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It is commendable that the authors have tried to study the serum sample of such a large number of STD clinic attendees. However, a better-designed and interpreted study would have resulted in more useful information especially coming from such a leading centre. We hope our comments will be taken in the spirit they are meant to be.

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Response by the author

Sir,

With reference to the queries of Dr. Dogra and Dr. Kumar on our article "Serological study for sexually transmitted diseases in patients attending STD clinics in Calcutta", our comments on the various points raised by them are as follows:

1. As had been clearly mentioned in the Introduction, one of the main objectives of the study was "to ascertain the prevalence of syphilis, hepatitis B, chlamydia and HIV infection among patients attending different STD clinics in Calcutta".
2. Since the VDRL test is a low cost, rapid and a good screening test, all the samples were first subjected to qualitative tests and samples showing reactivity were then further subjected to a quantitative test. In fact, it has been clearly mentioned in the article, under Materials and Methods that "Quantitative test was performed on all reactive sera including those showing weak or rough reaction". Moreover, all the VDRL reactive sera, including all biological false positive sera were subjected to the TPHA test, which is a very specific test.

3. As mentioned under Materials and Methods, the TPHA test was performed using TPHA 200 kits, manufactured by Newmarket Laboratories Ltd., UK, strictly as per the manufacturer's instructions. If one goes through the literature of the kit in detail, it will be observed that the final dilution of the sample is 1:80. Hence this is again a quantitative test and not qualitative. This is the standard method for the TPHA test.
4. About the comment that adding up VDRL and TPHA tests "falsely increases total number of positive tests without any logical basis", it appears that the data has been misinterpreted. It must be clarified that Table V showing two tests positive, i.e. VDRL+TPHA, indicates that the same sample was positive for both VDRL as well as TPHA. So there is no question of increasing the total number.
5. Yes, we agree that IgM antibodies are more helpful in establishing acute chlamydia infections of the genital tract, but the present study was undertaken more from the epidemiological point of view. The values obtained from this assay are intended to be an aid for diagnosis only.
6. Table II clearly shows positivity for different serological tests among different age groups, infection being most commonly observed in the 15-30 years age group (20.13%), followed by the 30-45 years age group (12.69%) and the > 45 years age group (4.38%). It thus shows that the 15-30 years age group is the most vulnerable, and because of their risky behavior, this age group is susceptible to multiple infections.
7. Single sample positivity proves that only one infection was positive. All the serological tests, viz. HBsAg, TPHA, chlamydia-IgG, were done by kits, as per the manufacturers' instructions and guidelines. The HIV test was done at the School of Tropical Medicine, which is a NACO centre for HIV detection. All the sera were tested by the ELISA method, using kits from INNOTECH HIV-1/HIV-2 Sp. Innogenetics N.V., Belgium. ELISA reactives sera were confirmed

- by the Western Blot test, using kits from Innogenetics N.V., Belgium.
8. As we all know, without a cut off value no ELISA test is positive and the HBsAg, chlamydia and HIV tests are all ELISA tests. The TPHA test uses a dilution of 1:80 sample, which is a very specific test for syphilis. Cut off values are essential for any ELISA test and have been duly considered. As is well known, results cannot be calculated without a cut off value.
9. The Institute of Serology is the pioneer, Institute for the production of VDRL antigen. This institute initiated the production of cardiolipin antigen for serological tests of syphilis, with the assistance of WHO, in 1954. For the present study also, the methodology used was of a high standard and by using kits of highly reputed manufacturers, as indicated.

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