

can be explained with radiculopathy which might have resulted from extensive spinal involvement due to DCC. Thus, it was an interesting case of DCC, which itself was uncommon and it presented in a very unusual way which finally turned out to be DCC.

References

1. Bhalla A, Sood A, Sachdev A *et al*. Disseminated cysticercosis: a case report and review of the literature. *J Med Case Reports* 2008; 137.
2. Jain BK, Sankhe SS, Agrawal MD *et al*. Disseminated cysticercosis with pulmonary and cardiac involvement. *Ind J Radiol Imaging* 2010; 20: 310-13.
3. Kumar A, Bhagwani DK, Sharma RK *et al*. Disseminated cysticercosis. *Indian Pediatr* 1996; 33: 337-9.
4. Khandpur S, Kothiwala S, Basnet B *et al*. Extensive disseminated cysticercosis. *Indian J Dermatol Venereol Leprol* 2014; 80 (2): 137.
5. O'Mahony J, Shroff M, Banwell B. Mimics and rare presentations of paediatric demyelination. *Neuroimaging Clin N Am* 2013; 23 (2): 321-36.
6. Banu A, Veena N. A rare case of disseminated cysticercosis: Case report and review of literature. *Indian J Med Microbiol* 2011; 29: 180-3.
7. El-Beshbishi SN, Ahmed NN, Mostafa SH *et al*. Parasitic infections and myositis. *Parasitol Res* 2012; 110: 1-18.
8. Pérez-Jacoiste Asín MA, Calleja-Castaño P, Hilario A. Lumbosacral Radiculopathy as the Clinical Presentation of Neurocysticercosis. *Am J Trop Med Hygiene* 2020; 102 (6): 1166-7.
9. Del Brutto OH, Rajshekhar V, White AC *et al*. Proposed diagnostic criteria for neurocysticercosis. *Neurology* 2001; 57: 177-83.
10. Park SY, Kong MH, Kim JH *et al*. Disseminated cysticercosis. *J Korean Neurosurg Soc* 2011; 49 (3): 190-93.

CASE REPORT

JIACM 2021; 22(3-4): 136-38

A Case of Hairy Cell Leukaemia Associated with Miliary Tuberculosis

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Abstract

A case of disseminated tuberculosis presented with fever, cough, and severe anaemia. Patient was initially diagnosed to have miliary tuberculosis based on radiological findings and bone marrow examination. Miliary tuberculosis is not a rare presentation of tuberculosis in India. Haematological malignancies and miliary tuberculosis may have many common presenting clinical symptoms, which may lead to masking of the underlying haematological malignancy. We hereby report a case of miliary tuberculosis which was later diagnosed to be a case of hairy cell leukaemia also.

Keywords: Anaemia, cladribine, pancytopenia, splenomegaly, tuberculosis, vemurafenib.

Introduction

Hairy cell leukaemia (HCL) was reported in 1958 by Bouroncle and colleagues as an indolent malignancy associated with pancytopenia and splenomegaly that accounted for 2% of all leukaemias[1]. Hairy cell leukaemia is primarily a disease of middle-aged men. The age of the patients reported has ranged from the 20's to the 80's, but the average age is in the low 50's. The male: female ratio is approximately 4:1[2]. HCL, typically results in reduction in the production of normal red blood cells, platelets, mature granulocytes and monocytes. The increased production of malignant cells, along with a reduction in these mature elements, results in a variety of systemic consequences, including splenomegaly, anaemia, bleeding, and an increased risk of infection[2].

Miliary tuberculosis (TB) is a potentially fatal form of tuberculosis caused by the spread of *Mycobacterium tuberculosis* bacilli. Miliary TB can arise as a result of progressive primary infection or via reactivation of a latent focus with subsequent spread via the blood stream. Since its first description by John Jacob Manget in 1700, it is estimated that miliary tuberculosis accounts for 2% of all cases of tuberculosis in immunocompetent individuals and up to 20% of all extrapulmonary tuberculosis cases[3]. The clinical presentation of miliary tuberculosis is variable, the most common extrapulmonary sites include the lymphatic system, bones and joints, liver, central nervous system (CNS), and adrenal glands. Disseminated TB can give rise to striking haematological changes, so much so that a primary blood disorder is mistakenly diagnosed. The reported haematological abnormalities include anaemia, leucocytopenia, and pancytopenia. Various theories put forward to explain the haematological manifestations of TB include abnormal splenic function and direct invasion of bone marrow[4].

Case report

A 38-year-old male patient, resident of Uttar Pradesh, India, presented to us on 16th of March, 2021, with a history of fever of 2 months duration. Fever was associated with dry cough, breathing difficulty, night sweats, generalised weakness, and loss of appetite. Patient was also complaining of diffuse vague abdominal pain associated with fullness. He initially developed generalised body ache with fatigue, followed by fever and loss of appetite, which made him to lose 5 - 6 kgs of weight over a period of two months. He also complained of shortness of breath on walking a few hundred yards – which was quite unusual for him. Fever was of moderate grade (101° F - 102° F) and intermittent in nature. Patient was pyrexial on admission and was running a temperature of 101.4° F with BP of 100/80 mm of Hg, respiratory rate of 24 per minute and pulse rate of 116 per minute. Patient was fully conscious with GCS 15/15. On general examination, the patient appeared pale, slightly icteric and also was noted to have skin bruises; but there was no obvious lymphadenopathy. Patient was found to have koilonychia. Cardiovascular and neurological examination was unremarkable. On chest auscultation, bilateral coarse bi-basal rales were heard, but breath sounds were not diminished. On abdominal examination, abdomen was distended; on palpation, liver was palpable three fingers below the right costal margin, but spleen was significantly enlarged and palpable with the splenic edge extending more than 8 cm below the left costal margin, which was also confirmed with a dull note on percussion. On imaging,

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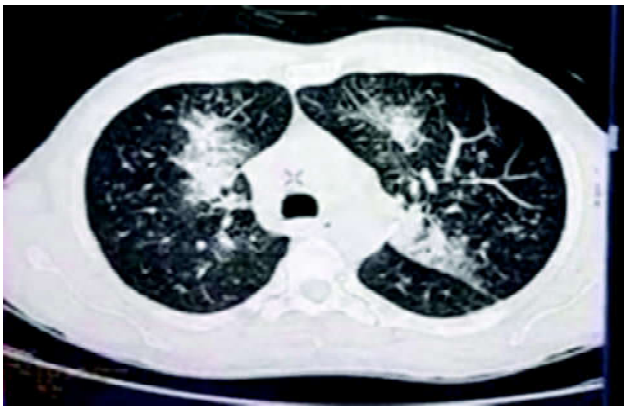

Fig. 1: *HRCT chest.*

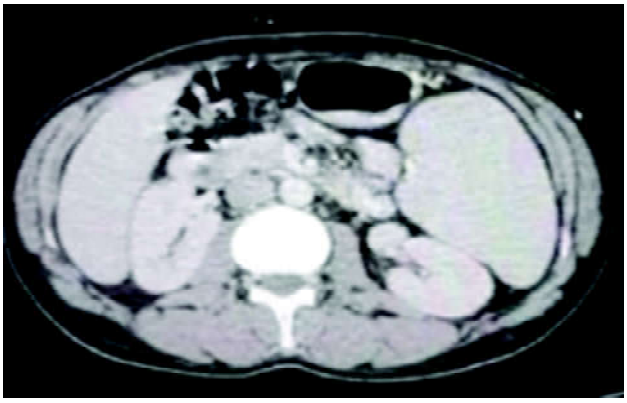

Fig. 2: *CE-CT abdomen.*

HRCT lung (in Fig. 1), showed multiple pretracheal, prevascular and carinal lymph nodes and also consolidation changes with miliary pattern, which are suggestive of thoracic tuberculosis. Abdominal CECT (Fig. 2) showed features suggestive of abdominal tuberculosis with splenomegaly with involvement of abdominal lymph nodes and ileal loops. Bone marrow aspiration/biopsy initially done at our hospital revealed pancytopenia with marked lymphocytosis and plasmacytosis and granulomatous inflammation (possibility of TB cannot be ruled-out). Lab parameters revealed ESR of 130, GBP (Table I) showed pancytopenia with macrocytic normochromic RBCs with lymphocytosis. Reticulocyte count was 0.30%, and his Vit B12 levels were in normal range, i.e., 391 pg/ml. Iron studies – Ferritin: 262 ng/ml, TIBC: 262 ug/dl, iron: 10 ug/dl. In consideration with the presenting clinical symptoms and bone marrow biopsy findings, along with the haematological and radiological reports, we diagnosed it as a disseminated tuberculosis with involvement of the bone marrow. Patient was initiated on anti-tuberculous therapy with low dose prednisone 20 mg OD in view of his bone marrow involvement. He was also transfused with 2 units of PRBC. The haematological abnormalities were only slightly reverted with a four-week course of anti-tuberculous treatment and low dose steroids. Patient remained apyrexial and haemodynamically stable at the end of the fourth week; hence his prednisone dose was tapered. However, in view of the COVID second surge, he was discharged on request with ATT medications prescribed on 19th April, 2021.

The patient was again reviewed in mid-May. We noticed that, his clinical condition was deteriorating further with recurrent episodes of fever. Unresponsive clinical condition with ongoing pancytopenia, even after six weeks of ATT, alerted us to contemplate a possibility of haematological malignancy. Hence, the patient was promptly referred to a haematologist for a repeat bone marrow aspiration/biopsy

Table I:

Inv.		16 March	18 March	19 March	20 March	21 March	24 March	25 March	28 March
CBC	Hb.	3.8	3.8	4		4.3	5	5.9	5.7
	TLC	0.48	0.36	0.37		0.26	0.43	0.52	0.3
	N/L	29/66	33/64	22/76		23/73	25/65	29/69	38/53
	RBC	1.16	1.07	1.29		1.45	1.63	1.95	1.38
	PCV	12.5	11.7	12.6		13.5	15.7	18.3	13.6
	MCV	107.8	109.3	97.7		95.2	96.3	93.8	98.6
	MCH	32.8	32.7	31		29.7	30.7	30.3	31.2
	MCHC	40.4	29.9	31.7		31.2	31.8	32.2	31.6
	PC	60	50	60		77	70	75	70
	ESR		150						
	ANC								
GBP			Pancytopenia: macrocytic normochromic RBCs, lymphocytosis						

and further expert opinion. He was reviewed by a haematologist and a repeat bone marrow aspiration/biopsy was done. The final impression of the repeat biopsy was hairy cell leukaemia and he was further advised to undergo Flow cytometery immuno-phenotyping and BRAF V600 Emutation testing. Comprehensive CLPD immuno-phenotype flow cytometry (25/5/21) concluded hairy cell leukaemia.

Discussion

Our patient was non-diabetic with no background co-morbidities. He was also screened for viral infections (HIV, Hepatitis B and Hepatitis C), which was found to be negative. Clinical manifestations of miliary TB are most likely to be subacute or chronic, the median duration of illness prior to clinical presentation was two months.The question of non-response to the primary treatment is clearly important and the alternative diagnosis should be reviewed with promptness. Hairy cell leukaemia (HCL), a rare and slow-progressive B-cell lymphoproliferative disease, enhances predisposition to infectious complications, especially to disseminated mycobacterial infections[6]. Most patients with HCL present with symptoms related to splenomegaly or cytopenias, (e.g., anaemia, thrombocytopenia, neutropenia, monocytopenia), including weakness and fatigue, infections of variable severity, and/or haemorrhagic findings such as gingival bleeding, ecchymoses, epistaxis[7]. Splenomegaly is frequently massive and may be the most prominent physical finding, mild hepatomegaly may be found in one-fifth of the patients. The most striking laboratory finding in about two-thirds of the patients with HCL is a moderate pancytopenia finding rarely present in patients with CLL[2] HCL is initially suspected in patients presenting with pancytopenia , splenomegaly without lymphadenopathy. Evaluation requires a bone marrow trephine biopsy and aspirate in conjunction with immunophenotyping and flow cytometry. Therapy is indicated only when the patient develops symptoms, significant cytopenias or symptomatic splenomegaly. Purine analogs, i.e., cladribine is the preferred initial treatment for most patients with symptomatic HCL and normal renal function[5]. Haematologist suggested chemotherapy with cladribine, a purine nucleoside analog. Our patient received five doses of cladribine (0.14 mg/kg = 8.5 mg), from 5/6/2021 to 10/6/21. He was reviewed in our OPD recently. Currently he remains apyrexial and his general condition is stable.

The BRAF inhibitor Vemurafenib is an investigational therapy for hairy cell leukaemia (HCL) with *BRAF* mutation[5]. In conclusion, hairy cell leukaemia, a predisposing clinical condition for disseminated tuberculosis.

Conclusion

In conclusion, our case elucidates that physicians must maintain a high index of suspicion when an immunocompetent or healthy young patient presents with a history of pancytopenia and prolonged fever. It is important to evaluate both the positive and negative symptoms at the same time when determining the treatment response.

Early diagnosis and timely referral of patients to a specialist is critical in preventing complications that can arise due to delayed treatment. Physician's awareness of symptoms and patient-physician communication is paramount to unravel the underlying unknown diagnosis that is contributing to the presenting primary infection.

References

1. Kreitman RJ, Arons E. *Clin Adv Hematol Oncol* 2018; 16: 205-15.
2. Golomb HM, Vardiman JM. Hairy cell leukaemia: Diagnosis and management. *CA: Cancer J Clin* 1978; 28: 265-77.
3. Dunphy L, Keating E, Parke T. Miliary tuberculosis in an immunocompetent male with a fatal outcome. *BMJ Case Rep* 2016; 2016: bcr2016216720.
4. Kashyap S, Puri DS, Bansal SK *et al*. Mycobacterium tuberculosis infection presenting as pancytopenia with hypocellular bone marrow. *J Assoc Physicians India* 1991; 39: 497-8.
5. Grever MR, Abdel-Wahab O, Andritsos LA *et al*. Consensus guidelines for the diagnosis and management of patients with classic hairy cell leukaemia. *Blood* 2017; 129: 553-60.
6. Arslan F, Batirel A, Ozer S *et al*. A predisposing clinical condition for disseminated tuberculosis: hairy cell leukaemia. *Mikrobiyol Bul* 2013; 47: 346-50.
7. Golomb HM, Catovsky D, Golde DW. Hairy cell leukaemia: a clinical review based on 71 cases. *Ann Intern Med* 1978; 89: 677-83.

CASE REPORT

JIACM 2021; 22(3-4): 139-43

Use of Steroids for Symptomatic Relief in Hepatitis A Virus-induced Cholestasis: A Case Series

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Abstract

***Aim:** We aimed to emphasize the potential role of steroids in hepatitis A virus infection (HAV) associated prolonged cholestatic jaundice in this case series.*

***Background:** Viral hepatitis is a significant healthcare burden in India. Hepatitis A and E virus infection are transmitted mainly through the faecal-oral route and are responsible for the epidemic and sporadic cases of acute viral hepatitis (AVH). In recent years changing trends in exposure to HAV are being observed with more cases being reported in adults than before. Though most of the cases with Hepatitis A resolve spontaneously, yet prolonged cholestasis had been reported in <1% of cases. Previous data had suggested the use of steroids in reducing cholestasis. In this case-series, we present our experience of steroids in HAV-associated prolonged cholestatic jaundice.*

***Results:** We had three young patients of HAV infection presenting with cholestatic features of more than 1 month duration. Inspite of all conventional therapies including ursodeoxycholic acid, L-ornithine L-aspartate, anti-histaminics, patients were not relieved of their symptoms. One patient had an episode of per rectal bleed secondary to coagulopathy as reflected by increased INR. After ruling-out other viral hepatitis (HBsAg, Anti HCV antibodies, IgM Anti HEV Ab for Hepatitis B,C and E respectively), all patients were given steroids (40 mg Prednisolone) till bilirubin levels fell <50% of its baseline levels and then steroids were rapidly tapered off. Patients had dramatic response to steroids with sharp fall in direct bilirunbin levels and marked improvement in cholestatic features within 5 - 10 days. All patients had complete resolution of their symptoms with normalisation of bilirubin levels on follow-up after 2 weeks.*

***Conclusion:** Steroids can be used for alleviation of HAV-induced prolonged cholestasis in selected patients after ruling-out other causes of viral hepatitis or reinfection with HAV.*

***Clinical significance:** Judicious use of steroids in prolonged cholestasis due to hepatitis A can be explored as a promising therapy for relief of cholestatic features.*

***Key words:** Hepatitis A, cholestasis, steroids, multidrug-resistance associated protein 2, case series.*

Background

Viral hepatitis is a significant healthcare burden in India. Approximately 400 million people all over the world have chronic hepatitis and the Asia-Pacific region constitutes the epicentre of this epidemic. It is equated as a threat comparable to the "big three" communicable diseases – Human immunodeficiency virus, malaria and tuberculosis[1]. Hepatitis A and E virus infection are transmitted mainly through the faecal-oral route and are responsible for the epidemic as well sporadic cases of acute viral hepatitis (AVH). Though HEV has been the leading cause of epidemics and sporadic acute and fulminant hepatitis among adults, in recent years changing trends in exposure to HAV are being increasingly reported[2].

Most cases of Hepatitis A resolve spontaneously with case fatality rate being approximately 0.3% in young adults. Prolonged cholestasis complicates <1% of cases and is associated with morbidity. Previous data suggests use of steroids for reducing cholestasis; however risks associated with the use of steroids hamper its use in patients. In this case-series, we present our experience of steroids in HAV-associated prolonged cholestatic jaundice.

Case 1

A 20-year-old male presented with complaints of gradually progressive yellowish discoloration of eyes for the past 2 months with decreased appetite for 1 month. He had clay-coloured stools for 1 month and intractable itching for the last 15 days. There was no history of blood transfusion/ surgery/dental exposure/high-risk behaviour/jaundice in past. There was no family history of prolonged jaundice. He was managed with tablet cetrizine, multivitamins, and

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ursodeoxycholic acid (UDCA) 300 mg tablets thrice daily with no symptomatic relief. No specific investigations relevant to the cause of jaundice were done. At presentation to our center, he was having complaints of significant itching and inability to sleep because of itching. Icterus was present with rest of the general physical examination being normal. On examination, there were no peripheral signs of chronic liver disease (CLD) and no evidence of ascites/hepatic encephalopathy (HE). *Per abdomen* examination revealed non-tender hepatomegaly (3 cm below right costal margin) with no other organomegaly. His liver function tests showed predominantly conjugated hyperbilirubinemia (Table I). As a protocol, we ordered serology for hepatitis viruses (Hepatitis A, B, C and E). Ultrasound abdomen showed hepatomegaly with no evidence of IHBRD. He was continued with the same supportive management with the addition of Calamine lotion, tab Hydroxyzine 25 mg at night and Cholestyramine sachets 4 g thrice daily before meals. His serology for Hepatitis A virus (IgM Anti HAV antibodies) came out to be positive (4.38 U/ml; normal <0.9 U/ml) with the rest of the viral markers being negative. The autoimmune profile (ANA, Anti LKM1, ASMA, IgG) was done and it turned out to be negative. Consequently, a diagnosis of prolonged cholestasis due to hepatitis A was made. The patient was admitted and planned for a challenge of steroids. Prior to that, LFTs were repeated and his SGOT and SGPT were <2 times the normal value which largely negated the possibility of any reinfection with hepatitis A virus/any other ongoing liver injury. His international normalised ratio (INR) was deranged with a value of 2.98. On second day, he had complaints of mild *per rectal* bleed. Consequently, parenteral Vitamin K and fresh frozen plasma (FFP) were given and INR normalised within 3 days with no fresh episodes of bleeding from any site. Later on, Prednisolone 40 mg was started in view of persistent cholestasis. On the 7th day of steroid treatment, the patient started having improvement in itching with a direct fraction of the bilirubin falling <50% of the level compared with that at time of admission (Table I, Fig. 1). The steroids were then rapidly tapered within the next 2 weeks (10 mg every 4th day and then on 12th day 5 mg for 3 days). He continued to have a persistent fall in direct bilirubin levels during this course. After 4 weeks of stoppage of steroids, the patient was asymptomatic with normal LFTs.

Case 2

A 15-year-old male presented with complaints of yellowish discoloration of eyes for the last 2 months, clay-coloured stools, and progressive itching for 1 month. There was no history of blood transfusion/surgery/jaundice/any prolonged jaundice in siblings. The itching was getting bothersome for the patient inspite of the treatment being given outside in the form of tablet UDCA, L ornithine L Aspartate (LOLA) sachets and tablet cetrizine. His Serology for Hepatitis B and C virus was negative. Chronic liver disease (CLD) was ruled-out on the basis of absence of peripheral signs suggestive of CLD, no evidence of ascites, encephalopathy, and USG imaging showing no signs of cirrhosis. On general physical and systemic examination, icterus was the only finding. LFTs showed hyperbilirubinaemia with a predominant conjugated fraction (Table II). The serology for Hepatitis A turned out to

Table I: LFTs in case 1 (before and after addition of steroids).

	16.11.18	09.12.18	09.01.19	Prednisolone Added	12.01.19	16.01.19	23.01.19	30.01.19	22.02.19
S. Bil (T/D)(mg/dl)	10.2/6.5	12.7/8.6	23.9/16.8		19.6/14.2	11.6/7.2	8/4.2	3.5/2	1.1/0.8
SGOT (U/L)	47	61	29		43	25	45	44	44
SGPT (U/L)	53	65	47		54	43	42	32	32
SAP (U/L)	112		271		224	165	134	154	
TP/Alb (g/dl)	7.2/4		7.2/4.6			7/4.2		7.2/4.2	

Table II: LFTs in case 2 (before and after addition of steroids).

	20.12.18	25.01.19	Prdnisolone Added	28.01.19	31.01.19	03.02.19	11.02.19	15.02.19	18.02.19
S. Bil (T/D)(mg/dl)	9.9/8.3	10.5/8.6		9/6.6	8/5.4	4/2.1	3/2	2/1.2	1.2/0.6
SGOT (U/L)	61	43		54	43	56	44	45	32
SGPT (U/L)	156	27		45	36	34	32	24	22
SAP (U/L)	589	576		485	345	372	274	270	302
TP/Alb (g/dl)	8.1/4.2	7.5/3.8		7.3/4		7/4			7/4.5

be positive (Anti HAV Ab: 10.09 U/ml vs Normal: < 0.9 U/ml). A diagnosis of prolonged cholestasis due to hepatitis A was made. He was started on a similar protocol of steroids as the first patient. The direct bilirubin decreased to < 50% of baseline level after 10 days of daily prednisolone 40 mg. Later on, the steroids were tapered off (10 mg every 3rd day) and the patient had normal bilirubin level after 25 days of therapy institution with no recurrence of symptoms at 3 months follow-up.

Case 3

An 18-year-male presented with progressively increasing yellowish discoloration of eyes for the last 1.5 months, and itching for 1 month. There was no history of clay-coloured stools/bleeding from any other site. There was no history of recurrent jaundice/family history of any prolonged jaundice in siblings/blood transfusion/surgery/dental exposure/high-risk behaviour. He was evaluated by a gastroenterologist at another hospital outside for jaundice and found to be IgM Anti HAV antibody positive. The rest of the viral markers were negative. He was being treated symptomatically for jaundice but the onset of itching and its gradual progression was worrisome for the patient. He was started on tablet UDCA 300 mg TDS , LOLA sachets 5 g twice daily, tab Hydroxyzine 25 mg twice daily with no relief. One week later, he was given cholestyramine sachet 4 gm thrice a day before meals and tab ondansetron 4 mg thrice daily with no significant improvement. His ultrasound abdomen showed mild hepatomegaly with a liver size of 17 cm and no other organomegaly/lymphadenopathy. There was no evidence of intrahepatic biliary radical dilatation (IHBRD). He came to us for no relief in his symptoms. The physical examination revealed icterus and scratch marks on extremities – likely due to itching. Systemic examination revealed no abnormality. The biochemical investigation showed persistently increased bilirubin levels with predominantly conjugated hyperbilirubinaemia. There was a decremental trend in aminotransferase levels with the last SGOT and SGPT being 56 and 23 U/L respectively (Table III). Having a good experience with judicious use of steroids in cholestatic hepatitis A virus and the patient profile being similar to the previous cases, Prednisolone was given in a dose of 40 mg and there was a drop of 75% of the direct bilirubin level within 5 days of therapy. Prednisone was tapered off 10 mg every 3 days in the next 10 - 12 days with normalisation of bilirubin levels at end of therapy. At 3 months' follow-up the patient was doing well with no complaints.

Discussion

HAV infection is frequently mild and asymptomatic in childhood. In developing countries, HAV infection is common during childhood, is often subclinical, and confers immunity to a large proportion of the population[3]. Therefore, HAV hepatitis usually occurs in children, and infection in adults is extremely infrequent.

In contrast, in the developed world, lack of exposure to HAV during childhood results in a large non-immune adult population. Due to better sanitation and personal hygiene practices being adopted in our country, adult cases with hepatitis A infection are infrequent nowadays. In adults, HAV infection has been reported to cause more severe liver disease such as cholestatic and relapsing hepatitis, which has a prolonged course[4,5]. Though the mortality due to HAV is extremely low (0.05% - 0.1%), associated intractable itching may be troublesome.

Initially, acute cholestatic hepatitis A was defined as clinical jaundice for at least 12 weeks, with a peak serum bilirubin greater than 10 mg/dl at a time when the serum aspartate aminotransferase level was rapidly declining[6]. However the criterion has been changed with elevated total bilirubin > 5 mg/dl more than 4 weeks as it is not advisable to wait until 12 weeks in view of increased morbidity[7]. Therefore, in the current scenario, any patient having hepatitis A infection and jaundice for more than four weeks, should raise the suspicion about prolonged cholestasis[8]. Classical signs and symptoms of prolonged cholestasis are pruritic skin, fatigue, weight loss, and loose, clay coloured stools. We had the three cases of prolonged cholestatic hepatitis A, one adolescent and 2 adult male patients. All these patients were receiving the best possible symptomatic treatment for cholestasis including UDCA, LOLA, Cetrizine, Hydroxyzine, Ondansetron with no/little relief. In addition,

Table III: LFTs in case 3 (before and after addition of steroids).

	20.01.19	22.02.19	08.03.19	Prednisolone Added	10.03.19	13.03.19	16.03.19	20.03.19	23.03.19
S. Bil (T/D)(mg/dl)	25.7/17.5	27.5/20.9	22.8/18.8		16/8.6	7.2/4.2	6.6/3.6	3.4/1.8	1.5/0.8
SGOT (U/L)	110	143	56		54	44	54	43	32
SGPT (U/L)	147	176	23		44	34	45	33	24
SAP (U/L)	287	205	218		216	143	137	207	187
TP/Alb (g/dl)	6.6/4.8	7.3/4			7/4.2			7/4	7.6/4.2

the other possible causes of cholestatic jaundice, viz., drug-induced and Primary Biliary Cirrhosis (PBC)/Primary Sclerosis Cholangitis (PSC) were also excluded. There was no history of any drug intake which might have caused the cholestasis. PBC was largely ruled-out as all 3 patients were male and age of onset of disease was early as compared to female preponderance and late onset disease (30 - 50 years) in PBC. Moreover, incidence of PBC is much less in our population as compared to the western world. PSC was ruled-out on basis of imaging showing no evidence of intra/extra hepatic biliary ductal dilatation.

In view of persistent symptoms, we planned to give a course of steroids as previously described by various authors[8-11]. Jain *et al*, in a series of 21 patients with prolonged cholestatic hepatitis A randomised eleven patients in ursodeoxycholic acid and prednisolone arm (Group A) vs Ten patients in ursodeoxycholic acid and placebo arm (Group B). Pruritus responded within a mean of 5 days (range 4 - 8 days) and 24 days (range 18 - 45 days) in group A and group B, respectively with anorexia and performance status improvement occurring early in group A. In addition, mean normalisation of serum bilirubin time was much less in group A patients (44 days) than in group B (94 days). The authors concluded that prednisolone resulted in symptomatic relief and a rapid initial drop in serum bilirubin levels followed by a persistent fall with adverse event[10]. Initially we were skeptical about the use of steroids in these cases due to lack of large series/studies available regarding its use and moreover with an inherent risk of flare of any underlying viral hepatitis virus, but we decided to give steroid challenge to these patients. All the three patients were give short course of steroids and they responded dramatically with normalisation of bilirubin and alleviation of all cholestatic features (Fig. 1-3).

Hepatitis A infection is a self-limiting viral illness with a clinical spectrum ranging from anicteric hepatitis, acute hepatitis, cholestatic jaundice lasting 10 weeks or more, Relapsing with 2 or more bouts of acute HAV infection occurring over a 6- to 10-week period to acute liver failure. Cholestasis has been reported in 0.4 - 0.8 % of cases[8]. In hepatitis A, cholestasis is presumed to occur because of the underlying inflammatory process. Endotoxin and pro-inflammatory cytokines like TNF alfa (TNF α) and IL-1 are released from liver and also as systemic response which inhibits mrp 2 (multidrug-resistance associated protein 2), one of the proteins having a role in bilirubin excretion[11]. Secondly, *in vitro* and animal studies on lymphocyte cultures of patients with alcoholic hepatitis and acute viral hepatitis have suggested that cellular or humoral immune phenomena might be involved in the pathogenesis for prolonged cholestasis[12]. The other proposed mechanism is an interruption in the continuity of bile flow secondary to periportal spotty necrosis[13]. The lympholytic action of corticosteroids may be the reason for their efficacy in these cases. In addition, alleviation of cholestasis by stimulating the alternate efflux pathway for bile salts has also been proposed as a possible mechanism.

Our findings add strength to existing data supporting the use of steroids in patients with HAV-related severe pruritus[8-11]. Since we did not estimate any molecular/cytological derangements described as potential mechanism of steroids so we are not in a position to comment about the exact underlying mechanism of these outcomes with steroids.

Conclusion

Prolonged cholestasis due to hepatitis A may be troublesome in a few patients. We suggest that a short course of steroids may be attempted in patients of HAV infection and associated cholestasis with following strategies:

1. Patients with prolonged cholestasis (total bilirubin > 5 mg/dl for more than 4 weeks).
2. Wait till 4 weeks is advised before starting of steroids as most of the patients recover from cholestasis by this time.
3. Prednisolone has to be instituted at a dose of 40 mg/d till direct bilirubin falls to < 50% of its baseline levels and then it should be tapered off over a period of 10 - 14 days, keeping a close watch on the serum bilirubin levels.
4. Steroid should only be advised in settings of no acute infections, after ruling-out other viral hepatitis infections (Hepatitis B, C and E) and reactivation of HAV (evidenced by nearly normal SGOT/SGPT).

References

1. Satsangi S, Chawla YK. Viral hepatitis: Indian scenario. *Med J Armed Forces India* 2016; 72 (3): 204-10.
2. Abraham P. Viral hepatitis in India. *Clinics Laboratory Med* 2012; 32 (2): 159-74.
3. Acharya SK, Batra Y, Bhatkal B *et al*. Seroepidemiology of hepatitis A virus infection among school children in Delhi and north Indian patients with chronic liver disease: Implications for HAV vaccination. *J Gastroenterol Hepatol* 2003; 18: 822-7.
4. Brown GR, Persley K. Hepatitis A epidemic in the elderly. *South Med J* 2002; 95: 826-33.
5. Lednar WM, Lemon SM, Kirkpatrick JW *et al*. Frequency of illness associated with epidemic hepatitis A virus infections in adults. *Am J Epidemiol* 1985; 122: 226-33.
6. Gordon SC, Reddy KR, Schiff L *et al*. Prolonged intrahepatic cholestasis secondary to acute hepatitis A. *Ann Intern Med* 1984; 101: 635-7.

7. Jung YM, Park SJ, Kim JS *et al*. A typical manifestations of hepatitis A infection: a prospective, multicentre study in Korea. *J Med Virol* 1982; 1318-26.
8. Darnindro N, Lesmana RA. Prolonged Cholestatic as a Typical Manifestation of Hepatitis A Infection: Diagnosis and Management. *Indonesian J Gastroenterolo Hepatolo Digestive Endoscopy* 2013; 14 (2): 120-5.
9. Kumar P, Bhatia V. Prolonged cholestasis due to hepatitis A virus Infection. *Indian Pediatr* 2011; 48: 485-6.
10. Jain P, Rai RR, Nijhawan S. Randomised placebo-controlled trial on the effect of corticosteroid on prolonged cholestasis due to acute hepatitis A. *Hung Med J* 2008; 2: 435-41.
11. Yoon EL, Yim HJ, Kim SY *et al*. Clinical courses after administration of oral corticosteroids in patients with severely colestatic acute hepatitis A; three cases. *Korean J Hepatol* 2010; 16: 329-33.
12. Marbet UA, Shefer S, Leevy CM. Intrahepatic cholestasis: modulation by immunological factors. *Schweiz Med Wochenschr* 1986; 116: 969-70.
13. Sciot R, Van Damme B, Desmet VJ. Cholestatic features in hepatitis A. *J Hepatol* 1986; 3: 172-81.

CASE REPORT

JIACM 2021; 22(3-4): 144-46

Necrotising Lymphadenitis in a Rare Overlap of SLE with Ankylosing Spondylitis

Somdatta Giri, Harpreet Singh**, Sunita Singh***, Gourab Bhaduri*, Gurjinder**

Key words: Systemic lupus erythematosus, Kikuchi's disease, Necrotising lymphadenitis.

Introduction

Lymphadenopathy results from reticulo-endothelial cell proliferation secondary to infection, autoimmunity, or malignancies. Getting a necrotising lymph node in a back ground of Disease Modifying Antirheumatic Drugs (DMARDs) therapy, tuberculosis is most likely. We are presenting a case report of a patient with a rare overlap of Ankylosing spondylitis (AS) and Systemic Lupus Erythematosus (SLE) presenting with a necrotising lymph node.

Case presentation

A 16-year-old female presented with fever for 3 months and persistent headache. Fever was moderate to high grade and was associated with anorexia and weight loss. She also gave a history of recurrent oral ulcers. On examination, she had bilateral cervical lymphadenopathy involving the posterior triangles, which were non tender, firm in consistency, and surface temperature was normal. Apart from lymphadenopathy, there was mild hepatosplenomegaly. There was no sternal tenderness or any bleeding manifestation. Her sensorium was normal and there was no neurological deficit. Fundus examination revealed grade 1 papilloedema.

In the past (nine years ago) she had been diagnosed as Ankylosing Spondylitis (AS) for her backache, bilateral sacroiliac joint involvement, positive HLA B 27 and family history. She was on Sulfasalazine for the same.

Her haemogram revealed leukopenia and thrombocytopenia. Her renal and liver function tests (including Prothrombin time) were normal. Her albumin was low and globulin was high suggesting a chronic inflammatory state. Inflammatory markers were found to be elevated (Erythrocyte sedimentation rate was 106 mm in first hour and ferritin was 3,455 ng/ml). But Procalcitonin and C-reactive protein was low. Mantoux test was negative and serology for HIV was non-reactive. Blood and urine cultures were sterile and Brucella serology was negative. Her chest X-ray was normal. A contrast-enhanced CT was done which revealed mediastinal, cervical, axillary, retroperitoneal and iliac lymphadenopathy with hepatosplenomegaly, but no focal lesion in the lung parenchyma, no fluid collection in serosa or bowel thickening (Fig. 1). Radiological diagnosis was lymphoma. MRI brain showed hyperintensity in bilateral thalamoganglionic regions, mesial temporal lobe and subtle cortical gyral hyperintensity with minimal post-contrast enhancement. CSF revealed mildly elevated protein (95 mg/dl), normal sugar and mild lymphocytosis pleocytosis (35 cells, 95% lymphocyte) and Viral panel and TB PCR were negative in CSF, thereby suggesting sterile meningitis.

A lymph node biopsy was done and she was put on intravenous antibiotics but there was no improvement in the fever pattern even after two weeks. The work-up for multisystem involvement revealed ANA to be elevated (1:1000, speckled pattern) with high titre of anti ds DNA, anti Smith antibody and low C3 and C4 level. Her urine examination was unremarkable and proteinuria was 120 mg/day. Based on clinical and immunological evidence, the patient fulfilled the 2012 SLICC criteria for the diagnosis of SLE. A diagnosis of SLE with high disease activity with

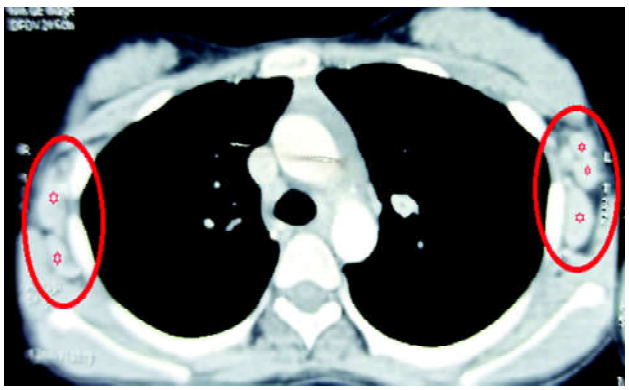

Fig. 1: *Contrast-enhanced CT showing heterogeneously enhancing bilateral axillary lympadenopathy. Largest LN 3.8 cm in the left axilla.*

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haematological, mucocutaneous, musculoskeletal and neurological involvement was made.

Biopsy report revealed necrotizing lymphadenitis with epithelioid histiocytosis with no architectural distortion (ruling-out lymphoma). And also the associated likelihood of Kikuchi's disease in a background of SLE needed to be looked in. Detailed review suggested foci of necrosis within the LN with numerous karyorrhexic bodies, fibrin deposits surrounded by Histiocytes which were CD 68 positive (Fig. 2) and lymphocytes predominantly CD 8 and few CD 4 cells, with absence of neutrophils and eosinophils. There was no caseation, no granuloma, nor any giant cell aggregation (thereby ruling-out TB). There was presence of Haematoxylin bodies (Fig. 3) and cells were negative for CD 30 marker ruling out KFD in a background of SLE (Fig. 4). Hence a final diagnosis of SLE lymphadenitis was made.

Accordingly, we gave her Methylprednisolone pulse therapy

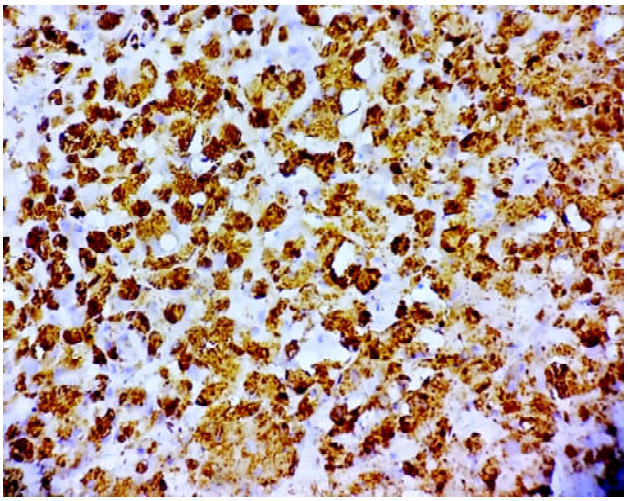

Fig. 2: *Cells stains with CD 68 marker suggesting presence of Histiocytes.*

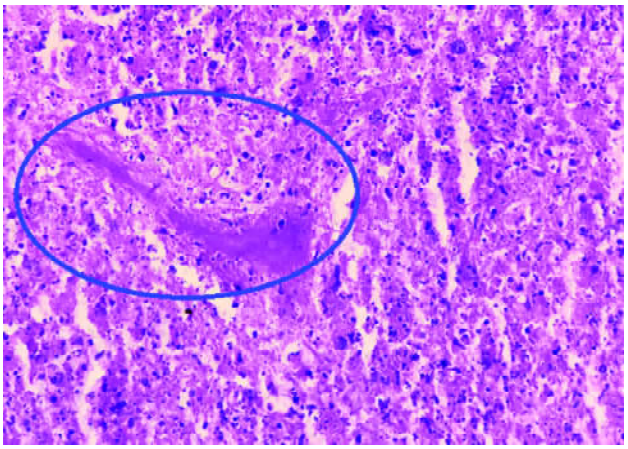

Fig. 3: *Showing high power view of LN biopsy. The encircled area marked above shows Haematoxylin bodies.*

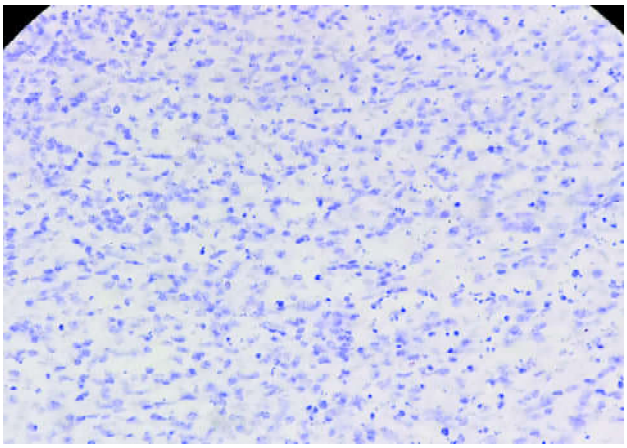

Fig. 4: *CD 30 marker is negative thus ruling-out KFD.*

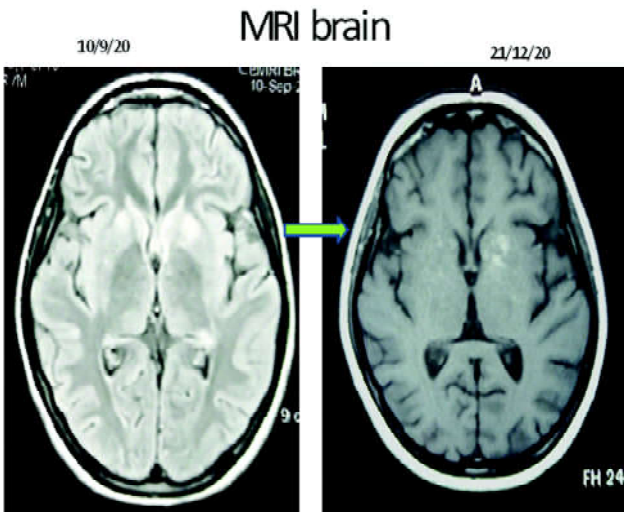

Fig. 5: *Showing comparison between the images of two MRI of brain at the level of Basal ganglia. The left side image (10.09.2020) is showing hyper-intensity in the ganglio-thalamic region bilaterally. The right side image (21.12.2020) is showing normal intensity with some residual changes in the left caudate nucleus.*

(1 g intravenous for 3 days) followed by oral prednisolone 1 mg/kg/d for one month and then tapered off. She dramatically responded to it with regression of the lymphadenopathy and fever. Her biochemical reports and MRI brain (Fig. 5) also showed marked improvement. Hydroxychloroquine was added. She showed no signs of relapse till reporting this case.

Discussion

Lupus lymphadenopathy has an estimated prevalence of 12 to 59%[1]. But, it rarely is an initial manifestation of SLE (5 - 7% at the onset). Usually it involves the cervical and axillary region. Rarely, mediastinal and retroperitoneal involvement may occur in a fulminant form of the disease. Lymph nodes (LNs) are soft, mobile, painful, and non-adherent to the

deep planes. Biopsy commonly shows reactive follicular hyperplasia which is a non-specific finding. Coagulative necrosis with haematoxylin bodies (condensed complexes of DNA and anti dsDNA antibodies) are typical findings in SLE, but it is rarely seen[2]. There is always an increased risk of lymphomas, especially non- Hodgkin's lymphoma in lupus patients that should be kept in mind while approaching such patients[3].

KFD is a rare entity, also termed as histiocytic necrotizing lymphadenitis. It is a disease of young people, predominantly females, of Asian descent. KFD generally presents with lymphadenopathy (localised – especially cervical, or generalised), organomegaly and constitutional symptoms. Diagnosis is made by LN biopsy. The disease is self-limiting. It usually takes 4 - 6 months. Treatment is of supportive care. However, corticosteroids are of benefit in severe or relapsing disease.

Review of the literature from 1991 onwards indicates that SLE predominantly predates or concurs with KFD[4]. An autoimmune origin with an infectious trigger of viruses has been suspected as aetiology of KFD. A strong autoimmune link with SLE has been implicated. A positive ANA has been found to be associated with a high chance of relapse. There is a significant overlap between pathomorphological features of KFD and lupus lymphadenitis. Pathognomonic feature of SLE being the presence of haematoxylin bodies that are condensed complexes of DNA and anti-dsDNA-antibodies, but it is very difficult to identify. Some of the studies documented that CD 30 cells are increased more in KFD than SLE that might also help them to separate[5]. Cramer *et al* in their report suggested that KFD may be a histopathologic alternative form of lupus lymphadenitis representing a '*forme fruste*' rather than being an independent disease entity[6]. Lupus lymphadenitis and KFD could in fact belong to the same entity. It is possible that the factors that induce lymph node proliferation are also responsible for the development of auto-antibodies. KFD should be a differential in SLE patients with necrotizing lymphadenopathy, due to its benignity and self limitedness.

In the present case, patient was on follow-up for AS and was on DMARDs – Sulfasalazine. In the light of necrotizing lymphadenopathy and background of DMARDs intake, secondary infection like tuberculosis was more a possibility. But during work-up, patients was found to have SLE (due to haematologlical, mucocutaneous, musculoskeletal, and neurologic involvement along with ANA positivity) as overlapping disease. The overlap of SLE with AS is very rare and only ten cases have been reported in the literature till date[7].

References

1. Cervera R, Khamashta MA, Font J *et al*. Systemic lupus erythematosus: clinical and immunologic patterns of disease expression in a cohort of 1,000 patients. *Medicine* 1993; 72: 113-24.
2. Behdadnia A, Allameh SF, Gharabaghi MA *et al*. Systemic Kikuchi-Fujimoto disease bordering lupus lymphadenitis: A fresh look? *Intractable Rare Dis Res* 2016; 5: 301-05.
3. Bernatsky S, Ramsey-Goldman R, Joseph L *et al*. Lymphoma risk in systemic lupus: effects of disease activity versus treatment. *Ann Rheum Dis* 2014; 73 (1): 138-42.
4. Kuo TT. Kikuchi's disease (histiocytic necrotizing lymphadenitis). A clinicopathologic study of 79 cases with an analysis of histologic subtypes, immunohistology and DNA ploidy. *Am J Surg Pathol* 1995; 19: 798-809.
5. Tabata T, Takata K, Miyata-Takata T *et al*. Characteristic Distribution Pattern of CD30-positive Cytotoxic T Cells Aids Diagnosis of Kikuchi-Fujimoto Disease. *Appl Immunohistochem Mol Morphol* 2018; 26 (4): 274-82.
6. Cramer J, Schmiedel S, Alegre NG *et al*. Necrotizing lymphadenitis: Kikuchi- Fujimoto disease alias lupus lymphadenitis? *Lupus* 2010; 19: 89-92.
7. Akbaryan M, Soltani Z. Overlap of Ankylosing Spondylitis and Systemic Lupus Erythematosus: A case report. *Int J Med Res Health Sci* 2016; 5 (4):100-103.

CASE REPORT

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COVID-19-Related Multisystem Inflammatory Syndrome in Adults: An Uncommon Case

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Abstract

Multisystem inflammatory syndrome in adults (MIS-A) is being recognised as a syndrome related to COVD-19 infection – more commonly in children, but occasionally in adults also.

A 28-year-old male with no co-morbidities presented with fever, abdominal pain, vomiting, chest and shoulder pains, and redness of eyes. He developed these symptoms 14 days post-COVID-19 vaccination. He was diagnosed with COVID-19 infection in September 2020. After excluding other causes for his clinical features, the diagnosis of MIS-A was made and IV immunoglobulin along with methylprednisolone was given. Patient responded to the therapy and remained symptom free on regular follow-up for 3 months.

We suggest that in a patient who presents with fever post-COVID-19 and/or post-immunisation with COVID-19 vaccination, the possibility of MIS-A should also be considered.

Key words: COVID-19, MIS-A, SARS-COV-2.

Introduction

COVID-19-related multisystem inflammatory syndrome (MIS) has been reported in children (MIS-C) and rarely in adults (MIS-A). Fever is generally the main finding of this syndrome and cardiovascular, gastrointestinal, haematological, and dermatological findings are prominent[1]. Involvement of lungs is rather uncommon in this entity. This is the only case of MIS-A at our hospital till date following the 1st and 2nd wave of COVID-19 infection. Our hospital is a designated COVID-19 (Level-3) hospital.

Centres for Disease Control and Prevention (CDC) gave the working case definition for MIS-A[2]. Here we report the case of a 28-year-old male who satisfied the case definition of MIS-A by the CDC (Centres for Disease Control), USA.

There is no set of guidelines available for the treatment of MIS-A and the options which have been tried are very limited. In many small studies including case reports, pulse therapy with intravenous (IV) corticosteroids are used with or without IV immunoglobulin (IVIG)[1]. Our patient responded to IV corticosteroid and IVIG treatment significantly.

Case report

A 28-year-old male patient was admitted to our hospital with the complaints of fever for 15 days, left upper abdominal pain for 15 days, and h/o redness over the chest and back with redness of eye 8 days back. He also complained of pain in chest over left side and left shoulder. The fever was of insidious onset, high-grade, 2 - 3 episodes per day which was relieved by taking paracetamol. During this period, he had occasional vomiting also.

There was no history of diarrhoea, shortness of breath, loss of consciousness, abnormal body movement, or bowel or bladder involvement. He had no co-morbidities like diabetes mellitus, hypertension, thyroid dysfunction. There was no history of any illicit drug use. Patient was diagnosed with mild COVID-19 (8 months before presentation), his 3 family members suffered from COVID-19 (3 months back) and he took 1st dose of a COVID-19 vaccine, 14 days before onset of symptoms.

On examination, the patient was conscious, alert, and oriented to time, place and person. His vitals included pulse rate of 108/min, blood pressure of 128/74 mm Hg, respiratory rate of 18/min, SpO2 of 97% on ambient air, and temperature of 102° F. Systemic examination was unremarkable.

On laboratory work-up as shown in Table I, it was found that the patient had significantly raised inflammatory markers like CRP, ESR, IL-6, D-dimer (mildly raised) before and at time of admission. ECG revealed T-wave inversion in leads II, III, aVF, V3, V4, V5 and V6 as shown in Fig. 1. The blood culture and urine culture showed no growth. Chest X-ray PA-view, HRCT thorax, CECT abdomen and 2-D echocardiography were within normal limits. Peripheral

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smear for malaria parasite, S. ANA and rheumatoid factor were negative. Urine (routine and microscopy), S. vitamin B-12, 25-OH Vitamin-D, S. amylase, S. lipase and thyroid profile were within normal limits. RT-PCR for COVID-19 was negative. Table I gives the details of other investigations.

This patient was diagnosed as a case of Multisystem Inflammatory Syndrome in Adults (MIS-A) according to CDC criteria[2]. He was managed with IV methylprednisolone (120 mg/day BD for 3 days), IVIG (2 gm/kg in 2 divided doses over 2 days), Inj. Ceftriaxone, tab. Ivabradine, tab. Apixaban, tab. Aspirin, multivitamin tablet, tab. Paracetamol and steroids (were tapered within 4 weeks). Patient's fever subsided within a day and his inflammatory makers decreased over the tab. aspirin, multivitamin tablet, tab. Paracetamol and next few days as shown in Table I. The ECG changes also reverted back to near-normal after 2 days as shown in Fig. 2. On day 10 of admission, the patient was discharged. On follow-up at 14 days, and 42 days post-discharge, the patient remained symptom-free, and his laboratory parameters were within limits as shown in Table I. Coronary angiography done on 10th August 2021 was within normal limits.

Table I: Serial investigations of the patient.

	19 May	25 May	27 May	04 Jun	08 Jun	09 Jun	10 Jun	11 Jun	13 Jun	24 Jun	27 Jun	29 Jun	20 July	05 Aug	10 Aug	Range	Units
Hb	14.9	14.6	12.8	13.6						14.6		14.3	15.2	14.1		13 - 17	g/dl
TLC	9800	7300	10300	8500						10100		9000	7900	5600		4000 - 11000	/mm^3
Platelets	283000	270000	313000	3250000						234000		165000	289000	238000		150-41000	/mm^3
ESR				22		60				22		17	5	8		0 - 15	mm/hr
CRP	21.18	64.35	21	25	102.5	96			12	2.58		24.21	1.01	1.72		< 5.0	mg/l
Ferritin			151.9		335	273			194	314			141.3			30 - 400	ng/ml
IL-6		4.58	30.4	26						2.5						< 7	pg/ml
D-Dimer	0.31	0.28	0.23		0.05	0.62			0.22	0.31			0.16			< 0.5	ug FEU/ml
Troponin T		14.04			110.5					13.68	18.64	18.64	19.09	22.44		< 14	pg/ml
Troponin I									0.32						0.03	< 0.02	ng/ml
S. LDH			245		267	447			264	211			199			120 - 246	U/L
Pro-Calcitonin						0.045				0.05			0.02			< 0.5	ng/ml
CPK-MB					21					2.7			11			0 - 16	U/L
NT-ProBNP				123						79			67			< 115	pg/ml
SGOP	33		20						21		24	21				< 50	U/L
SGPT	58	28	25						31	38		37	25	28.8		< 50	U/L
RET	WNL			WNL						WNL			WNL		WNL		
PT/INR			14/1.0														
Lipid profile												LDL = 147 triglyceride 292					
COVID-19	>400			22516						6546.2			3173.2				AU/ml
	SARSCoV-2 IgG antibody			anti-spike SARS CoV-2 IgG Ab						anti-spike SARS CoV-2 IgG Ab		anti-spike SARS CoV-2 IgG Ab				detects	
	< 12			< 50						< 50			< 50			range	
	CLIA			CMIA						CMIA			CMIA			method	
Mantoux			No induration														
Quantiferon Tb IGRA			Negative														
ECG	T wave inv		T wave inv		T wave inv		T wave inv		T wave inv		T wave inv		T wave inv		T wave inv		
			in aVF, II, III, V3, 4, 5, 6			in aVF, II, III, V3, 4, 5, 6			in aVF, III	in aVF, III	in aVF, III		in aVF, III		in aVF, III		
2-D ECHO		WNL		WNL			WNL					WNL			WNL		

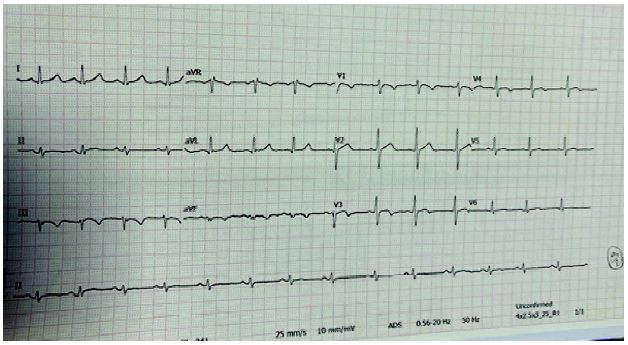

Fig. 1: *ECG on 09-06-2021.*

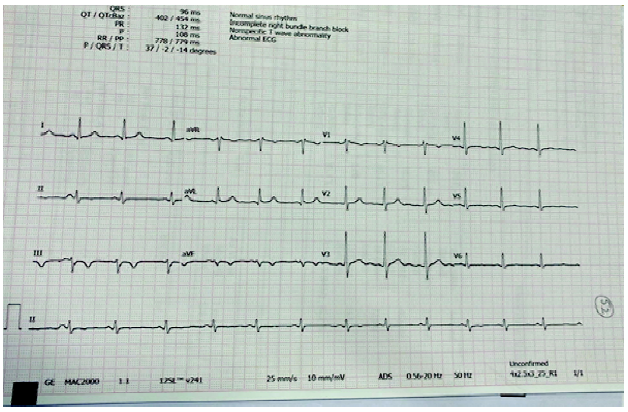

Fig. 2: *ECG on 11-06-2021.*

Discussion

The case definition for MIS-A as per CDC (Table II), includes a patient aged ≥ 21 years hospitalised for ≥ 24 hours, or with an illness resulting in death, who meets the clinical and laboratory criteria. The patient should not have a more likely alternative diagnosis for the illness[2]. In our patient, the clinical and laboratory criteria both were fulfilled including fever >38° C, myocarditis, rash, non-purulent conjunctivitis, abdominal pain, vomiting, elevated markers (like CRP, IL-6, ESR, procalcitonin) and antibodies against SARS-CoV-2 in high titres. The case series by Varadaraj *et al*[3] used morbidity and mortality weekly report[4] (MMWR) criteria for MIS-A but our patient satisfied the case definition of MIS-A as per CDC.

Our patient developed symptoms post-COVID-19 vaccination which could have been a triggering factor for the hyper-inflammation which is very similar to a case reported by Uwaydah *et al*[5]. They reported a 22-year-male who received inactivated SARS-CoV-2 vaccine, 6 weeks following a mild COVID-19 infection and developed MIS-A like symptoms. He responded well to steroids (IV dexamethasone 6 mg daily followed by oral steroids in tapering dose).

Table II: CDC case definition for MIS-A.

A patient aged ≥ 21 years hospitalised for ≥ 24 hours, or with an illness resulting in death, who meets the following clinical and laboratory criteria.

The patient should be not have a more likely alternative diagnosis for the illness (e.g., bacterial sepsis, exacerbation of a chronic medical condition).

I. Clinical criteria

Subjective fever or documented fever (≥ 38.0 C) for ≥ 24 hours prior to hospitalisation or within the first THREE days of hospitalisation* and at least THREE of the following clinical criteria occuring prior to hospitalisation or within the first THREE days of hospitalisation*. At least ONE must be a primary clinical criterion.

A. Primary clinical criteria

1. Severe cardiac illness includes myocarditis, pericarditis, coronary artery dilatation/aneurysm, or new-onset right or left ventricular dysfunction (LVEF < 50%), 2nd/3rd degree A-V block, or ventricular tachycardia.
2. Rash AND non-purulent conjunctivitis

B. Secondary clinical criteria

1. New onset neurologic signs and symptoms includes encephatopathy in a patient without prior cognitive impairment, seizures, meningeal signs, or peripheral neuropathy (including Guillain Barrè syndrome)
2. Shock or hypotension not attributable to medical therapy (e.g., sedation, renal replacement therapy)
3. Abdominal pain, vomiting, or diarrhoea
4. Thrombacytopenia (Platelet count <150,000/microliter)

II. Laboratory evidence

The presence of laboratory evidence of inflammation AND SARS-CoV-2 infection.

A. Elevated levels of at least TWO of the following: C-reative protein, ferritin, IL-6, erythrocyte dedimentation rate, procalcitonin

B. A positive SARS-CoV-2 test during the current illness by RT-PCR, serology, or antigen detection

Note: **These criteria must be met by the end of hospital day 3, where the date of hospital admission is hospital day 0.*

Although the exact underlying immunopathology is not well understood, adaptive immunity may be the cause responsible for these features[6]. In a study by Morris *et al*, it was believed to be a post-infectious syndrome rather than an infection in acute stage of development which causes direct endothelial damage and associated thrombo-inflammation[4]. In our case, whether the patient had a re-exposure to COVID-19 virus when his family members developed symptoms or the immunisation with COVID-19 vaccine acted as a trigger, or both these events could have led to a hyper-immune response over a period of time.

There is no exact treatment guideline for MIS-A, but most of the reviews, case series and case reports have mentioned the use of IV corticosteroids with or without the use of IV immunoglobulin along with supportive measures[1,4-8]. The treatment with a combination of IV corticosteroids and IVIG had a better response than either of them when given alone[1,4-8]. The IV

immunoglobulin acts at multiple levels and has an anti-inflammatory role[9]. The dose used in various cases ranged from 1 - 2 gm/kg in divided dose/single dose[4-8]. In this case, we used it in a dose of 2 gm/kg in two divided doses over 2 days along with IV methylprednisolone which was switched to oral prednisolone tapering over a 4-week course. The patient response was satisfactory and over 3 months of follow-up as shown in Table I, the patient remained symptom free and his inflammatory markers were also within normal limits as shown in Table I.

We suggest that in a patient who presents with fever post-COVID-19 and/or post-immunisation with COVID-19 vaccination, the possibility of MIS-A should be considered after ruling out other causes.

Abbreviations

COVID-19, Coronavirus disease 2019; MIS-A, Multisystem Inflammatory Syndrome in Adults; IV, intravenous; MIS-C, Multisystem Inflammatory Syndrome in Children; CDC, Centers for Disease Control and Prevention; OPD, out-patient department; CRP, C-Reactive protein; ESR, erythrocyte sedimentation rate; ECG, electrocardiogram; HRCT, High resolution computerised tomography; CBC, complete blood count.

References

1. Bastug A, Aslaner H, Bilir YA *et al*. Multiple system inflammatory syndrome associated with SARS CoV 2 infection in an adult and an adolescent. *Rheumatol Int* 2021; 41: 993-1008.
2. Centres for disease control and prevention. Multisystem Inflammatory Syndrome in Adults (MIS-A) Case Definition Information for Healthcare Providers. Accessed on 01 July 2021. Available from : https://www.cdc.gov/mis/mis-a/hcp.html.
3. Varadaraj G, Sangeetha B, Sandhu S *et al*. Four cases of Multisystem Inflammatory Syndrome in adults associated with SARS-COV-2 Infection – An overview of clinical features, diagnosis and treatment. *JAPI* 2021; 69: 24-7.
4. Morris SB, Schwartz NG, Patel P *et al*. Case series of multisystem inflammatory syndrome in adults associated with SARS-CoV-2 infection – United Kingdom and United States, March-August 2020. *Morb Mortal Wkly Rep* 2020; 69: 1450-6.
5. Uwaydah AK, Hassan NMM, Abu Ghoush MS *et al*. Adult multisystem inflammatory syndrome in a patient who recovered from COVID-19 postvaccination. *BMJ Case Rep* 2021; 14: e242060.
6. Kabeerdoss J, Pilania RK, Karkhele R *et al*. Severe COVID-19, multisystem inflammatory syndrome in children, and Kawasaki disease: immunological mechanisms, clinical manifestations and management. *Rheumatol Int* 2021; 41: 19-32.
7. Kofman AD, Sizemore EK, Detelich JF *et al*. A young adult with COVID-19 and multisystem inflammatory syndrome in children (MIS-C)-like illness: a case report. *BMC Infect Dis* 2020; 20: 716.
8. Ahmad F, Ahmed A, Rajendraprasad SS *et al*. Multisystem inflammatory syndrome in adults: A rare sequela of SARS-CoV-2 infection. *Int J Infect Dis* 2021; 108: 209-11.
9. Patil V, Kaveri SV. The mechanisms of action of IVIG in autoimmune and inûammatory diseases. *ISBT Science Series* 2013; 8: 185-8.

CASE REPORT

JIACM 2021; 22(3-4): 151-53

Brucellosis in a Patient with Ochronosis – A Rare Case

Poonam Ashok Kamath, Nandakrishna B**, Sudha Vidyasagar***, Cynthia Amrutha**, Muralidhar Varma****, Charan Thej Reddy**

Abstract

Alkaptonuria, an inborn error of metabolism, is due to deficient activity of homogentisic acid dioxygenase(HGD) enzyme. It is known to commonly present as spondyloarthritis, especially in the third decade. We discuss the case of a 49-year-old lady, who had presented with fever, backache, and multiple joint pains for 8 months, aggravated since the past 1 week. This case highlights the need to consider an additional diagnosis especially when there is no clinical improvement and the fever persists. Having a high index of clinical suspicion for a treatable cause like brucellosis is favourable, as it can lead to a better outcome.

Key words: Brucellosis, ochronosis, spondyloarthritis.

Introduction

Alkaptonuria is an autosomal recessive inborn error of metabolism characterised by a defect in the catabolic pathway of tyrosine, due to deficient activity of homogentisic acid dioxygenase (HGD) enzyme. This results in elevated levels of homogentisic acid (HGA), which upon polymerisation, forms a pigment which gets deposited in tissues, typically in the ear cartilage and sclera, leading to a condition called ochronosis. Affected patients are usually asymptomatic in childhood. There is also pigment deposition in large joints and spine, typically in the lumbosacral region. Calcification of multiple intervertebral discs is a characteristic radiological finding. Development of ochronotic arthritis and subsequent ankylosis results in limitation of the range of motion. The diagnosis is confirmed by quantitative measurement of HGA in urine and mutation analysis of the HGD gene. Tyrosine levels are normal.

Case description

A 49-year-old lady, no premorbid conditions, post-menopausal, presented to our outpatient department with complaints of fever, backache, and multiple joint pains for 8 months, aggravated since the past 1 week. She denied any scaly skin lesions and prolonged diarrhoea. However, she had exposure to cattle as she was a farmer. General physical examination revealed a hyperpigmented lesion on the lateral aspect of left sclera (Fig. 1), hyperpigmentation of left concha, and hyperpigmented macules on the left forearm and left shin (Fig. 2). Local examination revealed severe limitation in the range of movement of bilateral hip joints, knee joints and ankle joints. Straight leg raising (SLR) was positive. Figure of 4 test was positive bilaterally, suggesting a possible sacroiliitis. Rest of the systemic examination was unremarkable.

Complete haemogram revealed high ESR (95 mm/hr). X-ray showed loss of lumbar lordosis, intervertebral disc calcification, and decrease in intervertebral joint spaces, lumbar vertebral body lesions, and sacroiliitis (Fig. 3). MRI spine was confirmative of the same. Due to the comparatively young age of onset of the above-mentioned bone pathologies, and general hyperpigmentation, ochronosis was suspected. Homogentisic aciduria was measured, which came as positive (by qualitative testing). Urine sample turned black on exposure to silver nitrate (Fig. 4).

Presence of fever during admission pointed to a

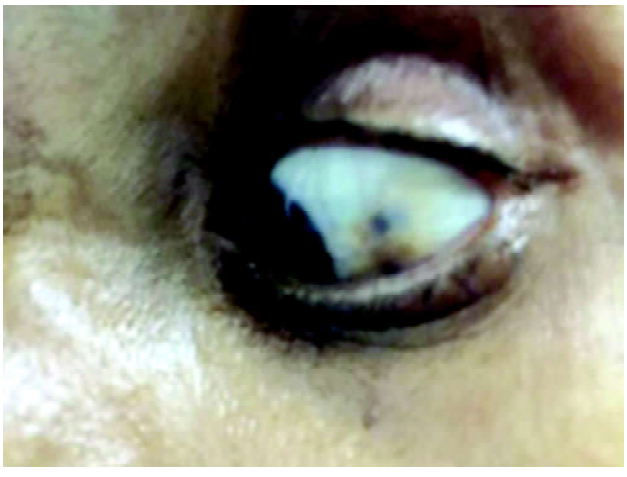

Fig. 1: *Hyperpigmented lesion in the left bulbar conjunctiva.*

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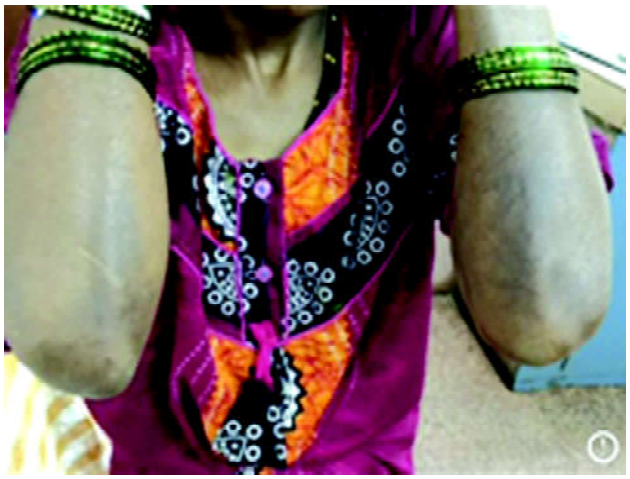


Fig. 2: *Figure showing forearm lesions.*

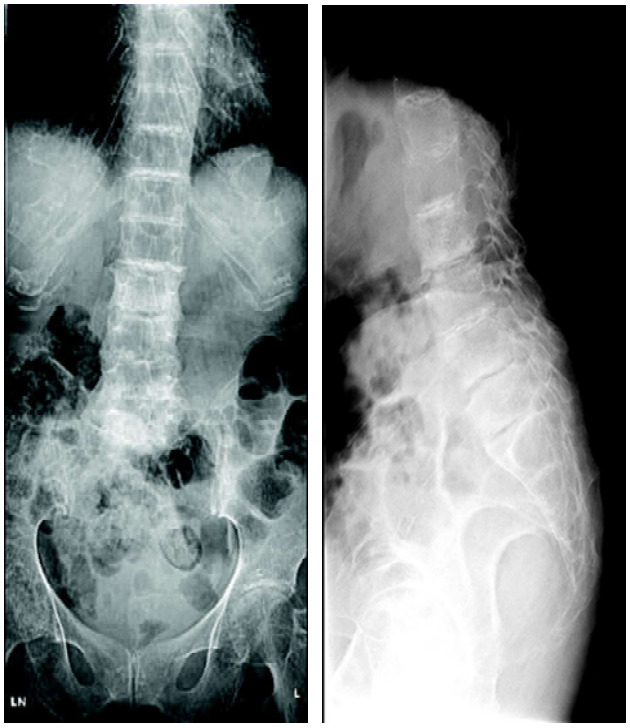


Fig 3: *X-rays showing calcification of intervertebral discs, reduced joint space, and reactive sclerosis.*

Fig. 4: *Change of colour of urine to black on exposure to silver nitrate.*

secondary cause, other than ochronosis. Her chest X-ray was normal and Mantoux test was negative – ruling-out the possibility of tuberculosis. Blood cultures were sterile. Echocardiogram ruled-out infective endocarditis. Brucella agglutination test was strongly positive (1:640) suggesting Brucellosis. Her clinical picture favoured coexistent Ochronosis with brucellosis. She was treated with a 6 week course of Rifampicin and Doxycycline after which her symptoms significantly improved, and she had no further fever.

For Ochronosis she was started on high-dose of oral vitamin C and N-acetylcysteine. Nitisinone could not be started due to financial constraints. Thereafter she came back for follow-up only after 1.5 years, at that time fever was absent, back ache had decreased, blood culture was sterile and Brucella agglutination titre had reduced.

Discussion

Axial spondyloarthritis is a potentially debilitating inflammatory arthritis of the spine, usually presenting as chronic back pain in the third decade of life[1]. It is often associated with one or more features like synovitis, enthesitis, dactylitis, and oligoarthritis. Non articular features include uveitis, psoriasis, and inflammatory bowel disease. Differential diagnoses to be considered are ankylosing spondylitis, psoriatic arthritis, and inflammatory bowel disease related arthropathy and reactive arthritis. Infective spondylodiscitis secondary to staphylococcus and tuberculosis should be borne in mind in such scenarios in the presence of fever and low back ache[2].

Ochronosis, an autosomal recessive disorder was first described by Virchow to denote a brownish-black pigmentation of connective tissue in patients with alkaptonuria[3]. The predominant deposition of homogentisic acid in cartilage (including the intervertebral discs and articular cartilage) causes collagen brittleness and consequent breakdown of the tissue, which in turn leads to spondylosis and large joint arthropathy. Though ochronosis is a rare entity, it must be suspected in classical presentations, like scleral and ear cartilage pigmentation. Characteristic findings on X-ray include articular space narrowing, osseous ankylosis, calcifications, osteophytosis, reactive sclerosis of the articular surfaces. Early identification is crucial, as delayed intervention after arthritis sets in, may not lead to resolution[4].

Clinically, there can be a significant overlap between ochronosis and brucellosis, especially regarding the presence of back pain. In patients with ochronosis, persistence of fever should encourage towards evaluating further, for another co-existing cause. Also, considering history of significant exposure to cattle, brucellosis is a

possibility that needs to be ruled-out. There is no approved treatment for alkaptonuria. Nitisinone has been studied and found to decrease the HGA levels by >95%[1,5]. But it is not beneficial in patients with well-established arthritis[5]. Dietary restriction of tyrosine and phenylalanine reduce HGA excretion, although the clinical effect is limited[5]. Ascorbic acid inhibits the conversion of HGA into polymers, but its efficacy has not been demonstrated for ochronosis[6].

Brucellosis, also known as undulant fever is a zoonotic infection caused by *Brucella spp* and is transmitted to humans from infected animals. It typically presents with insidious onset of fever, malaise, night sweats, and arthralgia. Osteoarticular disease can occur in up to 70% of patients. Other presentations include peripheral arthritis, sacroiliitis and spondylitis[7]. Other non-articular complications are genitourinary involvement, neurologic involvement (meningitis, encephalitis, brain abscess, radiculitis, neuritis), cardiovascular (endocarditis, myocarditis, pericarditis, endarteritis, thrombophlebitis, mycotic aneurysms), pulmonary involvement (bronchitis, interstitial pneumonitis, pleural effusion, empyema, hilar lymphadenopathy), hepatic or splenic abscesses, ocular involvement (uveitis, corneal ulcers, choroiditis, optic neuritis, papilloedema, endophthalmitis).

Hence, suspecting an additional diagnosis of brucellosis was beneficial in this patient, as it led to early diagnosis and prompt treatment with partial resolution of symptoms.

References

1. Suwannarat P *et al*. Use of nitisinone in patients with alkatonuria. *Metabolism Clinical and Experimental* 2005; 54: 719-28.
2. Medappil N, Adiga P. A 31-year-old female with fever and back pain. *J Emerg Trauma Shock* 2011; 4 (3): 385-8.
3. Bhattar PA, Zawar VP, Godse KV *et al*. Exogenous Ochronosis. *Indian J Dermatol* 2015; 60 (6): 537-43.
4. Drakoulakis E, Varvitsiotis D, Psarea G *et al*. Ochronotic arthropathy:diagnosis and management:a critical review. *Am J Orthop (Belle Mead NJ)* 2012; 41 (2): 80-83.
5. Milan *et al*. The effect of nitisinone on homogentisic acid and tyrosine: A two year survey of patients attending the National Alkaptonuria Centrex, Liverpool. *Annals of Clinical Biochemistry* 2017; 54 (3): 323-30.
6. Wolff *et al*. Effects of Ascorbic Acid in Alkaptonuria: Alterations in Benzoquinone Acetic acid and an Ontogenic Effect in Infancy. *Pediatric Research* 1989; 26 (2): 140-4.
7. Introne *et al*. A 3-year randomised therapeutic trial of nitisinone in alkaptonuria. *Molecular Genetics and Metabolism* 2011; 103 (4): 307-14.
8. Geyik MF *et al*. Musculoskeletal involvement of brucellosis in different age groups: a study of 195 cases. *Swiss Med Weekly* 2002; 132 (7-8): 98-105.

CASE REPORT

JIACM 2021; 22(3-4): 154-55

COVID-19 Vaccine-induced Skin Rash: A Case Study

Tulika Porwal, Anshul Agarwal***

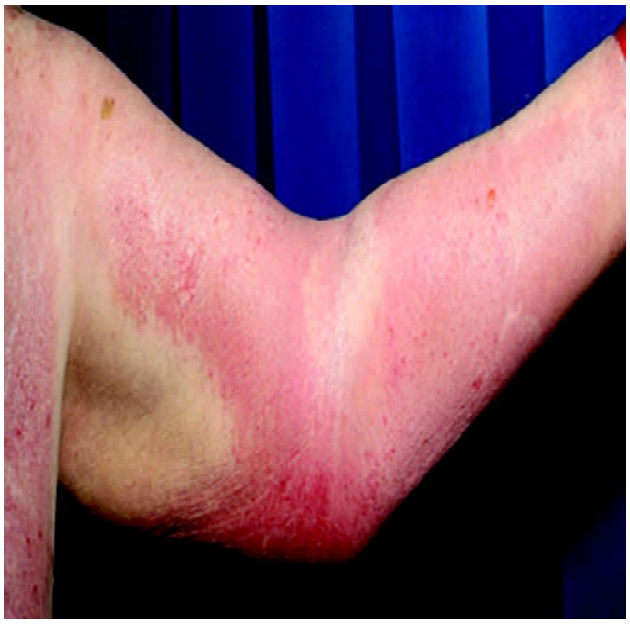
Abstract

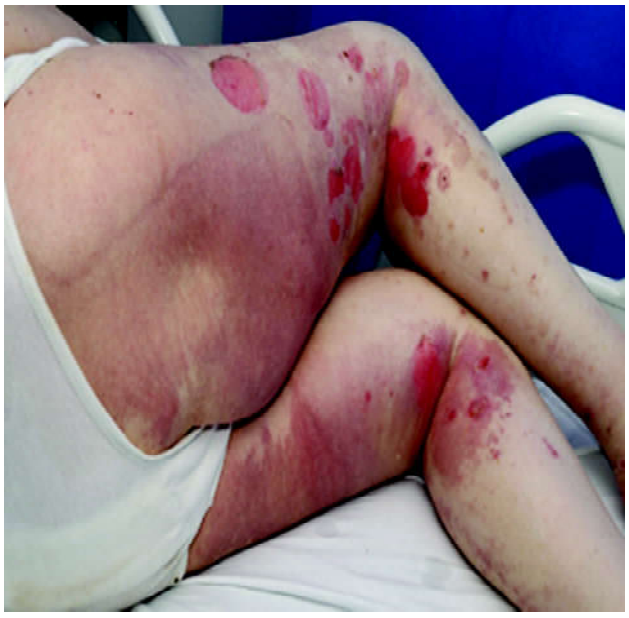
The past year saw a devastating effect from COVID-19, caused by a nRNA virus Coronavirus. This disease was declared as a global pandemic by WHO on 11th March 2020, since then has affected more than 175 million people and caused 3.8 million deaths worldwide[1]. There are several vaccines being manufactured to combat the virus causing COVID-19 throughout the world. The most widely used vaccines are the Moderna and Pfizer vaccines. While the vaccine is considered effective and safe, there have been a few side-effects reported with these vaccines. The most common side-effects reported can range from mild ones such as injection site irritation, to more severe and systemic side-effects such as venous clots. The causes of these skin reactions are attributed to both immediate and delayed hypersensitivity reactions, largely to the components of the vaccine[3]. Here, we report a case of COVID-19 vaccine induced erythematous skin rash with blisters (Pfizer-BioNTech).

Case

A 82-year-male with a background of prostatic cancer and peripheral neuropathy and no known drug allergies was admitted to the medicine emergency with rashes after 10 days of receiving the second dose of the Pfizer vaccine. (BNT162b2, Pfizer-BioNTech).

Initially it started with a maculopapular rash on the left arm and later spread to the chest, back, and limbs within 2 - 4 days. Oral and genital mucosa were not involved. On examination of the rash, it was very itchy and erythematous with inflamed plaques and blisters containing serosanguinous fluid. He had developed mild rash after the first dose of the vaccine but that had resolved within 2 - 3 days without any treatment. He did not have any fever, cough, arthralgia or sore throat. His blood showed a WBC count of 12,100 with mild eosinophilia 3.36 (0 - 0.4). Immunoglobulins, vasculitis profile, hepatitis screen, and HIV were negative. His complement levels including C3 and C4 were normal. A punch biopsy from the right thigh skin lesion revealed subepidermal bulla with prominent interstitial eosinophils and spongiosis. C3 staining was positive which was suggestive of bullous pemphigoid. He was treated with oral steroids starting with prednisolone 60 mg OD and chlorphenaramine. He demonstrated a significant improvement and was followed-up in the out-patient clinic.





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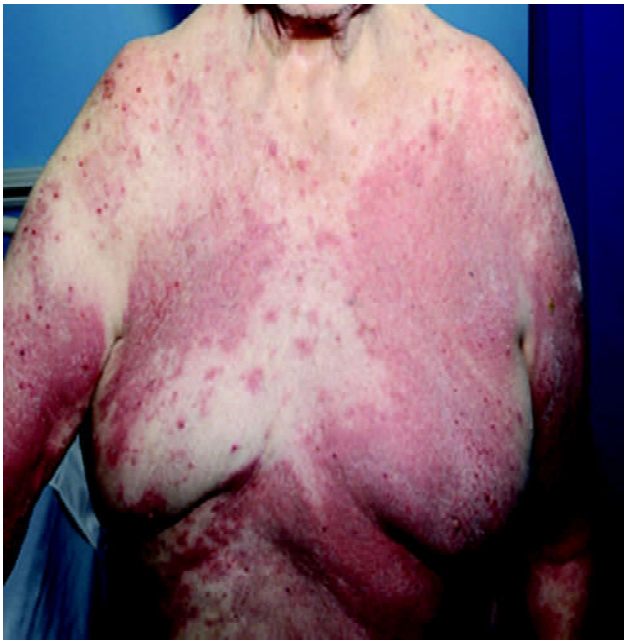

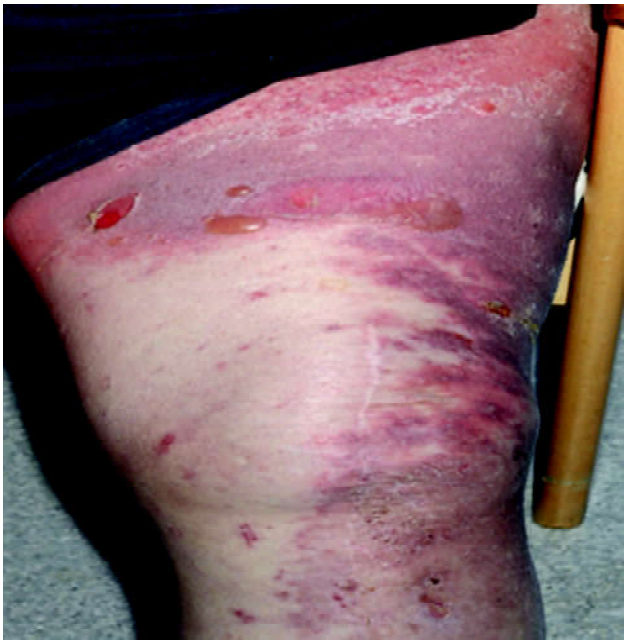

Fig. 1: *Patient's consent documented.*

Discussion

There are few reports of skin reactions with the COVID-19 Pfizer vaccine. The most common reactions are localised and generalised urticarial rash, and morbilliform rashes[3]. A few patients have reported rashes following both the doses of the vaccine, and a minority of them developed more generalised rash after receiving the second dose.

In this patient we observed a generalised erythematous rash with blisters developing after the second dose of Pfizer COVID-19 vaccine. The patient gave a history of milder skin reaction after the first dose. The patient did not develop other systemic reactions with the rash.

The presence of interstitial eosinophils and eosinophilia in blood suggests a delayed type hypersensitivity reaction. He responded promptly to systemic steroids and anti-histaminics.

References

1. Ritchie H, Ortiz-Ospina E, Beltekian D *et al*. (2020) - "Coronavirus Pandemic (COVID-19)". Published online at OurWorldInData.org. Retrieved from: 'https://ourworldindata.org/coronavirus' [Online Resource].
2. Features, Evaluation, and Treatment of Coronavirus (COVID-19) Marco Cascella; Michael Rajnik; Abdul Aleem; Scott C. Dulebohn; Raffaela Di Napoli.
3. McMahon DE, Amerson E, Rosenbach M *et al*. Freeman, Cutaneous reactions reported after Moderna and Pfizer COVID-19 vaccination: A registry-based study of 414 cases. *J Am Academy Dermatolo* 2021; 85 (1): 46-55. ISSN 0190-9622, https://doi.org/10.1016/j.jaad.2021.03.092. (https://www.sciencedirect.com/science/article/pii/S0190962221006587).

PICTORIAL CME

JIACM 2021; 22(3-4):156-58

Goldenhar Syndrome: A Rare Presentation to the Physician

Bijit Kumar Kundu, Rahul Sangwan***

Key words: External auditory canal atresia, Treacher-Collins syndrome, Pierre-Robin Syndrome, Crouzon syndrome, Goldenhar Syndrome.

An 18-year-old male delivered vaginally at term but with late cry after 5 minutes and delayed milestones, but presently fully independent in his daily activities of living, studying in high school with average academic performance, and normal social, motor, and language skills. He gave history of cleft lip operated at 1 year of age, branchial fistula operated at 3 years of age, cryptorchidism operated at 5 years of age, and left auditory canal atresia with reconstruction surgery at 12 years of age to create an external ear. He had two later siblings of which one sibling had died in infancy due to pneumonia. None of the siblings or any of his relatives had history of any similar affliction.

Clinical examination revealed facial asymmetry, low hair line, prognathism, no external auditory meatus on the right side, a reconstructed external ear on the left side, pre-auricular skin tags bilaterally, and genitalia underdeveloped for his age. X-ray of the cervical spine revealed synostosis of the upper 3 cervical vertebrae. Our patient had all the features of Goldenhar syndrome.

Few entities like Crouzon, Treacher-Collins, Pierre-Robins, and Goldenhar Syndromes have atresia of the external auditory canal as a common feature. Crouzon syndrome is inherited in an autosomal dominant pattern and is due to a

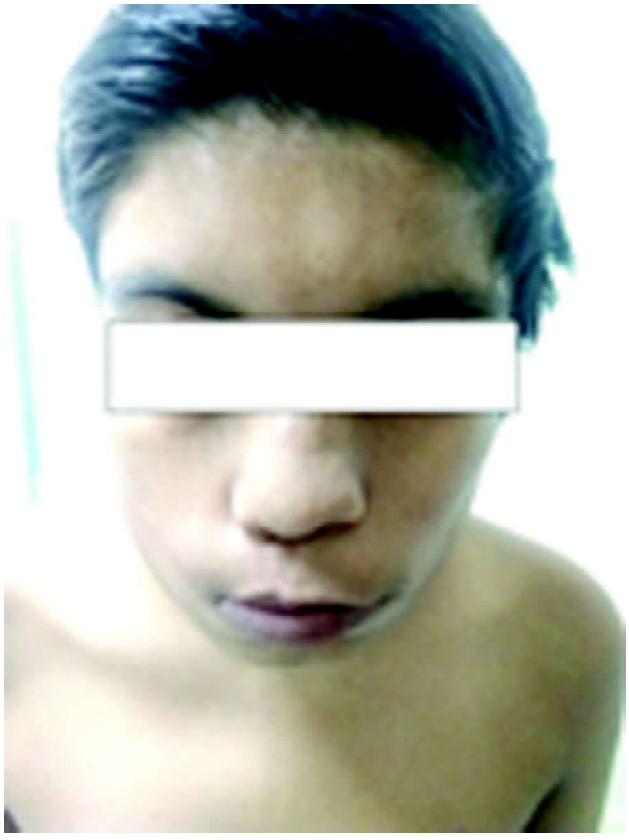

Fig. 1: *Asymmetry of the face.*

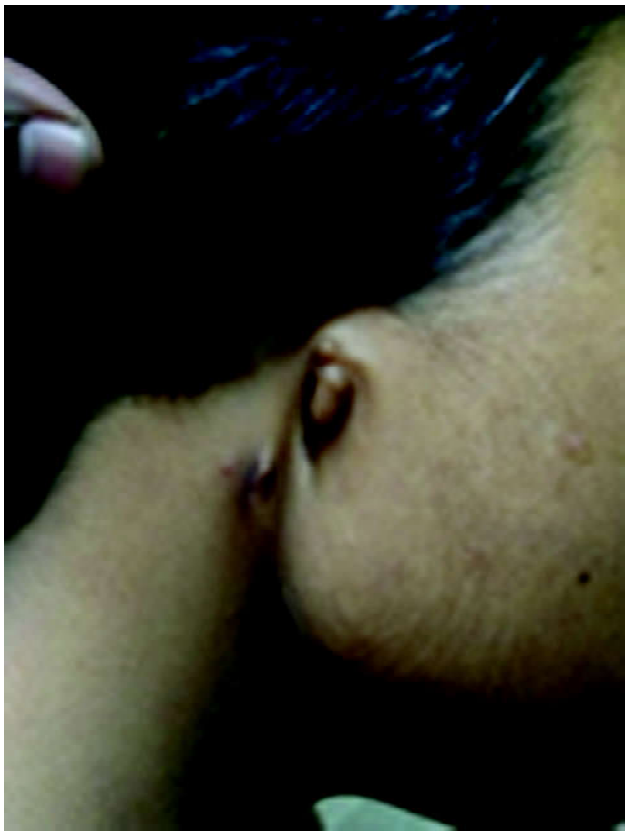

Fig. 2: *Absent external auditory meatus with skin tags.*

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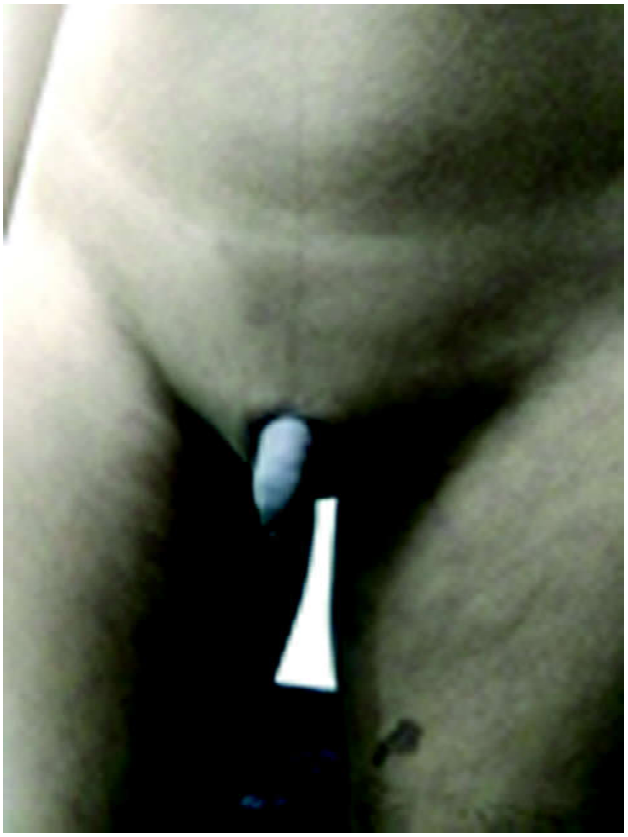

Fig. 3: *Under-developed genitals.*

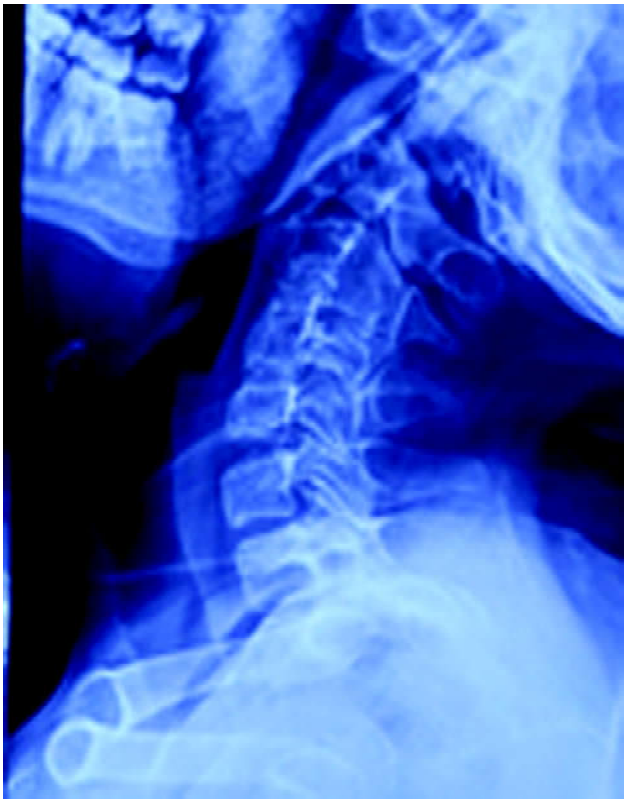

Fig. 4: *X-ray of cervical spine in Lateral view showing fusion of the 2nd, 3rd and 4th cervical vertebrae.*

mutation in fibroblast growth factor gone. It is characterised by premature craniosynostoses giving rise to cloverleaf skull. Exophthalmos, midfacial hypoplasia, Chiari I malformations (70% of cases), hydrocephalus, and stylohyoid ligament calcification (up to 50%) may also be present in patients[1].

Treacher-Collins syndrome, or mandibulofacial dysostosis, an autosomal dominant genetic abnormality, resulting from bilateral malformations of first and second branchial arches[2] due to mutations of the *TCOF1* gene, *POLR1C* and *POLR1D* [3], is characterised by dental and mandibular abnormalities including cleft palate, micrognathia, and zygomatic arch abnormalities. Patients have an obliterated nasofrontal angle. Spinal abnormalities are uncommon.

Pierre-Robin Syndrome (PRS) is the result of a sequence of events, and hence Robin Sequence should be the preferred term. The primary defect is abnormal development of the first pharyngeal arch causing a retrognathic mandible which prevents the tongue from descending which in turn prevents fusion of palatal shelves. Glossoptosis (retraction of tongue), high or U-shaped palate, micrognathia, and airway obstruction are the outcomes of this sequence. Polyhydramnios in mother is another result due to diminished swallowing in the foetus and its presence should prompt search for PRS[4].

Goldenhar syndrome, also called oculo-auriculo-vertebral spectrum (OAVS) is found in 1 in 3,000 - 5,000 newborns with a male to female ratio of 3:2. It is characterised by ear anomalies, commonest among which are pre-auricular appendages or skin tags, unilateral microphthalmia or anophthalmia, hemifacial microsomia, cleft palate, urogenital abnormalities, and asymmetry of skull and spinal anomalies[5,6]. Intelligence is usually normal. Radiological delineation of middle ear cavity volume, ossicles, inner ear structure, and course of internal carotid artery and facial nerve is important in surgical management of these cases[7].

References

1. Lowe LH, Booth TN, Joglar JM *et al*. Midface anomalies in children. *Radiographics* 2000; 20 (4): 907-22.
2. Johnson JM, Moonis G, Green GE *et al*. Syndromes of the first and second branchial arches, part 2: Syndromes. *Am J Neuroradiol* 2011; 32 (2): 230-7.
3. Dauwerse JG, Dixon J, Seland S *et al*. Mutations in genes encoding subunits of RNA polymerases I and III cause Treacher Collins syndrome. *Nat Genet* 2011; 43: 20-2.
4. Abramson ZR, Peacock ZS, Cohen HL *et al*. Radiology of cleft lip

and palate: Imaging for the prenatal period and throughout life. *Radiographics* 2015; 35 (7): 2053-63.

5. Kaissi AA, Chehida FB, Ganger R *et al*. Distinctive spine abnormalities in patients with Goldenhar syndrome: tomographic assessment. *Eur Spine J* 2015; 24: 594-9.
6. Tsirikos AI, McMaster MJ. Goldenhar-associated conditions (hemifacial microsomia) and congenital deformities of the spine. *Spine (Phila Pa 1976)* 2006; 31 (13): 400-7.
7. El-Feky M GF. https://radiopaedia.org/articles/external-auditory-canal-atresia [Internet]. Radiopaedia. [cited 2021 Jan 15]. Available from: https://radiopaedia.org/articles/external-auditory-canal-atresia?lang=gb.

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