# Impact of Sputum Volume in the Diagnosis of Smear-Positive Pulmonary Tuberculosis

#### Abstract

**Objective:** To find the impact of sputum volume in the diagnosis of smear-positive (SP) pulmonary tuberculosis (PT) and its association with gender. **Materials and Methods:** The study was conducted in the Department of Microbiology, GSL Medical College. PT patients were included and informed to provide good-quality sputum; volume was not mentioned. Smears were stained by Ziehl–Neelsen technique. Based on the volume of sample submitted, the participants were divided into three groups. Chi-square test was used to find the statistical significance; P < 0.05 was considered statistically significant. **Results:** Volume-wise, 22, 38, and 74 participants submitted sputum, respectively, in groups, <2 mL, 2–5 mL, and >5 mL; statistically, the difference was significant (P < 0.05). The smear positivity was 13% (15), 28% (32), and 59% (68), respectively, in groups, <2 mL, 2–5 mL, and >5 mL; statistically significant (P < 0.05). Among the missed cases, the difference was statistically significant (P > 0.05) in females. **Conclusions:** Notable number of SP cases are identified in  $\leq 5$  mL sputum. Hence, sample should not be discarded/rejected if the volume is <5 mL.

Keywords: Participants, sample, sputum, volume

## Introduction

Smear microscopy (SM) is the standard technique for the diagnosis of pulmonary tuberculosis (PT).<sup>[1]</sup> Inability to identify drug resistance and limited utility in the diagnosis of tuberculosis (TB) in HIV are the disadvantages of SM.<sup>[2]</sup> However, this is a rapid and economical technique. Due to the advantages and nonaffordability of the patients on rapid TB diagnostic tests, in high TB burden countries such as India, Pakistan, and Sub-Saharan Africa, SM is commonly used technique.<sup>[3]</sup>

As per Rashid *et al.* report, 5 mL is the minimum volume of sputum required<sup>[4]</sup> for SM. With this background, a study was undertaken to find the impact of sputum volume in the diagnosis of smear-positive (SP) PT and its association with gender.

## **Materials and Methods**

The study was conducted in the Department of Microbiology, GSL Medical College, from January 2017 to November 2017. The study protocol was approved by the Institutional Ethics Committee of GSL Medical College. Informed written consent was provided by all the volunteers who participated in the study. PT patients were included in the study. Participants were provided with sterile sample containers and informed to submit good-quality sputum; volume was not mentioned. Two samples were collected with a gap of 1 h, same day approach.<sup>[5]</sup> Smears were prepared and stained by Ziehl-Neelsen staining technique. Smear preparation, staining, screening under microscope, and grading were done as per the Revised National Tuberculosis Control Programme (RNTCP) guidelines.<sup>[6]</sup>

Based on the volume of sample submitted, the participants were divided into three groups, <2 mL, 2–5 mL, and >5 mL. Chi-square test was used to find the statistical significance among the SP cases between the groups and gender; P < 0.05 was considered statistically significant.

# **Results and Discussion**

A total of 134 patients were included in the study; the male-female ratio was 1.4. Volume-wise, 22 (16%), 38 (29%), and 74 (55%) participants submitted sputum,

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respectively, in groups, <2 mL, 2–5 mL, and >5 mL; statistically, the difference was significant (P < 0.05). Gender-wise, in males, respectively, 17 (13%), 29 (22%), and 32 (24%) participants submitted <2 mL, 2–5 mL, and >5 mL sputum; 96% (75/78) males were aware that 5 mL is the minimum volume of sputum required for SM. Inspite of this, just 41% (32/78) males were only submitted >5 mL sputum. However, in female category, respectively, 5 (3.7%), 9 (6.8%), and 42 (31%) participants submitted <2 mL, 2–5 mL, and >5 mL sputum. All the females were aware regarding the minimum volume of sputum to be submitted for SM. By remembering that, 75% (42/56) females submitted >5 mL sample.

Of the 134 study participants, 115 were SP. Among the SP cases, the male-female ratio was 1.3. According to the sputum volume, the smear positivity (SPT) was 13% (15), 28% (32) and 59% (68), respectively, in groups, <2 mL, 2–5 mL, and >5 mL; statistically, the difference was significant (P < 0.05). In males, volume-wise, 7, 4, and 2 cases were missed, respectively, in groups, <2 mL, 2–5 mL, and >5 mL; statistically, the difference was significant (P < 0.05). In females, volume-wise, 0, 2, and 4 cases were missed, respectively, in groups, <2 mL, 2–5 mL, and >5 mL; statistically, the difference was significant (P < 0.05). In females, volume-wise, 0, 2, and 4 cases were missed, respectively, in groups, <2 mL, 2–5 mL, and >5 mL; statistically, the difference was not significant (P > 0.05).

Bhat *et al.*<sup>[11]</sup> reported that upgradation of sputum quality and quantity increases the SPT. Even our previous studies<sup>[7-9]</sup> were also designed in such a way that 5 mL is the minimum volume of sputum to be submitted for SM. If the volume of sputum is <5 mL, the participants were informed to submit another sample and pooled sample was used for SM. Whereas, in the field conditions, laboratory technicians usually discard/reject the sputum if the volume is <5 mL. But, as per our study findings, 41% (47/115) SP cases are identified in the groups where the sputum volume is  $\leq$ 5 mL; gender-wise, it was 35 and 12, respectively, for male and female. It was mentioned in one report that the sputum volume is not critical for unconcentrated SM.<sup>[10]</sup> However, Warren *et al.*<sup>[11]</sup> reported that the sensitivity of SM will increase with >5 mL sputum.

### Conclusion

As per the current study findings, notable number of SP cases are identified in  $\leq 5$  mL sputum. Hence, sputum should not be discarded/rejected if the volume is <5 mL. In case if the smear is negative for acid-fast bacilli, as per the new RNTCP diagnostic algorithm, chest X-ray and cartridge-based nucleic acid amplification tests can be considered for the diagnosis of TB.<sup>[12]</sup> Study on confirmed PT patients and small sample size are the limitations of this research.

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#### **Conflicts of interest**

There are no conflicts of interest.

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