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## RESEARCH ARTICLE

# Impact of bioaerosol exposure on respiratory health of saw-mill workers

## Archana Sawane<sup>1</sup>, Manish Sawane<sup>2</sup>

<sup>1</sup>Department of Botany, Shri Mathuradas Mohota College of Science, Nagpur, Maharashtra, India, <sup>2</sup>Department of Physiology, NKP Salve Institute of Medical Sciences and Research Centre, Nagpur, Maharashtra, India

Correspondence to: Manish Sawane, E-mail: manishsawane@yahoo.co.in

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### **ABSTRACT**

Background: Occupational environments with organic dust and bioaerosols pose danger to the respiratory health of the personnel of such environments. Respiratory health of the workers of saw-mills in relation to airborne bacterial and fungal concentrations has rarely been studied in India. Aims and Objectives: This study was, therefore, designed to assess and compare the pulmonary functions (PFs) of saw-mill workers with those of matching controls. The study also aimed at assessment of bioaerosol exposure in saw-mills as compared to indoor air in a residential area as a control. Materials and Methods: The study design was cross-sectional case-control. PFs (percent predicted of forced vital capacity [FVC], forced expiratory volume 1 s/FVC ratio, peak expiratory flow rate, forced mid expiratory flow 25-75%, and maximum ventilatory volume) were assessed by computerized spirometer (Helios, Recorders and Medicare System, India) in 30 saw-mill workers with minimum exposure of more than 5 years. 30 age and sex matched controls were also evaluated for the same parameters. Bacterial and fungal concentration (CFU/m³) in the air of saw-mills with residential indoor air as a control was evaluated by volumetric air sampler (Hi-Air, Hi-Media, India). Appropriate statistical tests were used to compare respiratory symptoms, PFs, and bioaerosol levels. Results: Statistically significant lower values of PF parameters were observed in cases as compared to controls. Bacterial and Fungal concentrations in saw-mill air were significantly higher (P < 0.001) than control indoor air. Conclusion: The output of this study suggests a compromised respiratory status in workers of saw-mills with more exposure to bacteria and fungal spores. The results call for urgent measures to reduce environmental bioaerosol exposure in organic dust environments like saw-mills.

KEY WORDS: Bioaerosol; Organic Dust; Pulmonary Function Test; Saw-mill Workers

## INTRODUCTION

Bioaerosols may consist of pathogenic or non-pathogenic live or dead bacteria and fungi, viruses, high molecular weight allergens, bacterial endotoxins, mycotoxins, peptidoglycans,  $\beta$  (1 $\rightarrow$ 3)-glucans, pollen, plant fibers, etc., It is now appropriately recognized that exposures to biological agents

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in both the occupational and residential environment are associated with a wide range of adverse health effects with major public health impact, including contagious infectious diseases, acute toxic effects, allergies, and cancer.<sup>[1]</sup>

The pollution of air in saw-mills with microorganisms results from the primary or secondary infection of timber with bacteria and fungi, respectively. Secondary infection of wood proceeds on chopped wood chips and planks which are stored in sawmills in conditions favoring microbial growth. It is characterized by abundant growth of molds. Thus, sawmill workers may be exposed at work to the inhalation of various allergenic and immunotoxic agents, comprising wood derivatives and microorganisms associated with timber.<sup>[2]</sup> This exposure to bioaerosols in work environment

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presents hazards of allergy and respiratory symptoms. Pulmonary functions (PFs) in saw mill workers have been studied in India, however, correlation of respiratory symptoms and compromised PFs with bioaerosol exposure has not been explored. This study was, therefore, designed to assess and compare the respiratory symptoms and PFs of sawmill workers with those of matching controls in relation to bioaerosol exposure in sawmills.

## MATERIALS AND METHODS

The study design was cross-sectional case-control. PFs were assessed by portable computerized spirometer attached to a laptop (Helios, Recorders and Medicare Systems, Chandigarh, India) in 30 male saw-mill workers with minimum exposure of more than 5 years. All the workers included were nonsmokers. Those with a history of respiratory tract infection symptoms during previous 6 weeks and subjects suggestive of any active respiratory disorder were excluded by thorough history and clinical examination. Parameters studied were forced vital capacity (FVC), ratio of FVC at the end of the first second to the total FVC forced expiratory volume 1 s (FEV1)/FVC ratio, peak expiratory flow rate, forced midexpiratory flow rate (FEF) 25-75%, and maximum ventilatory volume (MVV). 30 age-matched non-smoker male controls in occupations without high bioaerosol exposure and residing in the area at least 2 km away from saw-mills but with similar socioeconomic status (as assessed by enquiring) were also evaluated for the same parameters. Measurements were performed following the recommendations of the American Thoracic Society.<sup>[5]</sup> Percentage of predicted values were considered for comparison instead of absolute values of different PF test (PFT) parameters to nullify the effect of most important confounding factors in PFs such as age, sex, height, and weight. [6] Prevalence of respiratory symptoms such as cough, phlegm, wheezing, and breathlessness was also assessed using American Thoracic Society's Division of Lung Diseases' respiratory symptoms questionnaire for adults.[7]

Volumetric air sampling for the survey of bacterial and fungal concentration (CFU/m³) in the air of saw-mills was conducted using Hi-Air Sampler (Hi Media Laboratories, India). Sampling was done in duplicate at two places at about 1.5 m above the surface, to simulate the human breathing zone fortnightly for 1 year duration. Hi-Air sampler is a centrifugal impactor where air is impacted onto isolation medium (separation volume of 40 L/min) contained in the strips. Nutrient agar with cycloheximide (0.5 mg/ml to suppress fungal growth)<sup>[8]</sup> and Martin's Rose Bengal agar medium strips were used for the isolation of bacteria and fungi, respectively. After sampling for 5 min at different sites, strips were incubated in an inverted position at 35°C for 2 days in an incubator for bacteria and at room temperature (25-30°C) for 5 days for fungi. Colonies were

counted and expressed as colony forming units per cubic meter of air.

To get an idea of the bioaerosol levels to which saw-mill workers are exposed, the microbial concentration of saw-mills was compared with an indoor air of residential area where the control subjects resided (sampling at two sites, fortnightly in duplicate for 1 year). PFT assessment was done in last month of bioaerosol survey.

Descriptive statistics was used to present the data. Unpaired Student's *t*-test assuming unequal variance was used to compare values of PFT of study and control groups. For correlation of PFs with duration of exposure, the correlation coefficient was calculated. Odds ratios with 95% confidence interval (CI) for case-control comparison were calculated for different respiratory symptoms. Odds ratios with a lower limit of 95% CI more than one were considered significant. Unpaired Student's *t*-test assuming unequal variance was also used to study differences of bacterial and fungal concentrations between saw-mills air and residential air.

### **RESULTS**

The composition of saw-mills workers (study group) and control group is shown in Table 1. The groups were comparable for age, height, and weight. Even after similarities in study and control group, percentage of predicted values for a particular sex, age, height, and weight were considered for comparison to nullify the effect of these most important confounding factors for PFs.

Reporting of different respiratory symptoms by control group and workers are shown in Table 2.

Percentages of predicted values for all PFT parameters along with their absolute values are given in Table 3. The statistical significance of the difference between mean values for cases and controls are also depicted in Table 3. Duration of occupational exposure in years was correlated (Pearson correlation coefficient "r") with different PF parameters. It was observed that all parameters were inversely correlated with duration of exposure. However, except FVC % predicted

**Table 1:** Composition of study and control groups with comparison of anthropometric data

Parameter	Control	Saw-mill	<i>t</i> -test	Remark
		workers		
Age (years)	39.46 (±2.014)	38.92 (±1.89)	>0.05	NS
Height (cm)	162.46 (±1.794)	163.15 (±1.19)	>0.05	NS
Weight (kg)	59.73 (±1.757)	58.90 (±1.942)	>0.05	NS

Parentheses show SEM. SEM: Standard error of mean, NS: Not significant

(r = -0.3621, P < 0.05), all other PFT correlations with duration of exposure were statistically insignificant.

Mean total bacterial and fungal concentrations in saw-mills air and indoor residential air are depicted in Table 4 along with the statistical significance of difference. Month-wise total fungal and bacterial concentration in the air of sawmills throughout 1 year study and the same in the air of residential area are shown in Figure 1. Fungal and bacterial concentrations in sawmill air  $(20.5 \times 10^3 \text{ to } 142.4 \times 10^3 \text{ CFU/m}^3 \text{ and } 34.2 \times 10^3 \text{ to } 132.0 \times 10^3 \text{ CFU/m}^3$ , respectively) were significantly more (P < 0.001) than control residential air  $(0.31 \times 10^3 \text{ to } 13.0 \times 10^3$ 

 $0.76 \times 10^3$  CFU/m<sup>3</sup> and  $0.43 \times 10^3$  to  $1.8 \times 10^3$  CFU/m<sup>3</sup>, respectively).

### DISCUSSION

Organic dust in saw-mill is responsible for high levels of bacteria and fungi. In this investigation average, fungal concentration in sawmill air was 178 times of indoor residential air whereas average bacterial concentration was 111 times in saw-mills as compared to indoor residential air. Exposure to a concentration of airborne microorganisms

**Table 2:** Number of cases and controls reporting different respiratory symptoms (n=30 in each group) Case-control odds ratio 95% CI **Symptoms** Number of person reporting Remark symptoms Saw-mill workers Control 14 3 Significant Cough 7.87 1.95-31.67 Phlegm 7 1 8.82 Significant 1.01-76.96 5 Wheezing 1 5.8 0.63-53.01 NS 8 Breathlessness 1 10.54 1.23-90.66 Significant

NS: Not significant, CI: Confidence interval

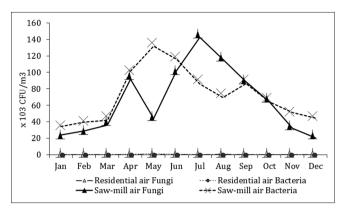
**Table 3:** Absolute values and percent predicted values (corrected for age, height and weight) of various PFT parameters in saw-mill workers and control group

PF Parameters	Control	Controls (n=30)		Cases (n=30)		
	Absolute	% predicted	Absolute	% predicted		
FVC (L)	2.71 (±0.09)	89.59 (±2.75)	2.58 (±0.13)	81.91 (±3.24)	>0.05	
FEV1	2.40 (±0.08)	96.77 (±3.30)	2.19 (±0.12)	86.50 (±3.2)	< 0.05	
FEV1/FVC ratio (%)	88.42 (±0.77)	-	84.93 (±1.21)	-	>0.05	
PEFR (L/s)	6.66 (±0.30)	80.05 (±3.38)	5.14 (±0.29)	60.28 (±2.72)	< 0.001	
FEF 25-75% (L/s)	3.08 (±0.21)	79.55 (±3.64)	2.39 (±0.14)	62.88 (±2.45)	< 0.001	
FEF 0.2-1.2 (L/s)	5.24 (±0.33)	80.26 (±4.55)	$3.82 (\pm 0.26)$	58.12 (±3.23)	< 0.001	
MVV (L/min)	92.16 (±4.17)	74.02 (±2.53)	$78.07 (\pm 3.35)$	60.27 (±2.22)	< 0.001	

Parentheses show SEM. Standard error of mean, \*t-test for independent samples with P value for difference (% predicted), FVC: Forced vital capacity, FEV: Forced expiratory volume 1 s, PEFR: Peak expiratory flow rate, MVV: Maximum ventilatory volume, FEF: Forced mid-expiratory flow rate

Table 4: Bacterial and fungal concentration in the air of saw-mills and indoor residential air and their comparison					
Bioaerosol Concentration	Indoor residential air (CFU/m³)	Saw-mill air (CFU/m³)	Unpaired <i>t</i> -test, <i>P</i> value		
Fungal concentration					
Rainy	$0.403 \times 10^3 \ (\pm 0.0204)$	$112.33 \times 10^3 \ (\pm 11.48)$	< 0.001		
Winter	$0.35 \times 10^3 \ (\pm 0.023)$	$35.18 \times 10^3 (\pm 9.95)$	< 0.001		
Summer	$0.356 \times 10^{3} \ (\pm 0.0277)$	$50.21 \times 10^3 \ (\pm 14.15)$	< 0.001		
Average of a year	$0.37 \times 10^3 \ (\pm 0.014)$	$65.91 \times 10^3 \ (\pm 11.85)$	< 0.001		
Bacterial concentration					
Rainy	$0.62 \times 10^3 \ (\pm 0.083)$	$89.44 \times 10^3 (\pm 9.67)$	< 0.001		
Winter	$0.478\times10^3~(\pm0.035)$	$48.54 \times 10^3 \ (\pm 6.28)$	< 0.001		
Summer	$0.860 \times 10^3 \ (\pm 0.243)$	$78.78 \times 10^3 \ (\pm 22.81)$	< 0.001		
Average of a year	$0.65 \times 10^3 (\pm 0.09)$	$72.26 \times 10^3 (\pm 9.31)$	< 0.001		

Parentheses show±SEM. SEM: Standard error of mean



**Figure 1:** Month-wise fungal and bacterial concentrations (10<sup>3</sup> CFUs/m³) in residential air and saw-mill air

revealed in the present study is higher than the degree of exposure found in saw mills in some of the studies<sup>[2,9,10]</sup> whereas similar high concentration has been reported from sawmills where there is more mold infection of wood.<sup>[11]</sup>

Increased prevalence of respiratory symptoms and decreased PF values, particularly highly significantly reduced FEF 25-75% in study group clearly indicate compromised PFs in these workers with the involvement of small airways. FEV1 is influenced not only by the elastic recoil of lung and the resistance of intrapulmonary airways but also by the effort dependent portion of a maximum forced expiration. Flow (and consequently volume change) which is generated in the effort dependent portion is a function of the strength, effort, and coordination of the subject as well as the resistance of the airways including the larynx.[12] Muscular endurance and power of the occupational workers in contrast to the control subjects may have contributed to a comparatively lower decrease in FEV1. The FEF (25-75%) has been reported to possess more sensitivity for determining airway obstruction than the FEV1, either as a percent of predicted or percent of FVC. The FEF 25-75% is believed to reflect frictional resistance of small airways particularly the diameter of small poorly supported airways.<sup>[7]</sup> Similar decrements in FEF 25-75% as obtained in this study have been reported in different occupational environments including sawmills with bioaerosol exposure.[10] This finding suggests early small airway obstruction changes in workers exposed to study environment.

Significantly reduced MVV in saw mill workers also points to restrictive lung function changes. Lung function impairment of restrictive nature in saw mill workers has been reported earlier. Statistically insignificant correlations of the duration of exposure with PF impairment may be due to a "healthy worker effect," Similarly, predisposing host factors might also play a role in differential impairment of lung functions with different exposure to organic dust. [14]

Other studies in occupational environments with high exposure to bioaerosols, have shown that exposure to bioaerosols may

cause a wide range of respiratory and mucosal symptoms that vary in severity from mucous membrane irritation to acute or chronic diseases such as toxic alveolitis (also called organic dust toxic syndrome [ODTS]), and allergic alveolitis and asthma.<sup>[15]</sup>

Many fungal species have been described as producers of Type I allergens (immunoglobulin E [IgE] binding allergens) and IgE sensitization to common outdoor and indoor fungal genera such as *Alternaria*, *Penicillium*, *Aspergillus*, and *Cladosporium* spp. is strongly associated with allergic respiratory disease, especially asthma. [16,17] Fungi are also a source of  $\beta$ (1ungiglucans which are suspected to induce inflammatory responses through release of cytokines (like tissue necrosis factor  $\alpha$ , interleukins [IL]-1 $\beta$ , IL-6, and IL-8) and cause non-allergic respiratory symptoms. [18-21]  $\beta$ (10iraglucans also originate from some bacteria and most of the higher plants. [22]  $\beta$ (1 $\rightarrow$ 3)-glucan may act synergistically with endotoxin in causing airway inflammation and may enhance the production of specific IgE. [23]

Bacterial cell wall components, such as endotoxin (present only in Gram-negative bacteria) and peptidoglycans (most prevalent in Gram-positive bacteria), are agents with important pro-inflammatory properties that may induce respiratory symptoms. Endotoxin has been recognized as an important factor in the etiology of occupational lung diseases including (non-allergic) asthma and ODTS.<sup>[24]</sup> Subjects exposed to endotoxin inorganic dust exposure have shown lung function impairment<sup>[25]</sup> and positive associations between endotoxin exposure and health effects including both reversible (asthma) and chronic airway obstruction, respiratory symptoms and increased airway responsiveness in a large variety of occupational environments characterized by different exposure levels and different compositions of the bioaerosol exposures.<sup>[26,27]</sup>

None of the workers in these saw-mills used a respiratory mask. This was confirmed by enquiring each worker in detail. This lack of preventive measure makes them vulnerable to more bioaerosol exposure.

# **CONCLUSION**

With a constraint of this study in the form of lack of valid quantitative multifactorial exposure assessment methods, the cause-effect relationship between specific components of bioaerosol and respiratory health could not be established. However, comparative study of bioaerosol exposure in saw-mills and residential air clearly suggest very high bioaerosol exposure in saw-mills. Similarly, though with a limitation of small sample size, decreased PF values in saw-mill workers in comparison with control subjects not exposed to any such occupational environment points toward a direct relationship between bioaerosol exposure in sawmills and respiratory

health of workers. The outcome of the study calls for urgent measures to reduce environmental bioaerosol exposure in sawmills like the use of respiratory masks and advanced technology in sawmills to reduce wood dust at source in addition to intermittent monitoring of bioaerosol.

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