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Original Research Article

Recent trends in seroprevalence of syphilis in different patient groups attending a regional centre for sexually transmitted infections in central India

Radha D. Datkar, Gopal N. Agrawal, Sonali S. Gosavi, Kalindi S. Deogade, Manisha K. Sharma, Vandana A. Agarwal*

Department of Microbiology, Government Medical College, Nagpur, Maharashtra, India

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***Correspondence:**
Dr. Vandana A. Agarwal,
E-mail: agarwal.gmc@gmail.com

ABSTRACT

Background: Easy access to Suraksha clinics for sexually transmitted infections calls for a review of seroprevalence of syphilis.
Methods: Serum samples from attendees of sexually transmitted infections (STI)/ reproductive tract infections (RTI) clinic/antenatal clinic, and samples of high-risk group (HRG: female sex workers and men having sex with men) brought by non-government organizations from 2017-22 were screened for syphilis by Venereal Disease Research Laboratory (VDRL) and Treponema pallidum hemagglutination (TPHA) tests. Samples positive by both tests were considered seropositive for syphilis. Statistical methods used for analysis were chi square test for linear trends and Kruskal Wallis test.
Results: In STI clinic percentage positivity for syphilis has shown a statistically significant decline from 2017-22. In RTI clinic the decline was significant from 2017-19 but not significant in the years 2019-22. In antenatal clinic and in HRGs the change in seroprevalence was not significant from 2017-22 and 2017-20 respectively. However, the HRGs showed a significantly increasing trend in syphilis seropositivity from 2020-22.
Conclusions: There is a significantly declining trend in the seroprevalence of syphilis in patients attending the STI/RTI clinic from 2017-22 and in HRGs from 2017-20. However, a significant increase in trend in HRG from 2020-22 may have been due to behavioural changes during the lockdown for covid 19 pandemic. A significant decline in syphilis in patients attending the STI and RTI clinic and in HRGs attending the regional centre indicates the effectiveness of consistent detection, treatment and counselling efforts of the national control program on STI in the region.

Keywords: Seroprevalence, Syphilis, TPHA, VDRL

INTRODUCTION

The seroprevalence of syphilis infection varies across different countries, as well as in different subpopulations of the same country.[1] The World Health Organization (WHO) estimates that 6 million new infections of syphilis occur every year globally.[2] World over the prevalence of syphilis in high-risk groups reported by World Health Organization in 2019 was 11.8% in MSM, 10.8% among FSWs and 3.2% in antenatal care attendees.[3] However exact data on prevalence of syphilis in different

population groups is not known due to the social stigma associated with sexually transmitted diseases.[4]

With easy access to Suraksha clinics, simple diagnostic tests, effective and reasonable treatment options syphilis can be fought successfully.[1] Further WHO global strategies to combat mother to child transmission has helped in reducing the prevalence of syphilis in antenatal patients. However key population comprising of high-risk groups (HRGs) like men having sex with men (MSM), female sex workers (FSW), transgender's (TGs) are still harbouring and transmitting syphilis.[3] Hence, present study was undertaken with the aim to study the trends of seroprevalence of syphilis in different population groups attending the regional centre for sexually transmitted infections in central India.

METHODS

A retrospective cross-sectional study was conducted at Regional STI Training, Research and Referral Laboratory (RSTRRL), Department of Microbiology, Government Medical College, and Hospital, Nagpur, Maharashtra, India from April 2017 to March 2022. A total of 96,621 patients selected for the study were categorised into four population groups. Group 1 was STI patients presenting with genital ulcer, Group 2 was RTI patients presenting with genital discharge, Group 3 was HRGs consisting of MSM and FSWs brought by Non-Government Organizations (NGOs) and Group 4 was antenatal patients attending the Antenatal Clinic (ANC) in the hospital. The only exclusion criteria was patients unwilling to give blood samples. Syphilis was diagnosed serologically by the non-treponemal VDRL test (VDRL antigen procured from The Institute of Serology, Kolkata) and a treponemal test viz TPHA test (Fortress Diagnostics Ltd, UK). Both tests were performed as per manufacturer's instructions. Samples that were positive by both VDRL and TPHA tests were labelled as seropositive for syphilis.

Both VDRL and TPHA test procedures consisted of antigen/reagent preparation, qualitative and quantitative testing.

VDRL test

Antigen preparation: The 0.4ml of buffered saline was pipetted into a 1 oz. flat bottomed reagent bottle. 0.5 ml of antigen was added directly on to the saline while rotating the bottle on a flat surface. The antigen was added drop by drop but rapidly so that it took approximately 6 seconds to complete the delivery. The last drop of the antigen was blown out and the bottle was rotated continuously for 10 seconds. Then 4.1ml of buffered saline was added from a 5ml pipette, the bottle was stoppered and shaken vigorously for approximately 10 seconds and left for 30 minutes for maturation.

Qualitative testing: The 0.05ml inactivated serum was pipetted out into one ring of a paraffin ringed glass slide. The serum was allowed to spread. One drop (1/60 ml) antigen emulsion was added to the serum. Slide was rotated for 4 minutes on a mechanical rotator. The tests were read immediately after rotation under microscope with low power objective (100x magnification). The results were read as non-reactive when there were no clumps, weakly reactive when small clumps and reactive when medium to large clumps were observed.

Quantitative testing: Quantitative test was performed on both weakly reactive and reactive serum samples. Successive two fold dilutions of the serum were made in 0.9% saline and each dilution was treated as an individual serum and was tested as described under qualitative testing. The results were reported in terms of the highest dilution of the serum that produced a definite reaction.

TPHA test

This test was performed on all VDRL reactive samples having titre ≥1:2.

Reagent preparation: All the reagents were supplied in a ready-to-use format. Test and control cells were thoroughly suspended prior to use. All the reagents were allowed to reach room temperature before use.

Qualitative testing: Each sample required three microwells. 190µl of diluent was added to well 1 to which 10µl of sample was added and mixed. 25µl of this solution was transferred from well 1 to wells 2 and 3. Further 75µl of control cells were added to well 2 and 75µl of test cells were added to well 3. Plate was covered and incubated for 45-60 minutes in an area which was away from heat, direct sunlight and vibration.

Quantitative testing: Each sample required 9 well of a microtitration plate. 190µl of diluent was added to well 1 and 25µl to wells 4 through to 9. A dilution of 1/20 was made by adding 10µl of serum to well 1 and then 25µl of 1/20 dilution was transferred to wells 2, 3 and 4. A dilution of 1/40 was then mixed in well 4 and 25µl of it was transferred to well 5. This step was repeated until the serial dilution was completed with discarding 25µl from the last dilution well. Further 75µl of test cells was added to wells 3, 4, 5, 6, 7, 8, 9 and 75µl control cells was added to well 2. Plate was covered and incubated for 45-60 minutes in an area away from heat, direct sunlight and vibration. The results were read as strong positive (4+) when uniform mat of cells covered the entire well base, positive (3+) when uniform mat of cells partly covered the well base, positive (2+) when uniform mat of cells was surrounded by a ring of cells, weak positive (1+) when smaller mat which was surrounded by a smaller more distinct ring of cells, indeterminate (+/-) when well defined button of cells with a clear centre, negative (-) when compact button in the well base.

For each population group and in each year (Table 1), A was the number of samples tested, B was the number of samples positive by both VDRL and TPHA tests and C was the percentage positivity.

The study was a part of the ongoing surveillance for sexually transmitted infections in our region that covered Vidarbha and adjoining states of Madhya Pradesh, Chhattisgarh, Andhra Pradesh, and Goa with a Memorandum of Undertaking on technical and administrative protocol with Maharashtra State AIDS Control Society (MSACS), Mumbai that was designed and ethically approved by National AIDS Control Organisation (NACO), New Delhi for implementation in all RSTRRLs in India.

The statistical analyses of data was done using SPSS version 21.0. Chi-square test for linear trends and Kruskal Wallis test was used to calculate p value for analysis of statistical significance of the data. P ≤0.05 was considered as statistically significant.

RESULTS

The population group attending the STI clinic showed a decline in the seropositivity of syphilis from 0.91% in 2017-18 to 0.73%, 0.64%, 0.55% and 0.74% in each consecutive year till 2021-22 (Table 1). The decline was statistically significant for each year, p value being 0.036, 0.010, 0.015, 0.024 respectively. The population group attending RTI clinic showed a significant decline from 0.1% in 2017-18 to 0.03% in 2018-19 (p=0.023). However, from 2019-22 although there was a decline in each consecutive year to 0.05%, 0.04% and 0.12%, it was statistically not significant (p=0.10, 0.24 and 0.45) for each year respectively. The population group attending Antenatal clinic showed a decline from 0.07% in 2017-18 to 0.02%, 0.02%, 0.07% in the years 2018-19, 2019-20 and 2021-22. The decline was however not significant with p value for each year p=0.10, 0.10, 0.15, 0.41 respectively. Amongst the HRG attendees the change in seroprevalence was not significant from year 2017-20, however a significant increase in seroprevalence was observed in the year 2020-21 (p=0.001) and 2021-22 (p=0.035).

Table 1: Seroprevalence of syphilis in different population groups attending a regional centre for sexually transmitted infections.

Population group	2017-2018			2018-2019			2019-2020			2020-2021			2021-2022		
	A	B	C (%)	A	B	C (%)	A	B	C (%)	A	B	C (%)	A	B	C (%)
Group 1: STI	3857	35	0.91	4906	36	0.73	6524	42	0.64	4521	25	0.55	5293	39	0.74
Group 2: RTI	4775	5	0.10	6446	2	0.03	6143	3	0.05	4558	2	0.04	6052	7	0.12
Group 3: HRGs	3216	1	0.03	668	0	0	566	0	0	162	14	8.02	67	1	1.49
Group 4: ANC	9895	7	0.07	9454	2	0.02	9049	2	0.02	3170	4	0.13	7299	5	0.07

DISCUSSION

In the present study seroprevalence of syphilis was determined in different population groups that attended the regional centre for diagnosis and treatment of STIs/RTIs. The STI and RTI population group showed a significantly decreasing trend in seroprevalence from the year 2017 to 2022 (Table 1). Khan et al, Nishal et al and Chopra et al have also reported statistically significant decrease from 4% (2004) to 1.39% (2008), from 8.30% (2008) to 2.24% (2012) and from 2.58% (2002) to 1.87% (2012) respectively in there STI group of patients.[5-7] These findings could be interpreted as indicators of effective implementation of STI control program in these population group in their respective regions. In contrast Sethi et al and Kashyap et al have reported rising trends of syphilis seroprevalence from 9.55% (2006) to 10.77% (2011) and 0.33% (2013) to 2.19% (2018) respectively which has been attributed to behavioural changes, socioeconomic factors and increasing prevalence of AIDS.[4,8] In the present study amongst HRG attendees decrease in seroprevalence was observed from 2017-20 though not significant. However, a highly significant increase was observed in year 2020-22. This can be due to increased spread of syphilis during COVID-19 pandemic because of behavioural changes in young population, unsafe sexual activities, and neglect towards STI counselling programs and services as the emphasis was more on control of pandemic.[9] Stanford et al, have also reported a significant increase in syphilis seroprevalence in a comparison between pre-pandemic (June 2019 - March 2020) and pandemic (April 2020 - June 2020) periods from 1.2% to 1.8% (p<0.01).[10] Study by Nazir et al also showed increase from year 2018 to 2020 which was attributed to a decrease in the in-person patient care and reduced screening during lockdown.[9] Therefore, though social distancing during covid pandemic has the potential to decrease STI transmission, impact of covid on healthcare resources and interruptions of STI diagnosis and treatment drives it in the opposite direction.[11] Among Antenatal clinic low prevalence was observed with no significant changes. This points towards the major success of WHO strategies for elimination of mother to child transmission.[12]

The study was not without limitations. Being a hospital based study trends in seroprevalence of syphilis in a proportion of patients not reporting to the regional centre remains unknown. Analysis of trends in rural versus urban population was not undertaken. Association of syphilis with other STIs needs to be explored.

CONCLUSION

Findings of this study can help us understand the disease trends at a broader scale. The overall seroprevalence of syphilis showed decreasing trends in all patient groups pointing towards effective STD control programs. However, Covid pandemic has caused some disruptions in STD prevention and management strategies leading to surge in syphilis in HRGs. Whereas, though the prevalence among ANC attendees is low, since it has significant adverse effects on pregnancy in 80% of cases, therefore it is very important to continue to emphasize on screening of antenatal women for syphilis. Sexual health education, prophylactic use of barrier contraceptives, treatment of cases and sexual partners can further help in lowering the prevalence.

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