

**OBSERVATION ON SPERM AGGLUTININS IN THE SERA AND
CERVICAL MUCUS OF UNEXPLAINED CASES OF
STERILITY IN WOMEN***

by

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According to Shulman (1978) 10 to 30% cases of sterility remain unexplained on any organic or apparent basis.

Material and Methods

One hundred cases of unexplained sterility were selected from the gynaecological out patient Department of Patna Medical College. Detailed history was elicited from the couple. The obvious causes of sterility were excluded after thorough history clinical examination and investigations.

Sperm agglutinating test was performed in the sera of the women and 50 control cases according to the method of Franklin and Dukas (1964 A) and modified micro double diffusion agar precipitin test of Wardsworth (1957).

Sperm agglutination test: Sperm specimens were adjusted to a concentration of fifty million spermatozoa per ml. It was mixed with wife's serum and physiological saline for sperm agglutination test as detailed in Table I.

Test tube 3, which contained sperm and

physiological saline was used to exclude non-specific agglutination so that cases showing non-specific agglutination could be discarded.

The three test tubes were incubated at 37°C for four hours and the specimens were examined microscopically at intervals of 30 minutes during the test period. The aggregation of 4 to 10 motile sperm per high power microscopic field was considered as positive, aggregation of 10 to 20 sperm per high power field as strongly positive agglutination test. Aggregation of sperm around the cell debris was not taken into consideration.

In cases where the sperm agglutination tests were positive, the test was repeated taking 3 additional tubes with serum from fertile female donor and semen from fertile male donor. Sperm agglutination test was also performed in 50 pregnant women as control.

The cervical mucus agglutinins were tested by micro double diffusion agar precipitin test. In cases where the test was positive, it was repeated again with the fertile donor male and female.

There were 50 control cases who had delivered within six months and were menstruating normally in whom micro double diffusion agar precipitin test was performed.

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The results which were analysed according to age, duration of sterility, blood group incompatibility anti sperm antibody test of serum and cervical mucus are set out in Tables I to V.

Out of 100, 12 had positive agglutination test.

Out of 12 cases, 6 had head to head agglutination, 3 had tail to tail and 3 had mixed type of agglutination.

TABLE I

Test tube	Wife's serum ml.	Physiological saline	Husband's sperm 50 x 10 ⁶ ml.
1	0.5	Nil	0.05
2	0.5 (1:5) dilution	Nil	0.05
3	Nil	0.5	0.05

TABLE II

Sperm Agglutinating Antibody in Unexplained Cases of Sterility
(Franklin Dukes Method)

Serial No.	Total No. of cases	Positive agglutination	
		No. of cases	Percentage
1.	Test Group 100 cases	12	12%
2.	Control Group 50 cases	Nil	—

TABLE III

Nature of Agglutination

Total No. of cases	No. of positive cases	Nature of agglutination		
		Head to Head	Tail to Tail	Mixed
100	12	6	3	3

TABLE IV

Micro Double Diffusion Precipitin Test in Serum

Serial No.	Total No.	Positive	
		No. of cases	Percentage
1. Test Group	100	8	8%
2. Control group	50	Nil	Nil

TABLE V

Micro Double Diffusion Precipitin Test in Cervical Mucus

Serial No.	Total No. of cases	Positive	
		No. of cases	Percentage
1. Test Group	100	8	8%
2. Control Group	50	Nil	Nil

Eight out of 100 cases of primary sterility showed a positive precipitation band. No positive band was obtained in the control group.

Out of 100, 8 cases showed precipitation band in cervical mucus. None of the control cases, however, showed precipitation band in their cervical mucus.

Discussion

The present work consisted of 100 cases of primary sterility of unknown aetiology.

Age of the female partner ranged between 20 to 29 years. Age of the male partners ranged between 23 to 45 years. Duration of sterility varied from 2 to 16 years.

Twelve out of 100 cases showed positive test by Franklin and Dukes (1964) technique. Nature of agglutination was head to head in 6 cases, tail to tail in 3 cases and mixed type in the remaining 3 cases. In the present work, the sample which showed agglutination in control (Physiological saline) were discarded.

Micro double diffusion agar precipitin test was also performed in the same 100 women. Out of 100, 8 were positive by this method. None of the control showed positive test. Undiluted serum was used to avoid negative precipitation due to dilution.

In the present series, 8 out of 100 cases had positive test by the Franklin and Dukes (1964) and Micro double diffusion method. The remaining 4 cases were positive only by Franklin and Dukes (1964) method. This variation may be attributed to the different techniques used in the two methods, sensitivity of the test and different immunoglobulins involved. Cervical mucus hostility is another important factor for sterility. With this object, cervical mucus hostility in 100 women

were tested by post-coital test and cervical mucus antibody by micro double diffusion precipitin test. Eight out of 100 cases had positive anti sperm antibody test in cervical mucus. It is interesting to note that these women had also poor post-coital test.

On analysis of the result it was found that out of 8 women, 5 had positive test (62.5%) both in serum and cervical mucus and 3 had positive test in cervical mucus only. These antibodies against sperm may only be found at local sites without showing their presence in systemic circulation according to Sudo *et al* (1977) and Dondero *et al* (1978). It is apparent that out of 12 serum positive cases, 7 positive cases failed to show positive test in cervical mucus.

Presence of antibodies in the cervical mucus may be due to its predominance at local site. Alternatively, it may be present only in serum which reflects the dilution of secretion due to the systemic predominance of sperm antibody with little transudation in the genital tract. Friedman and Shulman (1974) studied a total of 56 cervical mucus samples from 38 women. They all had positive antibody in serum. Infertile women with antibody positive serum had positive cervical mucus in 82% by Franklin and Dukes test and 67% by Kibrick test.

There is unanimity of opinion regarding poor post-coital test and presence of antibody in the cervical mucus. However, the opinion differs regarding the correlation of antibody in the sera and poor post-coital test.

The presence of antibody had definite correlation with the longer duration of sterility and advancing age of the patients.

As regards blood group no correlation could be established regarding the ABO

blood group and anti sperm antibody test in the present series.

It has been observed that reduction in antibody titre occurred after the use of condom in cases with positive serum and cervical mucus antibody test. Pregnancy has also resulted after reduction in antibody titre. In the present series, cases are being followed but none of them have returned with pregnancy.

Summary

Twelve cases out of 100 had positive test in the sera by Franklin and Dukes method and 8 of these cases had positive test in the sera by Micro-double diffusion method, 4 of the 12 cases failed to show any positive test by the micro double diffusion method.

Eight cases out of 100 showed positive antibody test in the cervical mucus. Only

5 of these cases had positive antisperm antibody test in the sera as well as the cervical mucus.

On analysis of the result it is apparent that cervix is definitely a local site for antibody production and possible factor for sterility of unknown aetiology. The sperm agglutinins had, however, definite relation with advancing age and longer duration of sterility.

References

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