

# Prevalence Survey of Bacillary Pulmonary Tuberculosis in Western Uttar Pradesh, India

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## Abstract

**Settings and Objectives:** A house to house, cross sectional, symptom elicitation, survey was conducted in two randomly selected districts of Western Uttar Pradesh, (Banda and Kanpur Nagar). This was as per the common national format to assess the prevalence of pulmonary tuberculosis in the community aged  $\geq 15$  years, developed by the Central Tuberculosis Division (CTD), Ministry of Health and Family Welfare, Government of India. The study was undertaken after taking informed written consent of all subjects.

**Methods and Study Population:** More than 98% of 96,476 population of selected proportional to size (PPS) clusters of these districts were screened by trained workers on symptomatology elicitation. Diagnosis of pulmonary tuberculosis was done by AFB sputum positivity on light microscopy and/or culture positivity for MTB on LJ medium.

**Results:** About 7.6% of the screened population was chest symptomatic. Three hundred and forty five patients were AFB positive/MTB culture positive in the screened population. The crude prevalence rate was 357.6/100000 and corrected prevalence for the whole registered population 385.2/100000 (range 328-441 with 95% CI). Pulmonary tuberculosis was observed more in males as compared to females, more in urban areas as compared to rural areas and increased progressively with age, highest rates were observed for 65 years and above age groups. About 76% of cases were AFB positive by light microscopy; 58% were culture positive on LJ medium; 23.5% were smear negative but culture positive.

**Conclusions:** Using a common national protocol this study for the first time provides baseline prevalence rate of bacillary pulmonary tuberculosis in the area. This prevalence is higher than the reported national average. Diagnosis by both smear microscopy and culture has been observed to be a better strategy for detection of tuberculosis.

**Key words:** AFB; Sputum positivity; Light microscopy; Culture positivity; MTB; DOTS

## Introduction

Tuberculosis is one of the ancient infectious diseases, which continues to be endemic in several parts of the world. To treat and contain the disease in India, Revised National Tuberculosis Control Programme (RNTCP) and DOTS (Directly Observed Treatment Supervised) was introduced from the year 1997 onwards and expanded in a phased manner. Uttar Pradesh is a large populous state, home to 199.5 billion inhabitants, (16%) of the country's population. It has 71 administrative districts (recently increased to 74). For operational reasons, ease of administration, management and supervision of the programme the State has been grouped into 2 sub divisions with 35 districts in Western UP and Bundelkhand region and remaining districts in Eastern and Central UP.

To estimate the prevalence of pulmonary tuberculosis, the Central Tuberculosis Division (CTD) of Government of India funded this study in Western Uttar Pradesh. On the basis of a common generic protocol, a cross sectional, house to house survey of chest symptomatics, aged  $\geq 15$  years was carried out in selected PPS (Population Proportional to Size) clusters of Kanpur Nagar and Banda during 2008-10 and subsequent microscopic and culture examination of sputa of chest symptomatics.

## Study Population and Methods

### Sample size estimation for the survey

Assuming a prevalence of 400/100,000 pulmonary tuberculosis cases,

design effect of 2, expected drop out of 10% (from the time of recruitment in the survey to completion of survey activities), 95% confidence level, the required sample size was estimated to be about 90,000 population. The calculation was based on the assumption that 60% of the cases would be picked up by symptom elicitation [1]. It was decided to allocate the estimated sample size to the two districts ensuring a minimum of 90% coverage of  $\geq 15$  year's population.

### Sampling methodology

Stratified cluster sampling was undertaken, taking a district as a sampling universe. Rural (village as unit) and urban (municipal wards as unit) were sampled. Sampling of these was done separately as well as collectively to observe differences if any between the two. A three stage sampling technique was adopted. In the first stage the districts of Kanpur Nagar and Banda were selected from 35 districts of the state, by Simple Random Sampling method taking into consideration the population size, density of the population / square Km. Kanpur Nagar from high density districts and Banda from medium to low density districts were selected. Population of 45000 was allocated to each of these two districts.

In the second stage proportional sampling for both rural and urban areas for the two districts was undertaken. In Banda district (1.5 million residents), 84% of population resided in rural areas while 16% in urban areas. The clusters were selected accordingly so that 84% of samples were envisaged to be collected from rural areas and the remaining from

urban clusters. In Kanpur Nagar (population of about 4.2 million) with 32.8% residing in rural and 67.2% in urban areas, clusters were selected accordingly so as to yield 65% samples from urban clusters and 35% from rural clusters.

In the 3<sup>rd</sup> stage clusters were selected with PPS (Probability Proportional to Size) sampling method with proportional allocation to both rural and urban strata. The clusters were chosen from the entire list of clusters in the district arranged in the ascending order of population (2001 census data). Fifty-two clusters were selected from both the above districts with the assumption that 90 % coverage of the clusters would yield a sample of 90000 persons of >15 years age. The sample included 35 clusters from Banda district (3 urban and 32 rural), and 17 clusters (6 urban and 11 rural) from Kanpur Nagar. These clusters were geographically well distributed.

### Ethical considerations

The project was approved by the Institute Ethical committee (IEC) before starting the survey. The IEC was informed periodically of the progress of the study, adverse events as well as final results of the survey. In the rural areas, the elected village representatives and elderly were informed of the proposed survey activities and explained about the symptomatology, investigations and treatment of tuberculosis. In the urban areas members of the legislative committees, and other leaders were involved. The individual households were visited subsequently; informed of the survey; written patient information sheet distributed, and, informed written consent taken at the start of the survey from each household member of  $\geq 15$  years age in Hindi (language used by the population).

### Training

Standard Operating Procedures (SOP) were prepared as per the approved protocol for all level of workers i.e. field workers, sample collectors, data entry operators, laboratory technicians, supervisors, medical officers. The staff was trained on the SOP as well as bio-safety measures, screening methodology of population, transportation, processing at NJIL & OMD, reporting of results etc in batches, both at the Institute as well as at the field stations.

### Survey methodology

Within every cluster, the entire eligible population (residing for 6 and more months in the area) of  $\geq 15$  years were registered by door to door census after obtaining their informed written consent. Each registered person was interviewed individually for the presence or absence of the following symptoms, and recorded in the approved data entry card:

- Continuous cough for  $\geq 2$  weeks [C]
- Chest pain of 1 month and more [P]
- Unexplained fever of 1 month or more [F]
- Hemoptysis [H]
- History of anti-TB treatment [T]

All persons with one or more of the above were eligible for sputum collection and registered. Findings were entered, both in the soft as well as hard copy. All efforts were made to elicit the information and investigations undertaken, by visiting the absentees in the population at different times of the day and during festivals/holidays.

Two sputum specimens, one spot and one early morning (OVM) were collected in sterile sputum containers and labeled. To each container, equal volume of 1% CPC was added using aseptic precautions to preserve as well as prevent over growth of other organisms during transportation.

The containers were transported at room temperature to the laboratory of NJIL & OMD within 72 hours of collection, with bio-safety precautions.

### Bacteriological examination

At the laboratory, patient details, naked eye examination of sputum specimen was recorded as per approved format. In cases of loss or inadequate sample, repeat sample was requested and examined. A direct smear was prepared under aseptic bio-safety conditions and examined using binocular light microscope after staining with Ziehl-Neelsen (ZN) stain. The light microscopy results were recorded after viewing about a hundred fields as per RNTCP and protocol guidelines on hard and soft data entry forms.

Each of the remaining specimen was inoculated on two slopes of Lowenstein Jensen medium (LJ) in BSL3 laboratory of the Institute. The slopes were incubated at 37°C and examined every day for first week and once every week for 8 weeks for observing the presence of mycobacterial colonies. The findings were recorded weekly as per protocol format. The mycobacterial colonies were confirmed as *Mycobacterium tuberculosis* (MTB) using Niacin test; 68°C Catalase test; and after further incubation on to LJ medium containing Para-nitro Benzoic acid (PNB). Colonies found to be niacin positive with no growth on PNB containing medium, were labeled as *Mycobacterium tuberculosis*.

### Quality assurance

For the quality check of the survey, 10% of the registered population was randomly resurveyed by supervisors (from different area) by random numbers generated by Minitab-12 statistical software for elicitation of eligibility for sputum collection. Results were cross verified and discrepancy if any was corrected by repeat survey of the locality. AFB staining, microscopy, and grading of smears, as well as culture and reporting were subjected to quality assurance protocols as per RNTCP guidelines by Intermediate Reference Laboratory (IRL) and Supra National Reference Laboratory as per National Programme.

Data from both the districts was digitalized by two independent persons, matched and rectified.

### Definitions used:

A person found to be smear and /or culture positive on one or both specimens was labeled as a case of pulmonary tuberculosis.

### Statistical Methods:

Crude prevalence was estimated by dividing the total number of smear and or culture positive by the total number of individuals screened.

Standard deviation was calculated using the formula  $SD = \sqrt{P(1-P)}$ .

Standard error (SE) was estimated as Standard deviation (SD)/ $\sqrt{n}$ .

Confidence intervals (95%) was calculated as mean of the binomial exact  $\pm 2SE$ . Individual level analysis was done using logistic regression model with robust standard error.

Chi-square test with continuity correction was used to test the significance of differences between proportions and p values <0.05 were considered significant.

### Results

The total registered population (Table 1) in the surveyed clusters was 96476 which included 46710 from Banda and 49766 from Kanpur Nagar. A total population of 95673 (99.2%) was covered in the survey (46177 [98.9%] from Banda and 49496 from Kanpur Nagar 98.6%). The coverage of the population during the survey was more than 98% (98.6 to 99.1% in males; 99.3-99.8 % in females) (Table 1).

In the registered population 7.6% (3.5% to 16.9% of different age groups) had symptoms related to pulmonary tuberculosis (chest symptomatics). These symptoms were more among the males as compared to females in the same age group, and also varied between the urban and rural clusters (Figure 1).

The proportion of chest symptomatics increased with age and was observed to be 15 to 17% in the age group of 55 to 75 years in the rural population and 10 to 15% in the same age group in the urban population (Figure 1). Most of the sputum eligible's had multiple symptoms i.e. combination of one or more of the symptoms with cough (Table 2). This was similar in the rural and urban population as well as in both the genders.

### Random quality control for verification of enumeration of chest symptomatics

Using Minitab -12 software, 8868 population was re-surveyed, selected households visited and symptoms enumerated by supervisors of different district and results recorded independently. Of these 690, cases were symptomatic, and 8159 persons were non- symptomatic by both the survey teams. The KAPPA value of concordance was 0.72.

### Bacteriological results

Over 97% of chest symptomatics sputum was collected, transported, examined and reported (highest reading tabulated). This was 97.2% (95.8 to 97.8%) for the spot sputum samples and 86.9% (85 to 89.2%) for OVM sputa (Figure 2).

Age group	Number Registered			Number Screened for symptoms % in brackets			Number with symptoms % in brackets		
	Male	Female	Total	Male	Female	Total	Male	Female	Total
15-24 yrs	15643	14075	29718	15501 (99.1)	14025 (99.6)	29526 (99.4)	591 (3.8)	446 (3.2)	1037 (3.5)
25-34 yrs	9779	10209	19988	9645 (98.6)	10145 (99.4)	19790 (99.0)	587 (6.1)	613 (6.0)	1200 (6.1)
35-44 yrs	8901	8782	17683	8788 (98.7)	8751 (99.6)	17539 (99.2)	746 (8.5)	711 (8.1)	1457 (8.3)
45-54 yrs	7319	6044	13363	7214 (98.6)	6034 (99.8)	13248 (99.1)	750 (10.4)	514 (8.5)	1264 (9.5)
55-64 yrs	4404	4448	8852	4342 (98.6)	4421 (99.4)	8763 (99.0)	673 (15.5)	503 (11.4)	1176 (13.4)
65-74 yrs	2865	2400	5265	2828 (98.7)	2384 (99.3)	5212 (99.0)	560 (19.8)	308 (12.9)	868 (16.7)
75+ yrs	927	680	1607	917 (98.9)	678 (99.7)	1595 (99.3)	190 (20.7)	80 (11.8)	270 (16.9)
All age groups (≥15yrs)	49838	46638	96476	49235 (98.8)	46438 (99.6)	95673 (99.2)	4097 (8.3)	3175 (6.8)	7272 (7.6)

Table 1: The age and gender wise distribution of population in the survey

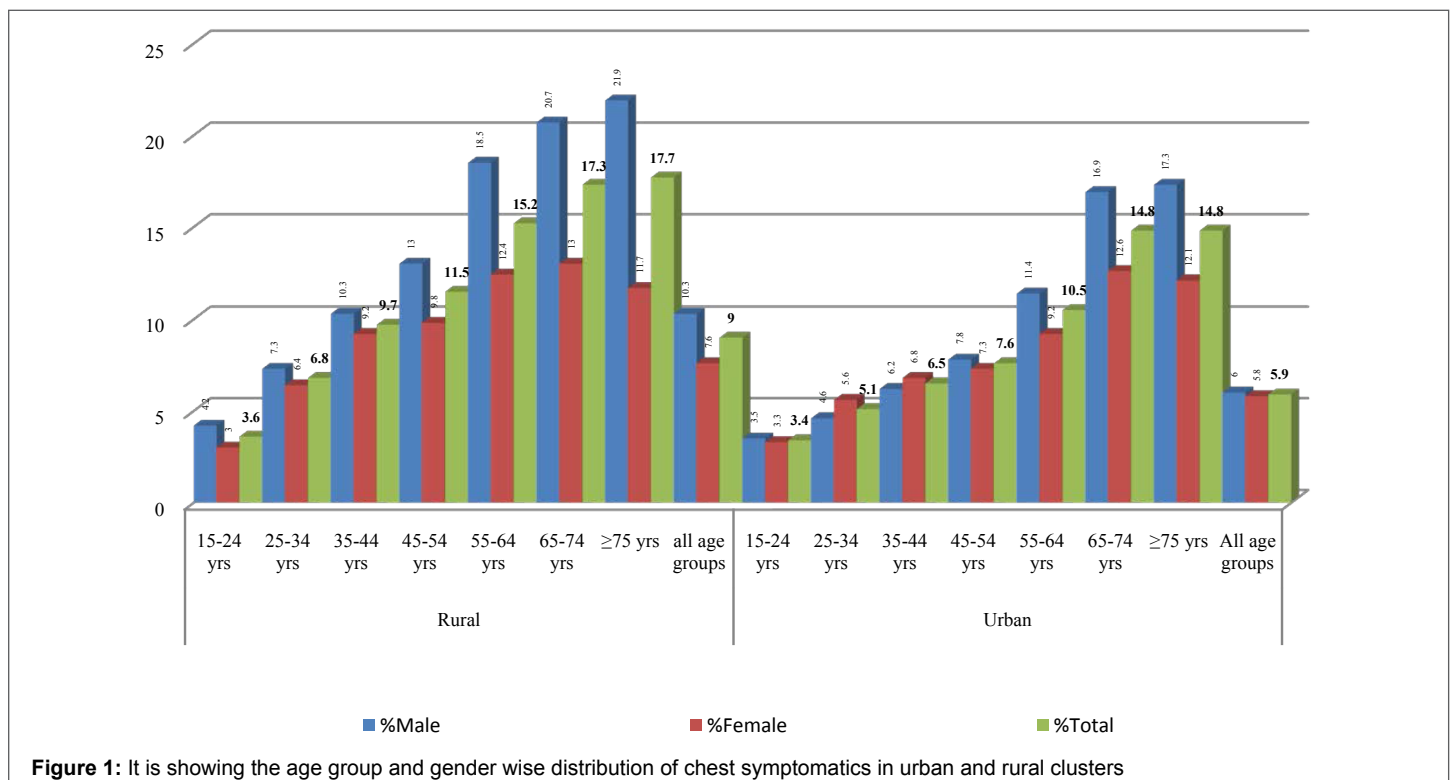
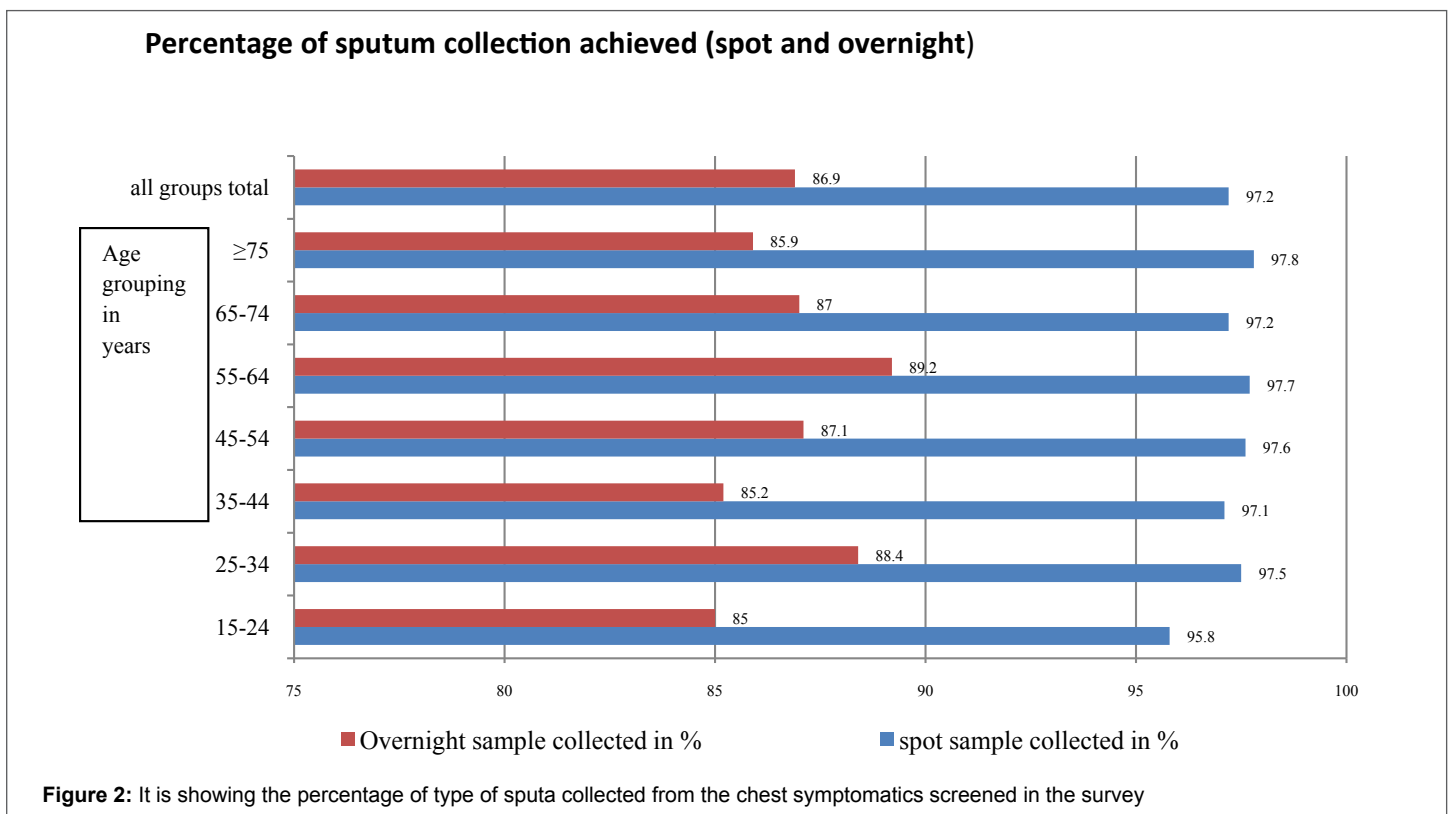


Figure 1: It is showing the age group and gender wise distribution of chest symptomatics in urban and rural clusters

Symptom	Rural	Urban	Combined
Cough alone	36.5	42.9	38.8
Cough with any of other symptoms	55.9	50.6	54.0
Chest pain alone	2.3	3.7	2.8
Fever alone	1.1	0.6	1.7
Haemoptysis alone	0.8	0.3	0.6
History of taking anti TB treatment irrespective of the above symptoms	30.8	31.3	30.9

**Table 2:** The proportion of various symptoms (%) in sputum eligibles in the survey



**Figure 2:** It is showing the percentage of type of sputa collected from the chest symptomatics screened in the survey

Sputum AFB was positive (spot /overnight or both) for 264/345 cases (76.5%). Sputum culture was done of both the samples (LJ medium). Biochemically confirmed MTB was observed in 57.98% (200/345) of the cases (Table 3). The proportion of contaminated culture samples was 0.6% (within the acceptable limits).

Of the 345 bacteriological positive cases 116 were females and 229 were males. Table 4 shows the calculated prevalence of pulmonary tuberculosis in the population. The total crude prevalence of pulmonary tuberculosis in the area was 357.6/ 100,000 population. After applying the corrections for the population not covered, standard error, as detailed in the methods the prevalence of bacillary tuberculosis was 385.2/100,000 and ranged from 288 .9 -473.9/1000,000 with 95% confidence limits.

Prevalence of pulmonary tuberculosis was significantly more in males as compared to females (Table 4: Chi square=30.04; p<0.0001). The female patients were about 1/3<sup>rd</sup> of the total cases. Prevalence ranged from 207.3 to 300.7/100,000 in them. The prevalence of disease in people residing in urban settings was higher than in residents of rural clusters (Table 4). The corrected prevalence observed was 250/100,000 in the rural population with a range of 203 -397/100,000/1000,000 with 95%

confidence limits. For the urban clusters this was 497/100,000 with a range of 436 -557/100,000. This difference was also statistically significant (Chi square=34.08; p<0.0001).

As expected, the prevalence of the disease varied among different age groups. The corrected prevalence was 186/100000 population for the age grouping of 15-24 years (lowest) and it increased progressively: 355.9/100,000 for age group of 25-34 years; 403.3/100,000 for 35-44 years (age group); 465.5/100,000 for age group 45-54 years; 804.8/100,000 for 55-64 years group; 802.7/100,000 for age group of 65-74 years and 987.8/100,000 for age group of ≥ 75years respectively. The difference in the prevalence among the age groups was statistically significant Chi square=28.90; p<0.0001 (Figure 3).

## Discussion

The study shows that cross sectional house to house survey, with more than 97% coverage of population can be successfully conducted under programme settings (Table 1). Using the same methodology and protocol of the survey 95.1% population were screened in Jabalpur district of Madhya Pradesh [2], 93.7% were surveyed in Faridabad district of Haryana [3], and 91.4% in Wardha district of Maharashtra [4].

Bacteriological profile	No. of cases (%)	Cases without ATT (% of total cases)	Cases with history of ATT (% of total cases)
Sputum smear positive & culture positive	119 (34.5%)	60 (17.4)	59 (17.1)
Sputum smear positive & culture negative	143(41.4%)	93 (26.9)	50 (14.5)
Sputum smear negative & culture positive	81 (23.3%)	44 (12.7)	37 10.7)
Sputum smear positive & culture contaminated	2 (0.6%)	1 (0.3)	1 (0.3)
<b>Total</b>	<b>345</b>	<b>198 (57.4)</b>	<b>147 (42.6)</b>

**Table 3:** The bacteriological profile of the TB cases

All clusters		Registered population	Examined population	No. of cases detected	Crude Prevalence / 100,000	Corrected prevalence/100,000	Range
Total population		96476	95673	345	357.6	385.2	288.9-473.9
Gender type	Male	49838	49235	229	459.5	500.7	409.1-532.1
	Female	46638	46438	116	248.7	264.4	207.3 -300.7
Type of cluster	Rural	52890	52226	102	234	250	203-397
	Urban	43586	43447	243	459.4	497	436-557
Age wise grouping	15-24 yrs	29718	29526	51	171.6	186.0	129 - 227.8
	25-34 yrs	19988	19790	66	330.1	355.9	258.9 – 425.6
	35-44 yrs	17683	17539	66	373	403.3	292.7-481
	45-54 yrs	13363	13248	58	434	465.5	334.3 – 568.5
	55-64 yrs	8852	8763	50	6.4.5	604.8	426.6 – 756.6
	65-74 yrs	5265	5212	39	740.7	802.7	602.8 -1044.9
	≥75 yrs	1607	1595	15	839.8	987.8	

**Table 4:** The prevalence of pulmonary tuberculosis in the population

Chi square for gender difference male/v/s female 30.04;  $p < 0.001$  : chi square for rural v/s urban cluster is 34.08;  $p < 0.0001$

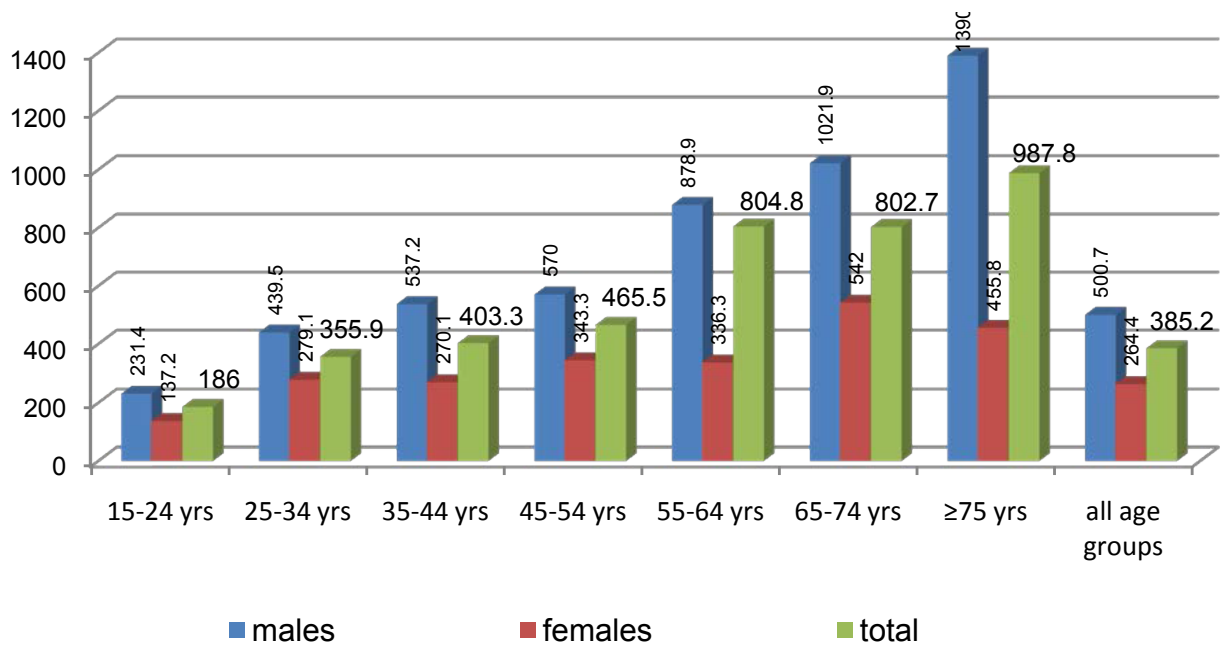
Among the population screened 7.6% (7272/95673) population (Table 1) were chest symptomatics. These ranged from 3.5% in the age group of 15-24 years to 16.9% in >75 years age group. There were more male chest symptomatics than female for the same age grouping, as well as more in rural clusters as compared to urban clusters (Figure 1). Similar findings have been observed by other investigators as well [2-5]. Most of the symptomatics had multiple symptoms with cough being the most common. About 31% of symptomatics had a previous history of taking anti-TB drugs which included monotherapy as well as multi drug therapy in various permutations (Table 2). This did not differ much in the rural and urban population or with the gender of the individuals. This big pool contributed significantly to the high prevalence of disease observed in this region.

Both spot and overnight sputum was collected from the inhabitants. In 86% of individuals OVM sputa was collected, while spot sample could be collected from >95% of individuals (Figure 2). Similar observations were also noted by other researchers [2-4], and highlight the importance of collecting and analysing both samples for better coverage.

One third of cases (119/345) were positive for both smear and culture for M.tb. About 58% of the sputum samples were culture positive on LJ media (Table 3). This was lower than that reported by others [2,4,5]. It is possible that a substantial number of the patients were under treatment or partially treated and were smear positive but did not harbor viable bacilli. Loss of viability due to addition of CPC could be a small contributory factor.

Two hundred and sixty-four patients (264/345; 76.5%) could be diagnosed as having tuberculosis on the basis of sputum smear positivity. Eighty one patients (23.5%; 81/345) were diagnosed as on basis of their culture results alone. This included 12.7% of patients with no H/O treatment and 10.7% who had history of taking anti-TB treatment earlier. Criteria for diagnosis of TB by sputum smear positivity alone would have therefore, missed about a fourth of cases and is an important finding. Culture along with smears can and should now be included in the programme settings as a routine. Recently, several laboratories have been accredited to perform culture and DST in the country [6] which can contribute for better programme management.

About two thirds of the total pulmonary TB cases detected in the survey were males. And the prevalence of the disease was nearly double that as compared to females (459.9/100000 v/s 248.7/100000). Higher prevalence of smoking in males might have contributed significantly to it. Moreover, in Uttar Pradesh tobacco chewing and sniffing is also very prevalent and may have contributed to the higher prevalence. About a third of all patients were females and the highest prevalence in them was in the age group of 65 to 74 years, followed by those in 55-64 yrs (Table 4) (Figure 3). Increase in prevalence in females in 25-34 years age group and above may be due to high exposure to bio-gas fuels in closed and ill-ventilated kitchens and living spaces. This increased considerably in the surviving females of older age groups. This may be due to malnutrition and post hormonal changes perpetuated by gender biased customs and eating practices in the region. In-depth studies on all possible contributory factors will be necessary.



**Figure 3:** It is showing the age wise and the gender differences observed in the prevalence of pulmonary tuberculosis in the study

The disease was more common in the urban clusters as compared to the rural clusters (chi square=34.08;  $p > 0.001$ ) although more chest symptoms were detected in the rural clusters (Figure 1). This was unlike that observed in the other prevalent surveys which showed a higher prevalence in the rural clusters [2-5]. Some of possible reasons for this could be that in Uttar Pradesh there is a higher density of population / sq metre, overcrowding, and unhygienic conditions in the urban clusters might have contributed to higher prevalence of the disease in these clusters. It is also possible that due to lack of facilities and ignorance, several people may have temporarily migrated from rural areas to urban areas (relatives in urban settings) for treatment, thus increasing the urban load.

The corrected prevalence for the registered population was 385.2 (range 288.9-473.9) with 95% CI (Table 4). This is more than that observed in Jabalpur [2], Faridabad [3] and at Wardha [4], reported recently. This was also higher than the national prevalence reported by RNTCP in India: 230/100,000 (range 150 to 319) [6], which of course is based on the reported prevalence. Chackraborty [7] had observed varied trends in the prevalence of tuberculosis in different populations, (especially tribal population) earlier. Reasons for higher prevalence in this area need to be investigated and addressed. This can be done by strengthening and monitoring of programme activities with focus on early and adequate access to services, coupled with better IEC activities so that the higher prevalence in this area can be controlled. Earlier no prevalence surveys for tuberculosis were conducted at such wide scale in Uttar Pradesh, the Indian state having largest population. Several reasons were attributed including poor approachability to population, vast culture diversity and practices, irregular and faulty data entry and collection. This survey highlights that this can be done in these difficult areas as well, with support of the state health services, information empowered people of the area. Although the state is way behind in registering a decline in tuberculosis at the National level as well as compared to other states, prompt IEC activities to health workers and elected representatives can motivate the population towards programme implementation. Better monitoring of the programme can be done to bring down the burden of pulmonary tuberculosis. This new original information about prevalence of bacillary pulmonary tuberculosis in this area and state will serve as baseline for strengthening

and evaluation of programme as well as research on dynamics of disease in these high endemic settings.

Not accounting for abacillary forms (X ray suggestive of tuberculosis) of the disease is a limiting factor of the present study, and prevalence would increase further if considered. Secondly there is no information on childhood tuberculosis (<15% yrs) and would contribute to higher TB burden in the community. Further no attempt has made to assess the HIV co-infection and other co-morbid conditions like diabetes, hepatitis, addiction and substance abuse in this study and this needs to be done.

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There is no Conflict of Interest with any of the agencies and the personnel in the study

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