T-MYCOPLASMA COLONISATION IN GENITAL TRACT AND INFERTILITY

by

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Abstract

Fifty couples who were married for 2 to 10 years were studied. Thirtyone (62%) females and 21 (42%) males showed mycoplasma colonisation in their genital tracts. Tetracycline therapy 250 mg 6 hourly for 10 days eradicated Mycoplasma in all but one couple. No substantial changes were noted in semen parameters after treatment except postcoital test which showed improvement. Five patients have conceived so far, 4 out of these 5 were after treatment with tetracycline. These findings are of great interest and certainly seem to justify further investigation.

Introduction

No major cause of infertility can be demonstrated in either husband or wife in about 10 to 20% of all infertile couples even after careful investigations (Southam, 1960). Recent studies have shown a trend towards more spontaneous abortions, stillbirths, infertility and low birth weight babies in women whose genital tracts were colonised with T-mycoplasma (Capsi *et al*, 1971; Kundsin and Driscol, 1970; Gnarp and Friberg, 1973). T-mycoplasma have been shown to occur with higher frequency in the ejaculate and cervical secretions of couples with unexplained infertility (Gnarp and Frigerb, 1972; Friberg and Gnarp, 1974). In addition, Gnarp and Friberg (1973) have suggested that T-mycoplasma might play a role in human reproductive failure, since pregnancies were achieved in approximately 30% of infertile couples after eradication of T-mycoplasma by antibiotics. Kapur *et al* (1976) reported the isolation of T-mycoplasma in human genital tract infection. The present work was carried out to study the role of T-mycoplasma (ureaplasma urealyticum) in human infertility with following objectives:

(i) Incidence of genito-urinary tract colonisation in fertile and infertile males and females.

(ii) Correlation of male genital tract colonisation with seminal parameters.

(iii) Therapeutic response to tetracy-

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cline in infertile couples harbouring T-mycoplasma.

Material and Methods

Among the patients who were attending the sterility clinic of the All India Institute of Medical Sciences Hospital, New Delhi from September 1975 to May, 1976, 50 couples who were infertile for 2 to 10 years were selected for the study. Forty-one out of 50 couples had not conceived at all and 9 had past history of premature deliveries or first trimester abortions. Detailed history was taken and thorough clinical examination was done in both the partners. Apart from routine investigations of blood, urine, x-ray chest and serological test for syphilis, special investigations like endometrial biopsy in premenstrual phase of the cycle, tubal patency test, post coital test and sperm analysis were done for all the cases under our study. Hysterosalpingography and antisperm antibodies test were done in few cases. Swabs from cervical canal were collected from female partners and 10 ml of urine from the male partners after prostatic massage for culture of T-mycoplasma on Shepard's uq colour test medium, Sbroth and A3 or A7 Agar plates. Growth of T-mycoplasma was recognised by the change in colour in uq medium to pink colour or typical colonies on agar. Blood was collected from female partners for antibody detection.

Control

Cervical swabs and blood for culture and antibody detection were collected from (1) 28 fertile non-pregnant patients who were having more than 2 children and attending the postnatal clinic of AIIMS. (2) Thirty fertile pregnant multiparas attending antenatal clinic of AIIMS. (3) Urine (after prostatic massage) 10 ml was collected from 25 fertile males (Husbands of the women for controls) for T-mycoplasma culture (Table I).

TABLE I

Sources of Specimens in Males and Females in Fertile and Infertile Group

Study Group	No. Studied	Specimen Cultured for T-Mycoplasma Cervical Swab	Urine Deposit
A. MALES			
Infertile	50		50
Fertile	25		25
B. FEMALES			
Infertile	50	50	
Fertile non-pregnant	28	28	
Fertile pregnant	30	30	

All the cases were followed up to detect any other demonstrable cause for their infertility and those couples from whose urine or cervical swabs 'T' mycoplasma were isolated, were given a course of tetracycline therapy (250 mg 6 hourly for 10 days) or doxycyline for 10 days. Treatment was given in the first part of the menstrual cycle. Urine and cervical swab culture were repeated after a month of the above therapy and followed every two months subsequently to see if 'T' mycoplasma had been eradicated completely and consequences of the therapy were noted. Seminal parameters

of the infertile males were studied and the effect of tetracycline therapy on these were observed.

Results and Observations

Table II shows the incidence of T-mycoplasma colonisation in the female genital tract in the women under our study. High incidence of positive culture for T-mycoplasma were seen in infertile females (62%) compared to control cases (21.0% and 37% respectively) which is statistically significant ($P < .01$).

TABLE IV

Incidence of T-Mycoplasma in Infertile Couples

Results	No. of Cases	Percentage
Both partners positive	20	40
Wife +ve and Husband —ve	10	20
Wife —ve and Husband +ve	1	2
Both partners negative	19	38

TABLE II

Incidence of T-Mycoplasma Colonisation in Fertile and Infertile Females

Study Group	No.	T-Mycoplasma No.	Positive %	Culture No.	Negative %
Infertile	50	31	62.0	19	38.0
Fertile non-pregnant	28	6	21.0	22	79.0
Fertile pregnant	30	11	37.0	19	63.0

Table III shows the incidence of T-mycoplasma colonisation in the male genito-urinary tract under our study. Incidence was high in infertile males, (42%) more than 5 times that in fertile males (8%) which is statistically highly significant ($P < 0.01$).

TABLE III

Incidence of T-Mycoplasma Colonisation in Fertile and Infertile Females

Study Group	No.	T-Mycoplasma No.	Positive %	Culture No.	Negative %
Infertile	50	21	42.0	29	58.0
Fertile	25	2	8.0	23	92.0

Table IV shows incidence of T-mycoplasma in 50 infertile couples. Thirty-one couples were harbouring T-mycoplasma, 25 couples were given a course of Tetracycline but only 19 could be followed up for a maximum period of 9 months.

Table V shows the causes of infertility and T-mycoplasma colonisation in 50 couples studied. In 29 cases (58%) some cause could be ascertained while no known cause was found in the remaining 21 (42%). The unknown group showed high incidence of T-mycoplasma (71.5%) in comparison with the known group (55.2%).

Tables VI and VII show the correlation between the T-mycoplasma conolisation in genito-urinary tract of the males under study and their sperm motility

TABLE V
Causes of Sterility and T-Mycoplasma Colonisation in 50 Infertile Couples

Causes of Sterility	No.	T-Mycoplasma Positive	Culture Negative
Tubal Block	15	8	7
C.L. Deficiency	1	0	1
Azoospermia	7	4	3
Oligospermia	5	3	2
Obstruction of vas	1	1	0
Total known causes for infertility	29 (58%)	16 (55.2%)	13 (44.8%)
Unknown	21 (42%)	15 (71.5%)	6 (28.5%)

TABLE VI
T-Mycoplasma and Sperm Motility in Infertile Males

Culture	No. of Cases	Sperm Motility					
		<40		41 to 70		70	
		No.	%	No.	%	No.	%
Positive	21	4	19	9	43	8	38
Negative	19	2	11	9	47	8	42

TABLE VII
T-Mycoplasma and Sperm Count in Infertile Males

Culture	No. of Cases	Sperm Count in Million/Ml					
		<20		21 to 40		40	
		No.	%	No.	%	No.	%
Positive	21	5	24	1	5	15	71
Negative	19	3	16	4	21	12	63

and count. Sperm motility less than 40% and sperm count less than 20 million/ml was seen more often in patients whose genital tract were colonised with T-mycoplasma.

Table VIII shows the correlation between post-coital test and mycoplasma colonisation. Post-coital test was twice as good in the negative mycoplasma group compared to those who were positive for

TABLE VIII
T-Mycoplasma and Post-Coital Test in Infertile Couples

Culture	No. of cases	Post-coital Test			
		Good		Fair/Poor	
		No.	%	No.	%
Positive	31	7	23	24	77
Negative	19	8	42	11	58

T-mycoplasma (42%) and 22.5% respectively). The criteria for good, fair and poor, post coital test were as follows.

Good P.C.T.

10 actively motile sperm/HPF

Fair P.C.T.

5 to 10 actively motile sperm/HPF

Poor P.C.T.

5 actively motile sperm/HPF

Besides the above criteria, amount of cervical mucus, spinnbarkeit and cellularity were taken into account. Good P.C.T. showed abundant cervical mucus and nil to occasional pus cells, whereas poor P.C.T. was characterised by scanty mucus and plenty of pus cells.

Tables IX, X and XI show the effect of tetracycline therapy on sperm count, motility and post coital test. There was no significant improvement noted in count and motility of spermatoza after tetracycline treatment but P.C.T. improved in 5 patients under our study. Three out of these 5 patients later conceived and delivered viable and healthy children.

TABLE IX

Tetracycline Therapy and Sperm Count

Sperm Count	No. of Cases		No. of Cases Improved
	Before Treatment	After Treatment	
Normal	9	10	0
Oligospermia	2	1	1
Azoospermia	4	4	0

TABLE X

Tetracycline Therapy and Sperm Motility

Sperm	No. of Cases		No. of Cases Improved
	Before Treatment	After Treatment	
Active	6	7	0
Moderately active	6	5	1
Sluggish	3	3	0

TABLE XI

Tetracycline Therapy and Post-coital Test

Post-coital Test	No. of Cases		No. of Cases Improved
	Before Treatment	After Treatment	
Good	2	4	0
Fair	6	7	2
Poor	7	4	3

Discussion

There has been a lot of controversy regarding the exact role of T-mycoplasma in infertility. According to Gnarp and Friberg (1972) 'T'-mycoplasma might be responsible for reproductive failure as 'T'-mycoplasma were isolated in high frequencies, 85% compared to the control females 23% and males 26%. In this study it was found that the colonisation was positive for 62% of cases compared to 21% and 37% in control cases, the difference being statistically significant. But Louvis *et al* (1974) found the incidence of T-mycoplasma colonisation not significant between fertile and infertile couples. Tayler Robinson *et al* (1969) found high frequency of this micro-organism in the genital tract of normal women and thus refuted the role of mycoplasma as a possible cause of infertility, though higher incidence of mycoplasma has been isolated from infertile couples, yet the possible underlying mechanism for reproductive failure is not known.

High incidence (68.4%) mycoplasma isolation has been reported (Mardth and Westrom, 1970) in fertile pregnant women compared to fertile non-pregnant women. A little higher incidence was noted in our study 37% and 21% respectively. The above authors have also shown that the incidence of colonisation is more in women taking oral contraceptives but low in prepubertal girls and postmenopausal women. It seems that hormones, oestrogen and progesterone have some role to play, in colonising and growth of T-mycoplasma. The other possibility that the change of pH of vaginal secretion during pregnancy might be congenial for the growth of mycoplasma. It is possible that only a limited number of 'T'-mycoserotype are of importance in cases of reproductive failure, which needs further investigations.

In seminal parameters, sperm motility is more important for the conception to occur (Mecleod and Gold, 1951). Pregnancy can occur with low count but good motility. Hence it seems that if mycoplasma has got any role on seminal parameters it is on the motility. In this study high motility (70%) was seen in 38.1% positive group and 42.1% in negative group for mycoplasma respectively. But there was no substantial difference in sperm count. Gnarp and Friberg (1973) and Fowlker *et al* (1975) have reported a close physical association between the spermatozoa and T-mycoplasma. It has been reported by Gnarp and Friberg (1975) that the infertile patients with T-mycoplasma infestation had an overall decrease in seminal quality compared to those lacking demonstrable T-mycoplasma. Seminal cytology in a group of 678 infertile men was distinctly poorer in those harbouring T-mycoplasma (Fowlker *et al*, 1975). It has been proved that the T-mycoplasma adhere to the middle piece of spermatozoa which can be seen by electron microscopy and this perhaps impairs the motility by interfering with normal metabolism of spermatozoa. T-mycoplasma which produce neuraminidase like substances may interfere with fertilisation and development of fertilised egg (Gnarp and Friberg, 1972). Because pregnancies could be inhibited in animal by neuramindase (Gasic and Gasic, 1970). If this attachment were permanent its influence would extend into the female reproductive tract. Horne *et al* (1973) have demonstrated an inflammatory reaction of the endometrium in association with positive T-mycoplasma culture of the cervix and thus is responsible for reproductive failure. Tetracy-

cline therapy eradicated T-mycoplasma in all cases except one who needed second course of therapy. Five patients got pregnant within 6 months, out of which 4 were sometime after treatment when the cervical swab and urine were negative for T-mycoplasma. One patient was irregular in her follow up and we do not know if she had any antibiotic therapy systemic or local during that time. Thus 21% of our patients conceived within six months after tetracycline therapy which is very near to the incidence reported by Friberg and Gnarp, 1973 i.e. 25%. These findings are of great interest and certainly seem to justify further investigations.

Thus T-mycoplasma appears to be a possible aetiological factor in human infertility. However, this needs to be proven by a larger study.

Summary

1. No other cause of infertility apart from mycoplasma could be found in twen'yone (42%) of the 50 infertile couples studied.

2. T-mycoplasma colonisation of the genito-urinary tract was higher in infertile cases as compared to fertile control.

3. P.C.T. was poor or fair in twice as many couples colonized as compared to those not colonized with T-mycoplasma.

4. Tetracycline therapy

(a) Eradicated T-mycoplasma in all except one case.

(b) Improved sperm count, motility 6% and P.C.T. in 33.33% cases.

(c) Resulted in 4 pregnancies.

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