DISCUSSION

Despite all the emphasis on population control, infertility is certainly an important subject in its own rights. The infertile couple is viewed with ambivalence by the activist, who is concerned in utilizing every possible mechanism to reduce the population growth. However, humanistic view of protection of individual human rights dictates that barren couple are not to be denied the right to achieve their goal of parenthood.

Fertility has been one of the man's most desired attributes since the beginning of the recorded history. Fertility remains as a driving need for young couple today. In the past even on complete treatment of existing pathology, favourable results kept waiting and unexplained infertility in married couple still remains the crux of the problem of infertility.

When the couple had tried for at least one year to produce a pregnancy, and has been sure to include coitus at the time of each ovulation, they are defined by most workers as being infertile.
Immunological aspect of infertility helped the gynaecologist to know the exact etiology of unexplained infertility and also to provide the comprehensive treatment with better results.

Case where no cause can be attributed to the infertility, it is supposed that there is some auto-immune reaction between husband and wife which is responsible for infertility.

In considering the diverse mechanism that may operate in problems of spontaneous infertility, we can segregate a group of patients who are said to have "immune infertility". This term has been proposed to suggest that their fertility has been greatly reduced by the occurrence of antibody activity to spermatozoon.

We may expect that this concept surely become broadened in the future, to include antibodies to human ova. This will be based in part on the existing findings of Shiverse and colleagues with regard to the occurrence of acute antibodies to the zona pellucida and the preliminary findings that some women may well have such antibodies to their own zona (Shiverse and Dunbar, 1977).
Ten to thirty percent infertility is estimated to be explained on any organic or apparent basis (Shulman, 1972). Investigation indicate that immunological incompatability may be a causative factor in about 20% of couple whose infertility is otherwise unexplained.

There are two main cause of infertility as such

1. Presence of antibodies against spermatozoa in women.


In the female antibodies are present in the genital tract due to local immune reaction against spermatozoa, or may be present in the serum because of systemic immune-reactions.

The sperm immobilizing phenomenon seen in the cervical mucus have been known since Sim's era. However, the microscopic agglutination of sperm in the cervical mucus has never been described or demonstrated before 1974, when Shulman demonstrated sperm agglutinating activity in extracts of cervical mucus from infertile women.

A recent report of Coeling (1974), Bennink and Menge (1974), stated that 'astonishingly' few
studies have been published on the detection of sperm antibodies in the secretion of genital tract fluids and obtained positive result of sperm agglutination in cervical mucus extract by microscopic agglutination test.

Keeping in view all these researches and interesting results, present study was undertaken. Only 'sperm agglutinating antibody' study has been done in case of unexplained infertility because it is much better established phenomenon and yet still creates controversy and confusion in some quarters.

For the purpose of discussion, a total of 106 women were studied. They were divided into two main groups.

Group I - included total number of 10 women of known fertility.

Group II - included 96 women having unexplained infertility (Table I).

Group II or study group has been further divided into two sub-groups depending on type of infertility.

A. Primary infertility - included 86 women.

B. Secondary infertility - included 10 women
(Table II).
The cases were thoroughly investigated to exclude any organic cause for infertility.

Age of women varied from 18 - 40 yrs. Majority of cases were present between the age group 21 - 30 yrs (Table III). The duration of infertility ranged between 2 - 20 yrs, most of the cases had duration of infertility between 6 - 10 yrs (Table IV).

Sperm agglutination test was done in all 106 women in serum by Franklin Dukes method and in cervical mucus by microscale test of cervical mucus extract (tube slide agglutination technique). Result observed in different group was as follows.

A. Control group -

(1) Sperm agglutinating activity in serum -

In the present series none of the 10 women of control group showed sperm agglutinating activity in serum (Table VI).

Franklin & Dukes (1964 a) reported the incidence of 11.8% sperm agglutinating activity in sera of control group. Isojima et al (1972) observed the incidence of 45.8%, Jones et al (1973 a, 1974) 18%.
Shulman et al (1975) 2.6%, Ingerslev (1980) reported the incidence of 1.7% in their control group.

Table XVII

Incidence of sperm agglutinating activity in serum of control group by various workers.

<table>
<thead>
<tr>
<th>Name of worker</th>
<th>Fertile group %</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Franklin and Dukes (1964 a)</td>
<td>11.6</td>
</tr>
<tr>
<td>2. Isojima et al (1972)</td>
<td>45.8</td>
</tr>
<tr>
<td>3. Jones et al (1973)</td>
<td>18.0</td>
</tr>
<tr>
<td>4. Shulman et al (1975)</td>
<td>2.6</td>
</tr>
<tr>
<td>5. Petrems et al (1976)</td>
<td>10.4</td>
</tr>
<tr>
<td>6. Ingerslev (1980)</td>
<td>1.7</td>
</tr>
</tbody>
</table>

Most of the Indian workers reported absence of antibodies in their control group (Hingorani et al, 1978; Mukerjee et al, 1978; Nanda and Pnigrahi, 1978; Upadhyay et al, 1979; Misra et al, 1984), while Joshi et al (1978) and
Misra et al (1981) reported an incidence of 3.3% and 2.8% respectively.

(ii) Sperm agglutinating activity in cervical mucus -

In the present study, none of the 10 women of known fertility showed the sperm agglutinating activity in cervical mucus (Table XI).


B. Study group (unexplained infertility)

(i) Sperm agglutinating activity in serum of study group -

In the present series sperm agglutinating activity in sera was present in 11.46% of cases having unexplained infertility (Table VI).
Table XVIII

Incidence of sperm agglutinating activity in sera of women having unexplained infertility.

<table>
<thead>
<tr>
<th>Name of worker</th>
<th>Unexplained infertility %</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Franklin and Dukes (1964 a)</td>
<td>78.9</td>
</tr>
<tr>
<td>3. Vaidya and Glass (1971)</td>
<td>20.0</td>
</tr>
<tr>
<td>4. Isojima et al (1972)</td>
<td>37.5</td>
</tr>
<tr>
<td>5. Jones et al (1973)</td>
<td>26.8</td>
</tr>
<tr>
<td>6. Mukerjee et al (1973)</td>
<td>19.0</td>
</tr>
<tr>
<td>7. Shulman et al (1975)</td>
<td>16.4</td>
</tr>
<tr>
<td>8. Petrunia et al (1976)</td>
<td>17.6</td>
</tr>
<tr>
<td>9. Mettler (1977)</td>
<td>16.8</td>
</tr>
<tr>
<td>13. Ingerslev (1980)</td>
<td>6.3</td>
</tr>
</tbody>
</table>

Our values in unexplained group were almost consistent with Shulman et al (1975) 16.4%, Petrunia et al (1976 19.6%, Mettler et al (1977) 16.8%.

A positive reaction of sperm agglutination has been carefully defined in several reports by Boettcher and Hay (1969), Kolodny et al (1971), Shulman et al (1975) and Mettler (1977), but they had used different criteria. Thus, Boettcher et al (1971) counted the number of agglutinates, per 100 motile, freely moving, non-agglutinated spermatozoa and accepted at least 6/100 as a positive reaction, whereas Shulman et al (1975) related the number of spermatozoa involved in the agglutinates to the total number of cells (agglutinated and non-agglutinated) and required that at least 10% of the spermatozoa are involved in the agglutinates.

Several investigation have been aware of the risk of non-specific agglutination of spermatozoa around leucocytes, cellular debris and dead sperm cells (Schwimer et al, 1967a; Boettcher and Hay, 1969; Israelstam, 1969; Glass and Vaidya, 1970; Jones et al, 1973a; Petrunia et al, 1976). The hazard of false positive reactions has been reduced in some studies by including known negative control sera in each experiment (Israelstam, 1969; Glass and Vaidya, 1970; Vaidya and Glass, 1971; Kolodny et al, 1971).
Although all sexually active women are exposed to a relatively extensive load of sperm antigens, few seem to develop humoral immunity against spermatozoal surface antigens. Hancock (1973) has presented several possible explanations against spermatozoa in women.

1. Spermatozoa are flushed down the female genital tract by secretions (Austin, 1976), reducing the number that gain success to the immune system.

2. Certain seminal plasma components seem to have an immune-suppressing effect (Lord et al., 1977).

3. Extensive phagocytosis of spermatozoa in the female genital tract (Moyer et al., 1970; Austin, 1976) may degrade the spermatozoa, reducing their immunogenicity.

The limited number of immune reactions in women exposed to spermatozoa may also be due to insufficient amount of antigen resulting in low non-tolerance rather than in antibody production.

The higher incidence observed by various workers could be explained by various reasons. It seems likely that genital infections or epithelial lesions of various kinds may act as
triggering factors for local and subsequently systemic immune reactions against spermatozoa.

A significantly higher frequency of sperm-agglutinating activity in sera has been found in prostitutes compared with normal fertile women (Schwimmer et al, 1967b; Kolodny et al, 1971). This may reflect either a more extensive exposure to sperm antigens or a higher occurrence of genital infections.

Incidence of positive sperm agglutination in cases of primary infertility was 11.62%, while in secondary infertility 10% in sera (Table VII). Incidence shown by various workers in primary and secondary infertility was shown in Table XIX.

Table XIX

Incidence of sperm agglutinating activity in serum of primary and secondary infertility.

<table>
<thead>
<tr>
<th>Name of workers</th>
<th>Primary infertility</th>
<th>Secondary infertility</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Ansbacher et al (1971)</td>
<td>20.4</td>
<td>6.0</td>
</tr>
<tr>
<td>2. Hingorani et al (1978)</td>
<td>13.8</td>
<td>8.8</td>
</tr>
<tr>
<td>4. Upadhyay et al (1979)</td>
<td>28.6</td>
<td>16.0</td>
</tr>
<tr>
<td>5. Mishra and Das (1981)</td>
<td>10.0</td>
<td>-</td>
</tr>
</tbody>
</table>
Our present series is very small to give any conclusion regarding the incidence of sperm agglutinating activity in primary and secondary infertility.

To establish the sensitivity of the test sperm agglutination test was also observed in diluted sera and agglutination activity was also recorded at different hours. In cervical mucus dilution was not done because of little amount of cervical mucus, sera had been tested in dilution by several investigators because of risk of non-specific reaction with the undiluted sera (Shulman, 1975).

(ii) **Sperm agglutinating activity in cervical mucus of study group**

In the present series sperm agglutinating activity in cervical mucus was present in 27.29% of women having unexplained infertility (Table XI).

Parish et al (1966) have demonstrated antisperm antibodies in 3 out of 11 patients (27.27%). Parish and Ward (1968) have demonstrated antisperm antibodies in 3 out of 48 cases (6.25%). Eyques and D'Almeida (1973) demonstrated antisperm antibodies in cervix mucus in 20% cases. Coelingh et al (1974)
demonstrated positive result in 3 out of 13 patients (23.77%). They supported the concept of local production of antibodies.

Shulman et al (1975) demonstrated the incidence of 15-30% sperm agglutinating activity in cervical mucus of women having unexplained infertility by tube slide agglutination techniques.

Sudo et al (1977) demonstrated the sperm agglutinating activity in 2.7% of cases. Ingerslev and Hjort (1979) demonstrated the antisperm antibodies in 3.8% of cases and Moghissi et al (1980) in 16.3% of cases having unexplained infertility. Mukerjee et al (1984) reported the incidence of 64% positive sperm agglutination in cervical mucus by MIS test. Misra et al (1984) reported the positive sperm agglutination in cervical mucus in 8% of cases having unexplained infertility (Table XX).
Table XX

Incidence of antisperm antibodies in cervical mucus reported by various workers.

<table>
<thead>
<tr>
<th>Name of workers</th>
<th>Unexplained infertility positive cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Parish et al (1966)</td>
<td>27.27</td>
</tr>
<tr>
<td>2. Parish and Ward (1968)</td>
<td>6.225</td>
</tr>
<tr>
<td>3. Eyquem &amp; D Almeida (1973)</td>
<td>20.0</td>
</tr>
<tr>
<td>5. Shulman et al (1975)</td>
<td>15 - 30%</td>
</tr>
<tr>
<td>7. Ingerslev (1979)</td>
<td>5.8</td>
</tr>
</tbody>
</table>

The variation in results could be explained on the basis of different techniques used to detect the sperm antibodies and presence of concurrent infections, because several bacterial species are spermicidal in vitro (Mathews & Buxton, 1951; Buxton et al, 1954; Kay et al 1954; Teague et al, 1971), and production of antibodies in cervical mucus following infection has been demonstrated by various workers (Kerr, 1955; Bell and Wolf, 1967; Omran and Hulka, 1971; Wilkie et al, 1972; Corbeil et al, 1974b; Yang & Schumacher, 1979).

The cervix seems to be the probable origin of local and systemic immune reactions against spermatozoa: (i) The cervix appears to contain a local immune system and the immune potential is higher than in any other segment of the female genital tract, as evaluated by immune-histological investigation and experimental immunization studies.

(ii) The cervix is exposed to a significantly greater number of spermatozoa and accordingly to a more extensive antigenic load, than higher part of the female genital tract (Ahlgren, 1969).

A significantly rising incidence of sperm agglutination in cervical mucus was observed with
increasing duration of infertility (Table XII), justifying the criteria that cervix is exposed to the greater number of antigenic spermatomes and also for longer period in these cases.

Incidently same pattern of rising incidence of sperm agglutinating activity in serum was also observed with increasing duration of infertility (Table VIII). Mishra et al (1981), Mukerjee et al (1984), Misra et al (1984) also observed that presence of antibodies had definite correlation with duration of infertility. With increasing duration of infertility, higher prevalence of positive cases have been demonstrated. However, no significant difference was observed in the incidence of positive cervical mucus result in primary and secondary infertility (Table XI).

**Correlation between sperm agglutinating activity in serum and cervical mucus**

(1) Agglutinating activity in cervical mucus in cases with positive sera -

Table XV indicates, majority of cases with positive serum showed positive result in the cervical mucus. Sperm antibodies in serum probably appears as a
consequence of sperm antigen being exposed to the immune system in the genital tract. It was therefore reasonable to believe that antibodies should be present concomitantly in serum and genital secretions. Shulman (1974) studied a total of 56 cervical mucus sample from 38 women who had positive antibodies in serum. They observed that infertile women with antibody positive serum had positive cervical mucus in 82% by F.D. test and 67% by Kibrick test.

However, several reports have demonstrated that only 40-60% of women with sperm agglutinating antibodies in serum have detectable antibodies in cervical mucus (Shulman and Guerrere, 1978; Maghissi et al, 1980; Ingerslev, 1980).

It is not likely that women with positive serum but negative cervical mucus, are immunized in the higher part of genital tract where immune potential is low. It is also unlikely that few spermatozoa entering the peritoneal cavity during mid cycle (Ahlgren, 1969) constitutes an insufficient stimulus to stimulate a systemic immune reactions.

The apparently shorter persistence of local immune reactions compared to systemic immune reactions
(Ogra and Karson, 1969; Rossen et al, 1970; Couch et al, 1971; O'Reilly et al, 1976), is more obvious explanation for negative test in cervical mucus from women with circulating sperm antibodies.

Another explanation could be that, the spontaneous cure of "triggering" condition like infection may lead to cessation of contact between sperm antigen and immune system, despite continued sexual activity. The local antibodies may subsequently disappeared, whereas, the antibodies in serum probably persist for much longer period.

Aghren (1969) suggested that few spermatozoa reaching the peritoneal cavity during the mid cycle may maintain systemic immune reactions once this has been established even in absence of local reaction.

(ii) Agglutinating activity in sera in cases with positive cervical mucus -

Table XVI shows that out of 26 women with positive sperm agglutinating activity in cervical mucus, 9 women had concurrent sperm agglutinating activity in serum (34.61%).

According to Sudo et al (1977) and Donadro et al (1978) antibodies against sperm may
only be found at local sites without showing their presence in systemic circulation.

Ingerslev (1980) investigated cervical mucus from 21 infertile women with sperm agglutinating antibodies in serum by applying the tray agglutination technique to bromaline treated cervical mucus, 8 of the women (38%) had concurrent sperm agglutinating antibodies in ovulatory cervical mucus.

Moghissi et al (1980) found negative serum in 22 of the 28 women with sperm agglutinating activity in cervical mucus with an incidence of only 21.4% positive result. Explanation for this could be that they tested the cervical mucus by more sensitive tray agglutination technique.

Sperm antibodies in serum probably appears as a consequence of sperm antigens being exposed to the immune systems in the genital tract. It was therefore reasonable to believe that antibodies should be present concomitantly in serum and genital secretions.

Correlation of antisperm antibodies with post coital test -

It was observed that poor, sluggish and no motility of sperm was associated with positive
sperm agglutination in cervical mucus. This suggest that sperm lose their power of motility due to sperm agglutinating antibodies in the cervical mucus. Therefore a poor post coital test could be taken as an indication for the study of antisperm antibodies.

Parish and Ward (1968) observed the same result and assigned the same significance to the study of antisperm antibodies in case with poor post coital test. However, Wall et al (1975) stated that poor result on post coital test were due to some factor, other than spermatozoal antibodies. Sinha et al (1977) suggested local immune reaction as a cause of poor post coital test and infertility.

Several subsequent investigations by Kremer and co-workers reported the relationship between the presence of sperm agglutinating antibodies in cervical mucus and poor post coital with poor sperm penetration test (Kremer and Jager, 1977; Kremer et al, 1977; Kremer et al, 1978 as Hansen & Hjort, 1979; Moghissi et al, 1980; Ingerslev, 1980). They demonstrated that sperm agglutinating antibodies in cervical mucus or in seminal plasma may cause infertility by impeding sperm penetration in cervical mucus.
Kremer and co-workers (1977 & 1980) have described 7 women with sperm agglutination antibodies in cervical mucus, all had poor post coital test.

Ingerslev (1980) observed lower titers and less affected sperm motility in cervical mucus. The apparent discrepancy between their result probably due to different selection of patients.

In most cases with a so called unexplained poor post coital test, occurring in the presence of optimal ovulatory cervical mucus and normal sperm sample, the cause is probably antibodies against spermatozoa. This evidence is further supported by Kremer et al (1978) and Moghissi et al (1980). They demonstrated that the number of good or excellent post coital test declined notably in the presence of sperm antibodies in cervical mucus in couples with normal cervical mucus and normal sperm sample. A negative or poor post coital test was observed in cervical mucus from 12 out of 18 women (66%) with sperm agglutinating antibodies in cervical mucus and in 9 and 11 (62%) with sperm immobilizing activity in the cervical secretions (Moghissi et al, 1980).
Regarding the pattern of agglutination there were either head to head, tail to tail and mixed type or combination depending upon the site of antigen present over the surface of spermatosoa. Glass and Vaidya (1971) have shown head to head pattern, as the most common pattern. Shulman (1973) showed tail to tail type to be most frequent. Mishra and Dass (1981) have shown all the three type of agglutination in their observation. In our study, head to head pattern was observed more commonly in cervical mucus as well as in serum (Table X & XIV).

From all these results, it appears that agglutination of any part of sperm may be taken as reliable index for presence of sperm agglutinating antibodies.

All the cases with positive sperm agglutinating antibodies were advised treatment with a view to destroy the antibodies and to prevent the further immunisation. Women with positive sperm agglutination test in sera and cervical mucus advised condom therapy along with Corticosteroids for 6 wk to 3 months. After stoppage of steroid therapy, patients were kept on estrogen therapy, Tab. ethinyl estradiol 50 μg O.D. from 5th to 25th day of period for 3 months.
The infertile couples having antisperm antibodies were kept on condom therapy from 2 - 12 months interval by Franklin and Dukes (1964), Schwimmer et al (1967), Glass & Vaidya (1970), Kolodny et al (1971), Mukerjee et al (1973), Jones et al (1976) and Srivannaboon et al (1982). They reported that significant number of women become pregnant subsequent to the resumption of unrestricted coitus. Ingerslev (1980) reported the significance decrease titer of sperm antibodies in cervical mucus with oestrogen therapy.

In the present series, cases are being followed but none of them have returned with pregnancy.

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