

Assessing the mask-wearing habit as a contributing factor for COVID-19–associated mucormycosis

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ABSTRACT

Introduction: Association of mask-wearing habit and mucormycosis. **Context:** During the second wave of the COVID-19 pandemic, there was an absurd surge in cases of mucormycosis. COVID-19–associated mucormycosis (CAM) was found to be associated with the presence of diabetes, use of systemic steroids, prolonged use of masks, and others. The improper use of masks was proposed to be treacherous. **Methods and Material:** A case-control study was planned in which the patients coming to the dental OPD over a period of 6 months were asked to fill out a questionnaire validated by experts. **Results:** A total of 100 participants were included in the study. Out of 100 patients, 43 opted for surgical/N95 masks, whereas others were seen using different fabric mass. Mask-wearing habits are not a contributing factor for CAM. (P value > 0.005). **Conclusions:** Such studies give us an idea of the impact of hygiene habits on infectious diseases, further studies are required on a larger sample.

Keywords: COVID-19, mask, mucormycosis, N-95, SARS-COV-2, viruses

Introduction

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is a single-stranded RNA virus. It is characterized by mild-to-moderate respiratory illness to severe multiorgan dysfunction, hospitalization, secondary infection, and death depending on the dysregulated immune responses of the individuals.^[1] Mucormycosis is one such life-threatening infection seen in patients recovering from COVID-19 infections. Various theories have been given for the increased prevalence of mucormycosis in patients who are immunocompromised because of diabetic ketoacidosis, neutropenia, organ transplantation, increased serum levels of iron, or prolonged steroid therapy.^[2]

Multiple studies have demonstrated the protective role of masks in preventing respiratory viral illnesses in health care and household settings.^[3,4] When used for a long time, it serves as a breeding ground for a large number of microorganisms including some opportunistic fungi.

It was seen that improper usage of masks was prevalent among the local population of Haryana which might be a risk factor for the increased incidence of invasive fungal infections. Hence, the present study was designed to study if mask-wearing habits were a risk factor in COVID-19–associated mucormycosis.

Subjects and Methods

The present questionnaire-based study was conducted in the Department of Oral Medicine and Radiology. As per the Declaration of Helsinki, the ethical approval was

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Received: 23-05-2024

Revised: 10-07-2024

Accepted: 29-07-2024

Published: 09-12-2024

Access this article online

Quick Response Code:



Website:
<http://journals.lww.com/JFMPC>

DOI:
10.4103/jfmprc.jfmprc_887_24

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How to cite this article: Saini B, Gupta A, Bisla S, Kumia K, Shukla S, Yama K. Assessing the mask-wearing habit as a contributing factor for COVID-19–associated mucormycosis. J Family Med Prim Care 2024;13:5766-74.

obtained by the institute's ethical committee with application number – PGIDS/2021/OMR/535. This is a prospective study on rare disease undertaken in the largest health centre of Haryana state over a population of 2.5 cr; thus, the sample size was taken based on the number of COVID-19-associated mucormycosis patients reporting to the OPD calculation was done using the Cochran formula. Post-COVID-19 patients with/without mucormycosis (diagnosed on based of KOH mount/culture/histopathology) with positive RT-PCR report within 3 months were included in the study. A total of 100 patients with a history of COVID-19 were screened. A detailed case history of COVID-19, medical, dental, and drug was recorded for each patient. After satisfying the inclusion and exclusion criteria, patients were assigned accordingly to two groups – Group I (post-COVID-19 patients diagnosed with mucormycosis) and Group II (post-COVID-19 patients without mucormycosis). To conduct the study, a questionnaire was designed, which included 3 sections, each consisting of multiple-choice questions in the English language [Annexure 1]. For validation of the questionnaire, it was discussed by a group of 5 experts in the field and it was then translated into the Hindi language by the person whose first language was Hindi and back-translated to English by another investigator. The purpose of the study and the assurance of confidentiality were included at the beginning of the questionnaire along with a statement that proceeding to question section implies informed consent to participate. Responding to all the questions was mandatory. The estimated time to complete the questionnaire was 10 minutes.

Statistical analysis

The data were analyzed using the Statistical software SPSS version 20.0. Frequency (percentage) was used to describe the

summary statistics. *P* values <0.05 were considered statistically significant.

Results

A total of 100 patients were included in the study which were randomly allocated into two groups, case and control which were matched in terms of age and disease (diabetic/nondiabetic). Forty-three of the 100 patients chose to wear surgical or N95 masks, while other individuals were observed wearing other fabric masses [Graph 2]. In the general section of the questionnaire, the majority were seen using fabric masks in both groups (Case - 50% and control - 52%). The mask was worn for less than 6 hours by the majority of the population (34% and 44%) Patients were well aware of the usage of masks to cover the nose, mouth, and chin area and to inspect for any tears or holes before the use of the mask. The intergroup comparison of all the general questions was statistically nonsignificant ($P > 0.005$) [Tables 1 and 2].

In 43 patients wearing surgical or N95 masks, 68% of cases and 61.9% of control were seen reusing the mask at least 2–4 times. Most of the patients were discarding the used N95 mask with general household waste (case - 90% and control - 76%). Those who were reusing the mask cleaned it with soap and water followed by sun drying (54.5% and 28.6%), and the intergroup comparison for this section of the questionnaire was statistically nonsignificant ($P > 0.05$) [Table 3].

Of the 57 patients using fabric masks, 57.1% of cases and 34.5% of control were using self-sewn masks made up of two layers of fabric. Patients were cleaning the mask after every use (case - 67.9% and control - 55.2%) using soap and

Table 1: General questions included in the questionnaire

	Patients with mucormycosis (n=50)	Patient without mucormycosis (n=50)	<i>P</i>
Type of mask does you use?			
Cloth	25 (50.0%)	26 (52%)	0.682
Surgical mask	12 (24.0%)	8 (16%)	
N95 Mask	5 (10%)	9 (18%)	
Both Cloth & Surgical	4 (8%)	2 (4%)	
Surgical & N95 Mask	1 (2%)	3 (6%)	
N95 Mask & Cloth	2 (4%)	1 (2%)	
All of the above	1 (2%)	1 (2%)	
Are you sure that your mask covers both your nose, mouth and chin?			
Yes	43 (86.0%)	45 (90.0%)	0.538
Do you inspect the mask for tears or holes every time before wearing?			
Yes	36 (72.0%)	42 (84.0%)	0.148
When do you use mask?			
In crowded settings	2 (4.0%)	8 (16.0%)	0.035
Where you cannot be at least 1 meter from others.	1 (2.0%)	0 (0.0%)	
In rooms with poor or unknown ventilation	1 (2.0%)	1 (2.0%)	
In the market place	3 (6.0%)	0 (0.0%)	
All the above	34 (68%)	29 (58%)	
Other	9 (18%)	12 (24%)	

detergent (75% and 89%). However, the intergroup comparison was statistically nonsignificant (P value > 0.05) [Table 4].

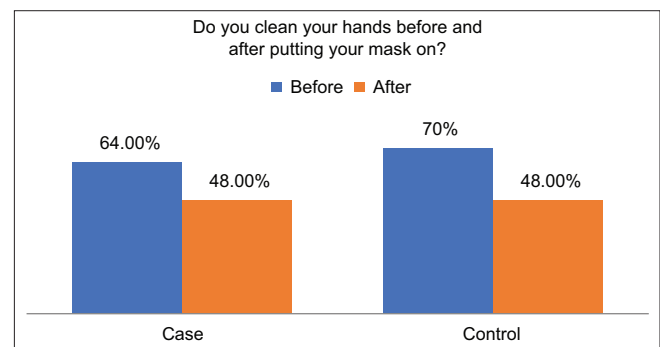
Discussion

Mucormycosis is an opportunistic infection known to have a seasonal and sporadic outburst. In India, there was a sudden gush of cases of fungal infection in COVID-19 infected patients.^[5] The proposed risk factors can be listed as – steroid use, poor glycaemic control, and immunocompromise. Spores of mucorales are ubiquitous in the environment, present in decayed organic matter. Although using masks contributed primarily to averting the COVID-19 infection, it has also been reported that masks might have acted as a risk factor for a surge in CAM. V.C. Keri *et al.*^[6] observed a high rate of fungal contamination in their pilot study emphasizing the need for better mask hygiene. Therefore, the present study looked at the various aspects of masking in patients who were infected with COVID-19 and subsequently developed mucormycosis.

According to the Centers for Disease Control and Prevention (CDC), proper hand hygiene is one of the most important practices in the reduction of the transmission of infection in the healthcare setting. Considering COVID-19 transmission spreads through direct, indirect (contaminated objects or surfaces), or close contact with infected people via mouth and nose secretions, washing hands with soap and running water is of critical importance.

Cowling BJ *et al.* in 2009 reported that hand hygiene with or without facemasks seemed to reduce influenza transmission, but when the observations were compared with the control group (Lifestyle education), the results were nonsignificant.^[7] In the present study, there was no significant difference reported between the cases and controls for hand sanitization, both before and after using the mask [see Graph 1].

Keri VC *et al.*^[6] did not report any significant association between the type of mask used by the patient and fungal contamination. Most of the patients in India used cloth masks, during the COVID-19 pandemic probably due to easy availability and low cost. In addition, they reused these masks, predisposing them to high chances for fungal contamination. Concerning the type of mask used, the present study reported



Graph 1: Hand hygiene habits before and after the use of a mask

Table 2: Usage of mask

Questions asked	Case n=50	Control n=50	P-Value
How long do you use your mask?			
12 h	15 (30.0%)	20 (40.0%)	0.096
12-6 h	15 (30.0%)	5 (10.0%)	
<6 h	17 (34.0%)	22 (44.0%)	
Occasionally	3 (6.0%)	3 (6.0%)	
How often you pull down your mask to your chin or take it off when speaking to other people?			
All the time	4 (8.0%)	4 (8.0%)	0.115
Very often	5 (10.0%)	8 (16.0%)	
Occasionally	21 (42.0%)	10 (20.0%)	
Never	20 (40%)	28 (56.0)	
Do you share the mask with others?			
No	50 (100.0%)	50 (100.0%)	a
How do you store your dry/new mask?			
Dry, breathable bag	4 (8.0%)	7 (14.0%)	0.385
Pocket or purse	11 (22.0%)	6 (12.0%)	
A plastic bag	14 (28.0%)	12 (24.0%)	
Your car/drawer/Almirah	19 (38.0%)	25 (50.0%)	
A + D	1 (2.0%)	0 (0.0%)	
C + D	1 (2.0%)	0 (0.0%)	
How do you store your wet/used mask?			
Sealed plastic bag until you can wash/discard it.	29 (58.0%)	25 (50.0%)	0.305
Pocket or purse	6 (12.0%)	4 (8.0%)	
Dry, breathable bag	2 (4.0%)	0 (0.0%)	
Car/drawer/Almirah	2 (4.0%)	2 (4.0%)	
Air dry	11 (22.0%)	19 (38.0%)	

a=No statistics are computed because q9 is a constant

nonsignificant results on the intergroup comparison. In both the study groups, a cloth mask was predominantly used followed by a surgical mask.

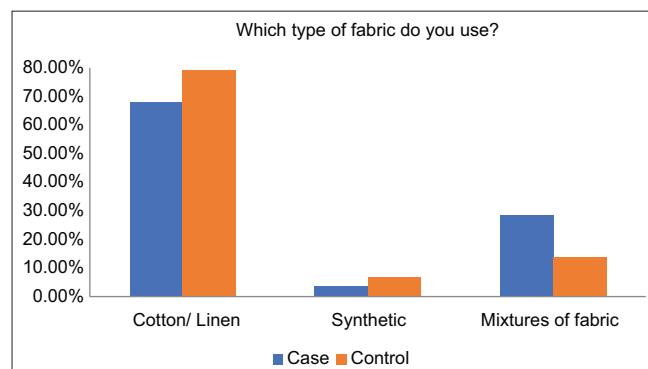
As advised by the World Health Organization (WHO), a face mask should cover the nose, mouth, and chin. The mask should be checked for any tears and holes. The majority study population in the present study was considerate about using the mask properly, and there was no difference reported between the two groups.

As recommended by WHO, face masks during COVID-19 should be worn in closed, crowded settings with poor ventilation, also indoors if one is not sure about ventilation then irrespective of distance from others. In the present study, 68% of cases and 58% of control had worn masks indoors and in crowded areas.

As stated by Dr. SN Chandan, prolonged use of face masks may lead to changes in the microbial flora of the respiratory tract and paranasal air sinuses, increasing the possibility of opportunistic fungal infections. In addition, it may lead to changes in the mucosal architecture or relative humidity of the paranasal sinuses, again raising the chances of fungal infections.^[8] A recent study reported higher odds of developing CAM in association with the prolonged use of cloth and surgical masks.^[9] In the present study, 60% population in the case and 50% in the control group were found to be using the face mask for more than 6 hours, although when compared, the difference among both the groups turned out as nonsignificant.

The mask should not be pulled down while speaking and should not be shared with others. No significant findings were observed regarding pulling down the mask while speaking. The study population was fully aware of not sharing their mask with others.

Regarding the storage of masks, it is recommended by CDC that wet or dirty masks should be stored in a sealed plastic bag until washed. Dry and clean masks can be stored in a breathable bag (paper or mesh fabric bag). Cloth masks should be washed at least once a day or as soon as they become wet or dirty. A washer and dryer can be used for this purpose. Disposable masks should be thrown away after they are worn once.^[10] In our study, 54% of the study population



Graph 2: Most commonly used fabric among the population of our study

Table 3: Usage of SURGICAL MASK/N-95 (n=43)

	Case - 22	Control - 21	P-value
Do you reuse your mask?			
Yes	15 (68.2%)	13 (61.9%)	0.906
No	6 (27.3%)	7 (33.3%)	
Sometime	1 (4.5%)	1 (4.8%)	
How many times do you use one mask?			
Once	0	1 (4.8%)	0.495
2-4 times	12 (54.5%)	12 (57.1%)	
Until it become humid/wet	2 (9.1%)	1 (4.8%)	
Until it gets dirty/wears off	2 (9.1%)	0	
How do you verify which side is the top?			
By metal strip	14 (63.6%)	18 (85.7%)	0.176
Does not matter	6 (27.3%)	3 (14.3%)	
Both sides are same	2 (9.1%)	0	
How do you discard your mask?			
With general household	20 (90.9%)	16 (76.2%)	
Separate plastic bag	2 (9.1%)	5 (23.8%)	
How do you clean your mask?			
Soap & water	12 (54.5%)	4 (19.0%)	0.073
Boiling in hot water	0	1 (4.8%)	
Did not clean	4 (18.2%)	9 (42.9%)	
How do you dry your mask?			
In sun	12 (54.5%)	6 (28.6%)	0.107
Air dry	1 (4.5%)	0	

Table 4: USAGE OF FABRIC MASK (n=57)

	Case-27	Control-30	P-value
Are you using a sewn mask?			
Yes	16 (57.1%)	10 (34.5%)	0.113
How many layers of fabric are there in your masks?			
Single layer	6 (21.4%)	1 (3.4%)	0.057
Two layers	10 (35.7%)	20 (69.0%)	
Three layers	3 (10.7%)	2 (6.9%)	
Do not know	7 (25.0%)	6 (20.7%)	
Do you Change your mask if it gets wet?			
Yes	22 (78.6%)	18 (62.1%)	0.248
How often do you clean your mask?			
After every use	19 (67.9%)	16 (55.2%)	0.406
After 2-3 days	7 (25.0%)	7 (24.1%)	
When it gets dirty	1 (3.6%)	5 (17.2%)	
Never	1 (3.6%)	1 (3.4%)	
How do you clean the mask?			
Soap/detergent	21 (75.0%)	26 (89.7%)	0.259
Boil in Hot water	0 (0.0%)	1 (3.4%)	
Sanitizer	2 (7.1%)	0 (0.0%)	
None of the above	2 (7.1%)	0	
A + D	2 (7.1%)	2 (6.9%)	
A + B	1 (3.6%)	0	
How do you dry your mask?			
Dried in the sun	9 (32.1%)	17 (59.6%)	0.085
Air dry	13 (46.4%)	11 (39.0%)	
Other	5 (21.6%)	0	

stored used masks properly in sealed plastic bags. New masks were stored in dry breathable bags by 12% study population. The intergroup comparison did not reveal any significant findings. Most of the patients who were using fabric masks in the present study used cotton or linen masks that were double-layered. As reported by one study, cotton, the most widely used material for cloth masks performs better at higher weave densities (*i.e.*, thread count) and can make a significant difference in filtration efficiencies.^[11] Most of the patients who were using fabric masks changed or washed their masks once they got wet or after every use. Intergroup comparison did not reveal any significant findings in the present study.

Out of 100 patients, 43 were found using surgical mask/N-95, but there was no significant difference between the case and control groups. CDC recommends that the number of donning for an N95 FFR should not be more than five per device. As reported by one study, fit performance is decreased over multiple, consecutive donning.^[12] Fifteen patients in the test group and 12 in the control group reused the surgical masks. Most of them used it 2–4 times. The top side of the mask should be verified by a metal strip. Most of the people using surgical/N-95 masks did the same in both groups.

According to CDC, the three most promising N95 mask disinfection methods are vaporous hydrogen peroxide, ultraviolet germicidal irradiation, and moist heat incubation. In the present study, it was found that most of the patients who cleaned their masks used soap and water. Out of 15 patients reusing masks in the case group, 12 were washing and sun drying their N95/surgical mask, whereas 16 patients in the control group were seen reusing their masks without washing them. The possible explanation for this is that with every wash, the integrity, fit, and effectiveness of the mask are lost; thus, these masks should be either discarded or sterilized. While hand washing the fabric masks, one should scrub them thoroughly with hot soapy water for at least 20 seconds. A detergent or bleach solution known to kill microbes can be used. After cleaning, they can be dried in the dryer machines or air-dried in direct sunlight. Ultraviolet rays of sunlight may help to kill microbes. In the present study, no significant difference was noted regarding the cleaning of fabric masks in both groups.^[13]

There was better perception and understanding of the questions as there was direct interaction with the patients. The control group was case-matched in terms of diseases. The small sample size and the questionable reliability of the answers, as patients in hospital setup try to fabricate the answers are the limitations of the study.

Conclusion

Possible contamination of the reused mask by fungal and bacterial pathogens is a matter of solicitude, and thus, proper mask hygiene is a must, and further studies with large sample size should be done.

Key messages

Mask-wearing habit whether it is made up of cloth or surgical/N95 is not the main causative factor leading to

mucormycosis; hence, it is important to rule out any debilitating conditions like diabetes or other comorbidities that could be predisposing the covid patients to this life-threatening condition.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

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Annexure I: Questionnaire

Q1. Do you clean your hands before you put your mask on?

- a) Yes b) No

Q2. Do you clean your hands after you take the mask off, and after you touch it at any time?

- a) Yes b) No

Q3. What type of mask do you use?

- a) Cloth mask
b) Surgical masks
c) N95 Mask

Q4. Are you sure that your mask covers both your nose, mouth and chin?

- a) Yes b) No

Q5. Do you inspect the mask for tears or holes everytime before wearing?

- a) Yes b) No

Q6. When do you use mask?

- a) In crowded settings
b) Where you can't be at least 1 metre from other
c) In rooms with poor or unknown ventilation
d) In the market place
e) all of the above

Q7. How long do you use your mask?

- a) 12 hours
b) 6-12 hours
c) < 6 hours
d) Occasionally

Q8. How often u pull down your mask to your chin or take it off when speaking to other people.

- a) All the time
- b) Very often
- c) Occasionally
- d) Never

Q9. Do you share the mask with others?

- a) Yes
- b) No
- c) Sometimes

Q10. How do you store your dry/new mask?

- a) In dry, breathable bag (like a paper or mesh fabric bag)
- b) In pocket or purse
- c) In a plastic bag
- d) In your car/drawer/ Almirah

Q11. How do u store your wet/used mask?

- a) In a sealed plastic bag until you can wash/discard it
- b) In pocket or purse
- c) In dry, breathable bag (like a paper or mesh fabric bag)
- d) In your car/drawer/ Almirah
- e) Air dry

SURGICAL MASK/N-95

Q1. Do you reuse your mask?

- a) Yes
- b) No
- c) Sometime/occasionally

Q2. How many times do you use one mask?

- a) Once
- b) 2-4 times
- c) Until it becomes humid/wet
- d) Until it gets dirty / wears off

Q3. How do you verify which side is the top?

- a) By the metal strip
- b) Does not matter
- c) Both sides are same

Q4. How do you discard your mask?

- a) In recycle bags
- b) Along with general household rubbish.
- c) In a separate plastic bag

Q5. How do u clean your mask?

- a) Using soap and water
- b) boiling in hot water
- c) Normal water
- d) using sanitizer
- e) none of the above

Q6. How do u dry your mask?

- a) Dried in the sun
- b) Ironed
- c) Air dry

For fabric mask

Q1. Which type of fabric do you use?

- a) Cotton/ Linen
- b) Synthetic
- c) mixtures of fabric
- d) Any of the above

Q2. Are you using a sewn mask?

- a) Yes
- b) No

Q3. How many layers of Fabric are there in your masks?

- a) single layer
- b) two-layer
- c) three layers
- d) not sure

