Original Article

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Co-infection at the Time of Hospital Admission in Patients with COVID-19 Infection

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Abstract

Background: Co-infections with *Severe Acute Respiratory Syndrome Corona Virus 2 (SARS-CoV-2)* lead to unfavourable outcomes. However, data on prevalence of various co-infections in *SARS-CoV-2* positive patients on admission to the hospital is sparse. This study focusses on assessing co-infection rates and common pathogenic bacteria and fungus involved in these patients.

Methods: Patients admitted between April 2020 and December 2020 with Corona Virus Disease-2019 (COVID-19) were included. Criteria for co-infection using definitions were developed. All microbiological investigations of these patients performed within first 48 hours of admission were analysed. Their demographic characteristics along with existing co-morbidities, presenting symptoms, other laboratory findings on admission and clinical outcome were also reviewed.

Results: Of 1566 patients, 60% were males.64% belonged to 13-65 years age-group. 4% of COVID-19 positive patients were co-infected. 451 samples were received for culture of which urine were 66%, blood 23% and sputum 11%. 15% samples showed growth of which urine were 19%, blood 10% and sputum 2%. Bacteria isolated were 91% and fungus 9%. The common bacteria isolated were *E. coli* 46%, *Klebsiella pneumoniae* 25%, *Stenotrophomonas maltophila* 6% and *Pseudomonas aeruginosa* 4%. The common fungus isolated was Candida species 7%.10% of COVID-19 positive patients with co-infection expired and 90% recovered.

Conclusion: We report a 4% rate of bacterial and fungal co-infection in COVID-19 positive patients on admission mostly related to *Enterobacterales*, Non-fermenting Gram-negative bacilli and *Candida species*. This data can be valuable in optimising the use of antibiotics and antifungals in our patients. Similar studies on co-infection and its various aspects are the need of the hour.

Keywords: Bacteria, Co-infection, COVID-19, Fungus, SARS-COV-2

Introduction

After a clutch of peculiar viral pneumonia cases, later named as Coronavirus disease-2019 (COVID-19), was first identified in December 2019 in Wuhan city of China, the world has witnessed a viral blizzard of Coronavirus disease 2019 (COVID-19) cases. India has become the second hard-hit country by COVID-19 [1]. With uncountable deaths and a major blow to economy and mankind due to it, human race is still battling between hope and despair. Co-infection in *Severe Acute Respiratory Syndrome Corona Virus-2 (SARS-CoV-2)* cases with other microorganisms like virus, bacteria and fungi is a well-known entity and these can pose significant challenge to diagnosis, treatment and prognosis of COVID-19 along with increase in morbidity and mortality. In severe cases, there can be increased need for intensive care and antibiotic treatment [2].

The widespread use of antibiotics in such patients is daunting which subsequently leads to increased drug-resistance in bacteria [3]. Thus, co-infections act as fuel to the fire by adding to requirement of antibiotics or antifungals. As per CDC, co-infection is one occurring concurrently with the initial infection and such infections that occur within 48 hours of hospital admission are said to be community-acquired [3]. *S. aureus, S. pneumoniae, Gram negative bacilli, Legionella, Mycoplasma* and Chlamydia among the bacteria and *Candida species* and *Aspergillus species* amongst fungi are commonly encountered as cause of co-infections [1,2]. Along with coinfections of the respiratory tract, others like blood stream and urine infections are also a cause of concern [4].

The probable mechanisms involved in co-infection are damage caused to respiratory epithelium by the virus and their effects on innate and adaptive immunity thus

enhancing bacterial adherence, colonisation, growth and invasion into healthy sites in the respiratory tract. A suitable environment for co-infections is created by downregulation and differential regulation of immune genes [3]. However, data on coinfections in COVID-19 positive patients is scarce [5]. As per available data, incidence of co-infection is variable in COVID-19 positive patients, bacterial ranging from 8% to 47% and 7.3% viral co-infections [1,5].

We aimed to describe the rates of community-acquired co-infections in patients admitted with Covid-19 infection in our institute. To address the limitation of overdiagnosis of infection in colonised rather than infected patients and underdiagnosis, definitions for co-infections based on microbiological and clinical criteria were developed. The culture results of various samples were correlated with other laboratory findings along with demographic characters, existing comorbidities, presenting symptoms and clinical outcome in these patients.

Material and methods

This was a retrospective observational study including patients from all age groups and both genders that were positive for COVID-19 on admission to our 390 bedded institute. Such patients from April 2020 to December 2020 were included.

Covid-19 test of all patients on admission was done by Rapid Antigen Detection (RAT) test or Reverse Transcriptase Polymerase Chain Reaction (RT-PCR) [6]. Clinical as well as microbiological criteria were used to define various co-infections considered present at the time or within 48 hours of admission. Samples received for bacterial and fungal culture on or within 48 hours of admission of confirmed Covid-19 cases fulfilling these criteria were included and processed for bacterial and fungal culture as per clinician's request using standard operation procedure [7,8]. Identification and antibiotic susceptibility testing of isolates were done on Vitek 2 /Compact system.

Other laboratory investigations like demographic characteristics, existing co-morbidities, presenting symptoms, presence of lymphopenia, ferritin levels, presenting symptoms and clinical outcome were obtained from hospital information system. A patient was considered as co-infected when at least one of the performed cultures isolated a pathogen.

Sample inclusion Criteria: 1) All admitted or in-patient confirmed cases of COVID19 [A person with a positive Nucleic Acid Amplification Test (NAAT) or A person with a positive *SARS-CoV2* Antigen -RDT and meeting either the probable case definition or suspect criteria A or B]. 2) Samples meeting criteria of co-infection considered present at the time of admission. [Table 1].

Sample exclusion Criteria:

1) Samples received after 48 hours of patient admission
2) Samples not fulfilling the above criteria

Data analysis and Presentation: Demographic data and other details of patients were accessed from Hospital Information System. For comorbidities and presenting symptoms, electronic progress notes and general case notes summary were screened, and summary was prepared. Data was cross-tabulated across total COVID-19 cases and those with and without growth in culture tests. Values were expressed in absolute numbers as well as percentages of the group. Data obtained was compared and statistical significance calculated. P value of <0.05 was considered significant.

Ethical consideration: The study was approved by the Ethics committee of the institute.

Result

1566 patients were positive for SARS-CoV-2 on admission. 935 (60%) were males and 631 (40%) females. 23 (1%),995 (64%) and 548 (35%) patients belonged to <13 years,13-65 years and >65 years age group respectively. 654 (42%) were known cases of diabetes mellitus, hypertension 258 (17%), cardiovascular disease 75 (5%), bronchial asthma 32 (2%), hypothyroidism 31 (2%), chronic kidney disease 25 (1.5%), malignancy 19 (1%), Chronic Obstructive Pulmonary Disease (COPD) 6 (0.4%), pregnancy 6(0.4%) and steroid intake 5 (0.3%).

Fever as presenting symptom was seen in 556 (36%) patients.349 (22%) patients had history of contact with a case of SARS-CoV-2 and asymptomatic were 247(16%). Other common symptoms were body-ache in 210 (13%), shortness of breath 174 (11%), throat pain 150 (10%), cough 100 (6%), diarrhoea 51(3%) and loss of smell 24 (1.5%). Lymphopenia on admission was seen in 1264 (80%) of patients and serum ferritin was raised in 48 (3%) patients. 1427(91%) patients recovered while 138(9%) expired. Of the patients whose samples showed growth, 30 (45%) patients were males and 37(55%) were females. 28(42%) and 39(58%) patients were from 13-65 years and > 65 years age group. 36(54%) were known cases of diabetes mellitus, hypertension 15(22%), cardiovascular disease 6(9%),

Table 1: Criteria for co-infection considered present at the time of admission.

Sample	Sample criteria	Clinical criteria	Microbiologic criteria	Type of coinfection
Sputum	Presence of squamous Epithelial cells/lpf:10 or less	Temp > 38ºC or < 36^0C with WBC >=12,000 AND Producing purulent sputum AND Chest imaging (x-ray or CT) consistent with bacterial infection AND requiring supplemental oxygen	Growth of bacterial pathogen and or fungal mould or yeast	Bacterial and or fungal respiratory co-infection
Endotracheal aspirates	Presence of squamous Epithelial cells/lpf:10 or less OR no organisms visible seen under oil immersion.	Temp > 38ºC or < 36^0C with WBC >=12,000 AND Patients unable to produce sputum	Growth of bacterial pathogen or yeast with colony count > 10^5 or fungal mould	Bacterial and or fungal respiratory co-infection
Blood	Single blood culture (in case of pathogen) or multiple samples if normal flora or contaminant grown	Signs and symptoms suggestive of BSI.	Growth of bacterial pathogen and or fungal mould or yeast	Blood Stream Infection
Urine	Mid-stream urine with >10 WBC/hpf OR Sample collected from port in catheterized patients	Signs and symptoms of lower or upper UTI	Growth of bacterial pathogen with significant colony count. and or fungal mould or yeast	Urinary Tract Infection
Stool	Sample showing pus cells and or red blood cells	Symptoms suggestive of GI infections	Growth of Salmonella, Shigella or Vibrio species and or fungal mould or yeast	Gastrointestinal Infection
Other infections present at the time of admission but not included in the list.				

chronic kidney disease 4(6%), bronchial asthma 2(3%), hypothyroidism 2(3%), malignancy 1(1.5%) and history of steroid intake 1(1.5%). The symptoms in these patients were fever in 23(34%), throat pain 14 (21%), cough 13 (19%), shortness of breath 9(13%), body-ache 8 (12%), contact with COVID-19 positive patients 8 (12%), loose motions 4 (6%), running nose 4 (6%), asymptomatic 3(4%), headache 1(1.5%) and loss of smell 1(1.5%). Lymphopenia was seen in 66(99%) while raised serum ferritin levels in 9(13%). 60(90%) patients recovered while 7(10%) expired.

Risk factors like diabetes mellitus and bronchial asthma and symptoms like contact with positive case and throat pain were statistically significant (Table 2). A total of 451 samples were received for culture of which urine were 300 (66%), blood 103 (23%) and sputum 48 (11%). 67 (14%) samples from different patients showed growth of which 56 (19%) were urine, blood 10 (10%) and sputum 1 (2%). Bacteria isolated were 91% and fungus 9% (Table 3). *E. coli* were 31(46%), *Klebsiella pneumoniae* 17 (25.5%), *Stenotrophomonas maltophila* 4 (6%), *Pseudomonas aeruginosa* 3 (4.5%), *Klebsiella oxytoca* 2 (3%), *Enterococcus faecalis* 2 (3%), *Candida tropicalis* 2 (3%), *Methicillin Sensitive Staphylococcus Aureus* 1 (1.5%), *Achromobacter xylosoxidans* 1(1.5%), *Candida albicans* 1(1.5%), *Candida glabrata* 1(1.5%), *Candida auris* 1(1.5%) and *Trichosporon species* 1(1.5%) (Fig.1).67 out of 1566 patients had co-infection (4%).

Discussion

In the present study, co-infection rate in our institute was 4%. Karaba S et al reported 1.1% rate of bacterial respiratory co-infection. Non-respiratory co-infections were 5% and urinary tract co-infection was the commonest in their study [8]. This was comparable to our findings and

Table 2: Comparison of baseline demographics, co-morbidities, symptoms, laboratory findings and clinical outcome of COVID-19 positive patients included in the study and those with co-infection.

Characteristics	Total Number included in the study (%)	Number with co-infection (%)	P value
Number of patients positive for SARS-COV2 -19	1566	67(4)	
Gender			
No of Males	935 (60)	30(45)	0.011
No. of Females	631 (40)	37(55)	
Age group			
<13 years	23 (1)	0	0.000
13-65 years	995 (64)	28(42)	
>65 years	548 (35)	39(58)	
Number of patients with co-morbidities			
Diabetes mellitus	654 (42)	36(54)	0.042 *
Hypertension	258 (17)	15(22)	0.182
Cardiovascular disease	75 (5)	6(9)	0.103
Bronchial Asthma	32 (2)	2(3)	0.020 *
Hypothyrodism	31 (2)	2(3)	0.546
Chronic kidney disease	25 (1.5)	4(6)	0.354
Malignancy	19 (1)	1(1.5)	0.831
Chronic Obstructive Pulmonary Disease	6 (0.4)	0	0.133
Pregnancy	6 (0.4)	0	0.000
Steroid	5 (0.3)	1(1.5)	0.000
Number of patients with symptoms at presentation			
Fever	556 (36)	23(34)	0.837
In contact with a known COVID positive case	349 (22)	8 (12)	0.038 *
Asymptomatic	247 (16)	3(4)	0.010
Body-ache	210 (13)	8(12)	0.718
Shortness of breath	174 (11)	9(13)	0.537
Throat pain	150 (10)	14(21)	0.001*
Miscellaneous (stomach ache, vomiting, chest pain, disorientation, etc)	118 (8)	0	0.017
Cough	100 (6)	13(19)	0.000
Running nose(cold)	94 (6)	4(6)	0.991
Diarrhoea	51 (3)	4(6)	0.201
Headache	36 (2)	1(1.5)	0.653
Loss of smell	24 (1.5)	1(1.5)	0.978
Confusion	1	0	0.833
Number of patients with Laboratory findings			
Lymphopenia	1264 (80)	66(99)	0.000
Raised serum ferritin-Males >322, Females>291	48 (3)	9(13)	0.000
Number of patients whose samples were received for microbiological investigations			
Urine	300 (66)	56(19)	0.000
Blood cultures	103 (23)	10(10)	0.005*
Sputum	48 (11)	1(2)	0.445
Clinical outcome			
Number of patients recovered	1428(91)	60(90)	0.629
Number of patients died	138(9)	7(10)	0.629
***Indicates significant p value**			

Table 3: Results of microbiological investigations performed in patients included in the study

Sample	Number of samples received (%)	Number of samples with growth (%)	Organism isolated	No. of isolates
Urine	300(66)	56(19)	*E. coli* *Klebsiella pneumoniae* *Pseudomonas aeruginosa* *Candida tropicalis* *Candida albicans* *Candida auris* *Candida glabrata* *Enterococcus faecalis* *Trichosporon species*	30 17 2 2 1 1 1 1 1
Blood	103(23)	10 (10)	*Stenotrophomonas maltophila* *Klebsiella oxytoca* *Escherichia coli* *Enterococcus faecalis* *Methicillin Sensitive Staphylococcus Aureus (MSSA)* *Achromobacter xylosoxidans*	4 2 1 1 1 1
Sputum	48(11%)	1(2)	*Pseudomonas aeruginosa*	1
Total samples **Bacteria** **Fungus**	451	67 (15) 61 (91%) 6 (9%)		67

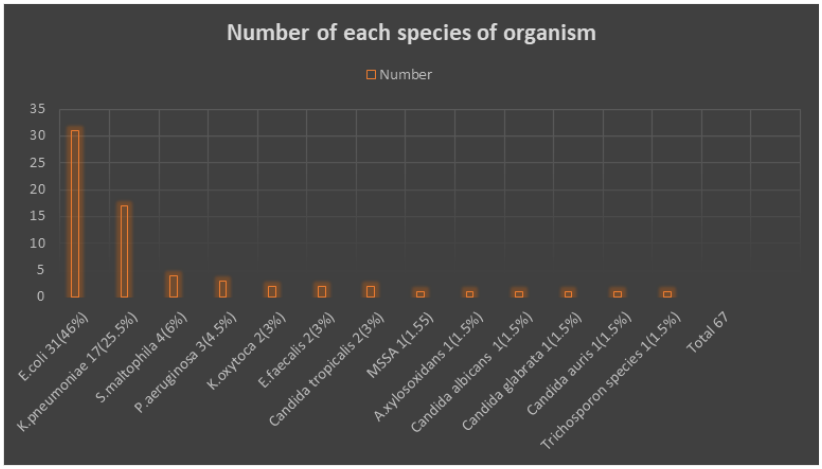

Figure 1: Number of each species of organism from all samples showing growth

urinary tract co-infection was the commonest in our population too. Vidal C et al found over-all co-infection rates to be 3.1% and bacterial pneumonia infection to be 2.1% [4] while Rawson T M et al reported it to be 8% [9] which was in accordance with our findings. It is noteworthy that bacterial pneumonia co-infections in patients hospitalised for COVID-19 was lower compared to those in other respiratory virus infections like influenza H1N1 or influenza H3N2.Aspergillosis as a complication of COVID-19 was not as frequent as that observed in patients with influenza [4] as seen in our study also. A 7.7 % bacterial co-infection rate was established by Zhang et al [10] Bacterial co-infection was found to be higher, 14% and 28% in patients admitted to intensive care unit as suppported by Jing R et al and Contou D et al [11,12].

According to Rothe K et al, bacterial and fungal co-infections are uncommon in COVID-19 patients and are mainly present in critically ill patients [5]. Patients that are severely ill are more likely to receive treatment with invasive catheters leading to increased susceptibility to secondary infections with multi-drug resistant pathogens[2].This is backed by Guin Zhang who observed that severely affected patients had a higher rate of bacterial

(25.5%) and fungal (10.9%) co-infections where-as patients with less severe disease had rates of co-infection of 0.8% and 0.6% respectively [13].This could be one of the reason of lower co-infection rates in our study as most of our patients were in the wards.

The rate of co-infection in COVID-19 patients is comparatively low probably due to some immunologic factor like macrophage hyper-activation and limited available studies [4,14]. However, in a study by K Shreenath,41% rate of co-infection was observed in respiratory samples from such patients. This is higher compared to our study as well as to other studies referred to above. Some of the factors responsible for this variation can be age group, comorbidities, disease severity, antibiotic exposure, detection method employed and variation in space and time [1]. Zhou et al also exhibited a contrastingly high rate of co-infections which was 50%[15].We observed an over-all lower rate of co-infection. This could be due to the method of testing which was culture and not Polymerase Chain Reaction (PCR). Viruses causing co-infection were not detected in our study. Most of our patients were administered an empirical antibiotic on admission. Application of a standard definition of various infections for sample inclusion could also be a reason. Number of sputum samples received by us were lesser due to lower number of patients presenting with cough.

Low rate of sputum culture positivity can be attributed to above reasons. The commonest organisms responsible for co-infections were *E. coli* followed by *K. pneumoniae* and *P. aeruginosa*. This finding was akin to that of K. Shreenath et al in which *Klebisella species* were one of the most commonly encountered bacteria among *Enterobacterales* causing co-infections [16-18]. Fontana C et al reported *Pseudomonas* as the commonest followed by *Enterobacterales* [19]. As per Karba et al, *E.coli* was one of the commonest blood pathogen and also the commonest organism causing urinary tract infection followed by *Proteus* and *Klebsiella species* [8].This was commensurable with our findings. Study by Chen at al showed *C. albicans* and *C. glabrata* among the common fungi causing co-infections in COVID-19 [20]. In *SARS-CoV* patients, Gram-negative bacilli and *Candida* are the most common types of bacteria and fungus [21] as was the case in our population too.

Conclusion

Rate of co-infection was 4 % in patients with COVID-19 infection on admission to our hospital which is low. The commonly involved bacteria were *E. coli* and *K. pneumoniae*. *Candida species* was the commonly involved fungus. This information will assist clinicians in judiciously choosing to use antibiotics which can be reserved for more serious cases. This will also be a step ahead in combating antimicrobial resistance.

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