SUBSEQUENT PREGNANCIES AMONG 161 COUPLES TREATED FOR T-MYCOPLASMA GENITAL-TRACT INFECTION

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THE role of T mycoplasma *(Ureaplasma urealyticum)* in reproductive failure is controversial. Since mycoplasma was first isolated from a genital-tract abscess,[1] an association between mycoplasma and various genital-tract conditions, reproductive failure, and relative or absolute infertility has been suggested. Several investigators have shown that the relative frequency of T-mycoplasma colonization in the genital tract depends on the sexual experiences of the subject[2,3] and that infertile couples tend to have higher colonization rates.[4] Several claims were made previously that by eradicating the organism, doxycycline improved fertility and resulted in higher pregnancy rates in a group of couples with unexplained infertility.[5] These findings were subsequently refuted by several other investigators who did not find a close correlation between doxycycline therapy and fertility.[6,7]

For the past three years our clinic has systematically cultured semen specimens from the husband and recommended therapy for both husband and wife when the husband was found to have a T-mycoplasma infection of the genital tract. The purpose of this paper is to present the results of a three-year follow-up study comparing pregnancy rates in women whose husbands' T-mycoplasma infections were successfully eradicated, as demonstrated by a negative post-therapy culture, with the rates in women whose husbands' infections were not eradicated by treatment. The rate of successful pregnancies after therapy was 60 per cent for the group in which T mycoplasma was eradicated (n = 129) and 5 per cent for the group in which it was not (n = 32). Our data suggest that T-mycoplasma infection in the genital tract, with or without other bacteria, may have an important role in infertility.

METHODS

One hundred sixty-one men with T-mycoplasma infection in their seminal fluid were treated, along with their wives, for four weeks with doxycycline (100 mg twice a day). Irrespective of the semen quality, the criteria for patient selection were a positive mycoplasma culture at the beginning of the study and the intent to conceive during the observation period. The seminal fluid was produced by masturbation after a three-day abstinence period. Part of the specimen was submitted for mycoplasma culture and another portion for routine semen analysis, as described previously.[8,9] Specimens were cultured for mycoplasma for one or more of the following reasons: a referring physician's request, a history of miscarriages, a history of genital-tract infection in either the man or the woman, or generally poor semen quality — especially suppressed motility. The variables examined in the analysis were age of the woman (≤35 years or >35), occupation of the husband (blue or white collar), order of marriage (first, second, or more), presence of children in current marriage, length of marriage, length of time spent trying to conceive, other fertility procedures performed (hysterosalpingogram, tuboplasty, myomectomy, dilation and curettage, treatment with fertility drugs), seminal-fluid motility (classified as either poor or good on a scale described elsewhere[10]), and sperm count (≤10 million, 10 million to 60 million, or >60 million). At the end of the therapy, the man's mycoplasma status was reclassified as either positive or negative; the mycoplasma status of the woman was not evaluated. At the end of the follow-up period, the records were updated by direct telephone contact with the patients and by reviewing the medical records of the referring physicians.

Statistical Analysis

Relations among variables were examined by means of the chi-square test or Fisher's exact test for contingency tables, as appropriate. The product–limit method[11] was used to estimate the proportion of women who were pregnant at a given time. The log-rank test[12] was used to determine whether there were differences in the distributions of time until pregnancy among different groups of patients. Data related to time until pregnancy were summarized in terms of the three-year pregnancy rates; the log-rank test was used to compare the pregnancy "experience" (i.e., the distribution of time until pregnancy) of two or more groups but not specifically to compare three-year rates. A multivariate analysis was carried out with Cox's regression model,[13] to determine what combination of variables best predicted the amount of time until successful pregnancy and the order of importance among these variables. (These three methods are commonly referred to as methods of "survival analysis," since they are frequently used to analyze data related to survival.)

In all analyses involving the time until pregnancy the interval measured was the time from the last determination of mycoplasma status until a miscarriage or until the last menstrual period that directly preceded a successful pregnancy, whichever came first. (A "successful pregnancy" was defined as the delivery of a viable infant at between 38 and 42 weeks of gestation.) If a woman had neither a miscarriage nor a pregnancy during this period, the time until the most recent follow-up contact was recorded. Consequently, a woman's post-therapy status was classified as either successful pregnancy or no successful pregnancy. Since the outcome of interest was a successful pregnancy, the intervals for patients falling into the second category were treated as "censored."

RESULTS

Of the 161 men whose semen cultures were positive for T-mycoplasma infection before therapy, 129 (80 per cent) had negative cultures at the conclusion of therapy.

Table 1 shows the distribution of variables studied and the proportion of patients whose specimens were negative for mycoplasma after antibiotic therapy. There was a significant association between post-therapy pregnancy status and post-therapy mycoplasma status ($P<0.001$). Among the couples who eventually had a successful pregnancy, almost all men (99 per cent) were free of mycoplasma after the course of antibiotic therapy; only two thirds of the men in the group of couples that did not have a successful pregnancy had specimens that were negative for mycoplasma after therapy.

The status of mycoplasma infection after therapy was not associated with age, occupation, order of marriage, presence of children in current marriage, length of marriage, length of time spent trying to conceive, a

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count or quality (Table 1).

Ninety-one women (57 per cent) had undergone at least one fertility procedure, and 55 (60 per cent) of them had undergone two or more. Thus, there was a fairly strong tendency toward a history of multiple fertility procedures.

Statistical analysis of post-therapy pregnancy status should take into account the differing lengths of follow-up among the women. For this reason, the methods commonly used for survival analysis were employed. Figure 1 shows the distribution of time until pregnancy according to the results of culture after therapy.

Table 1. Distribution of Variables Studied and Post-Therapy Mycoplasma Status in 161 Couples Given Antibiotic Therapy.

VARIABLE	NO. OF COUPLES (%)	PER CENT WITH NEGATIVE MYCOPLASMA CULTURE
Mycoplasma status		
Positive	32 (20)	—
Negative	129 (80)	—
Age (yr)		
≤35	140 (87)	81
>35	21 (13)	76
Occupation		
Blue-collar	33 (20)	76
White-collar	128 (80)	80
Order of marriage		
First	131 (81)	79
Second or more	30 (19)	80
Prior children in this marriage		
Yes	23 (14)	78
No	139 (86)	79
Length of marriage (yr)		
≤1	6 (4)	100
2–4	75 (46)	75
≥5	80 (50)	80
Length of time trying to conceive (yr)		
≤1	43 (27)	83
2–4	103 (64)	78
≥5	15 (9)	77
Hysterosalpingogram		
Yes	76 (47)	80
No	85 (53)	80
Tuboplasty		
Yes	33 (20)	80
No	128 (80)	82
Myomectomy		
Yes	8 (5)	88
No	153 (95)	80
Dilation and curettage		
Yes	44 (27)	80
No	117 (73)	80
Fertility drugs		
Yes	48 (30)	80
No	113 (70)	81
Sperm count (in millions)		
≤10	4 (2)	100
11–60	43 (27)	72
>60	114 (71)	83
Sperm quality		
Poor	27 (17)	89
Good	134 (83)	78
Post-therapy outcome		
Successful pregnancy	70 (44)	99 *
No successful pregnancy	91 (56)	66

*P<0.001 by Fisher's exact test.

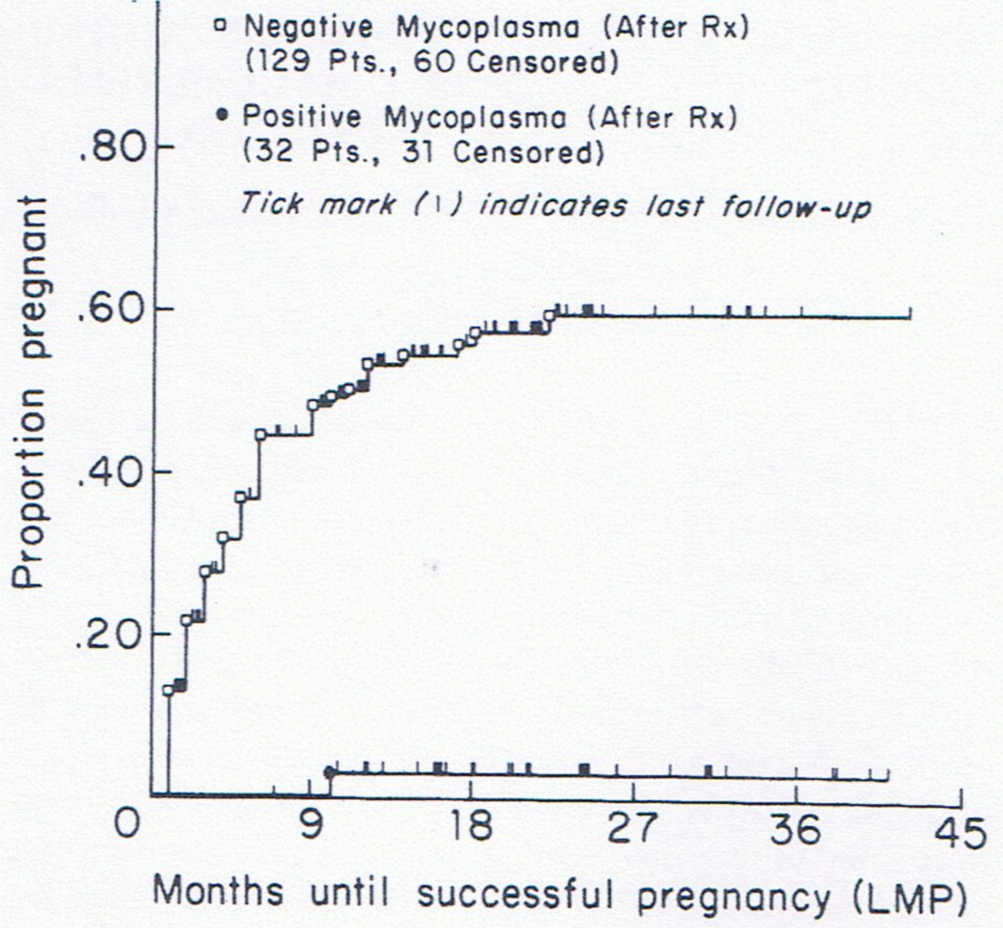

Figure 1. Time until Pregnancy in 161 Couples Given Antibiotic Therapy.

LMP denotes the last menstrual period. In couples with a negative culture for mycoplasma there was a significantly higher rate of pregnancy than in those with a positive culture (P<0.001). The three-year pregnancy rates, as estimated from the curves, were approximately 60 and 5 per cent for couples with negative and positive cultures, respectively.

Statistically significant differences among distributions of time until pregnancy were also related to hysterosalpingogram (P<0.001), tuboplasty (P<0.001), dilation and curettage (P<0.002), and fertility drugs (P<0.001). In each group of patients who had undergone a particular fertility procedure, the pregnancy rate was lower. Table 2 shows the estimated three-year pregnancy rates according to variable.

The Cox regression method was applied to the data to determine which combination of variables best predicted time until successful pregnancy, because the Cox model assumes proportional hazards and adequately fits the data. The procedure determined which variables were significant in the following order of importance: eradication of mycoplasma (P<0.001), no history of hysterosalpingogram (P<0.007), and no history of tuboplasty (P<0.03). Although other variables were associated with pregnancy outcome on a univariate basis (e.g., dilation and curettage and fertility drugs), the associations were not significant when considered independently of mycoplasma, hysterosalpingogram, and tuboplasty.

These results are interpreted to mean that the variable most significantly associated with the time until a successful pregnancy occurred was mycoplasma status after therapy. After mycoplasma status, hysterosalpingogram was the next most important variable, followed by tuboplasty.

DISCUSSION

A thorough understanding of the effect of T mycoplasma on fertility may be provided only by a double-

Table 2. Factors Associated with Pregnancy Outcome in 161 Couples Given Antibiotic Therapy.

VARIABLE	3-YEAR PREGNANCY RATE (%)	P VALUE *
Mycoplasma		
Negative	60	<0.001
Positive	5	
Hysterosalpingogram		
Yes	35	<0.001
No	57	
Tuboplasty		
Yes	14	<0.001
No	55	
Dilation and curettage		
Yes	32	<0.002
No	53	
Fertility drugs		
Yes	25	<0.001
No	57	

*Significance determined according to the log-rank test, which was used to compare distributions of time until pregnancy.

blind controlled prospective study in which well-defined cases of unexplained infertility are randomly assigned to placebo or antibiotic treatment. Nevertheless, our study establishes certain important associations between the status of T-mycoplasma infection and the predictability of pregnancy. Among the 129 couples in which the men were treated successfully for mycoplasma, with negative cultures after treatment, the three-year rate of successful pregnancy was 60 per cent, in contrast to the 5 per cent pregnancy rate among the 32 couples in which the men were treated unsuccessfully for mycoplasma, with positive cultures after treatment. We are not certain whether this difference is due solely to the successful eradication of mycoplasma or to the eradication of other aerobic and anaerobic bacteria and chlamydia for which doxycycline is an effective antibiotic. None of the examined variables was directly correlated with success in eradicating or failure to eradicate mycoplasma; therefore, we believe that at the end of the therapy positive cultures for T mycoplasma were colonized with doxycycline-resistant strains. The emergence of such strains is a growing concern,[10] and whether or not they represent a more pathogenic serotype is only speculative. We found a higher pregnancy rate among women under 35 years of age and in couples in which the man's sperm count and quality were higher. However, these differences were not significant. Patients who underwent tuboplasty, myomectomy, or dilation and curettage and those who received fertility drugs, clomiphene (Clomid), human menopausal gonadotropin (Pergonal), or human chorionic gonadotropin (Pregnyl) were less likely to become pregnant. According to the referring doctors' records, the patients who underwent such procedures as tuboplasty and myomectomy had prior pelvic damage established on the basis of a hysterosalpingogram. The negative correlation with fertility drugs suggests that there is a group of patients in whom the cause of infertility remains unknown, and that these patients are more likely than others to be treated with a variety of infertility procedures and medications empirically. The median time to achieve pregnancy after obtaining a negative mycoplasma culture was 10.6 months, and the bulk of the patients became pregnant without any additional fertility tests or drugs. According to the Cox regression model, the most important variable associated with time until pregnancy was mycoplasma status. In addition, a history of a hysterosalpingogram or tuboplasty had a significant negative association with time until pregnancy.

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