

## **PERSISTENCE OF CHLAMYDIAL DNA AND CHLAMYDIAL ANTIGENS AFTER THERAPY**

**National Chlamydia Laboratory Committee  
National Infertility Prevention Project (IPP)**

It is known that viable chlamydial organisms are not usually recoverable by tissue culture, after a compliant course of therapy with doxycycline (1-3). However, many studies have demonstrated that non-culture chlamydial tests can remain positive after chlamydial therapy, which is adequate in eradicating viable organisms as tested by tissue culture. Nachamkin, et al. demonstrated that 6 of 36 (6.7%) direct fluorescent antibody (DFA) tests remained positive at 7-10 days after therapy with doxycycline, while none of these patients were still culture positive (2). Additionally, they observed that 3 of 39 (7.7%) patients still had positive cultures, two of which also had a positive DFA.

Similarly, enzyme immunoassay (EIA) can remain positive immediately following therapy. From 106 infected women, Radcliffe et al. reported that 3 of 91 (3.3%), who returned within one week following antibiotic treatment with either doxycycline or oxytetracycline, were EIA positive (4). However, none of 90, who returned for a test-of-cure EIA test at 7-27 days remained positive. These findings are consistent with those of Lefebvre, et. al., who showed that, of 48 infected women tested 4 or more weeks after therapy, culture, EIA, and DFA were negative for all but one patient, who was positive by all three assays, and probably represented a reinfection (5). Cerin, et. al. monitored the disappearance of organisms detected by culture, EIA, and DFA of the cervix, as well as by culture and EIA of the urethra during doxycycline therapy of 48 women with chlamydial cervicitis (6). They demonstrated that two thirds of the patients were still positive by one or more tests on day 2, while 81% were negative by all tests on day 4, with all screening tests having turned negative by day 6 of treatment. At day 4, the only positive culture was for 1 urethral specimen, but 5 were positive by EIA of the cervix and 1 by cervical DFA. Conversely, Soren and Wills reported that chlamydial antigens persisted for 10% of specimens as long as 1-3 weeks following the eradication of viable organisms (7). In a study comparing chlamydial cell culture to EIA (Chlamydiazyme, Abbott Laboratories, Abbott Park IL) during erythromycin treatment of chlamydial genital infections, Havlichek, et. al. reported that by day 3, culture was positive for 17.2% (5/29), while EIA was positive for 37% (11/30) (3). At day 7, none were culture positive, while one was EIA positive and all tests were negative by day 14. The supposition by these authors was that the EIA and DFA tests were detecting antigens from dead organisms and thus, test-of-cure assays which employ non-culture tests must be interpreted cautiously.

Since PCR is much more sensitive than EIA or DFA, one would expect that DNA amplification tests would also remain positive longer after therapy than culture tests, which measure viable organisms. A study by Vogels, et. al. supports this supposition for cervical specimens (8). They compared cell culture and PCR for

follow-up of 35 women and 35 men positive for *C. trachomatis* by culture and/or PCR, and found that, while all post-treatment specimens taken at 2 weeks following treatment with doxycycline were culture negative, the PCR test remained positive for 3/70 (4.3%) patients (one female and two male samples) (8). Conversely, Claas, et. al. demonstrated that there was no residual DNA by PCR for 32 patients one week after doxycycline therapy (9).

Since compliance in taking antibiotics is always an issue in eradicating organisms, it is of interest that when Ossewaarde, et. al. compared the efficacy of azithromycin versus doxycycline for the treatment of chlamydial cervicitis they found that chlamydial DNA could be detected in patients from both treatment groups (10). Four patients (approximately 15%) had detectable DNA at one week after the start of therapy. One was in the doxycycline group and three were in the azithromycin group. After 4 weeks 7% (2/29) had detectable DNA by PCR, one from the doxycycline group and one from the azithromycin group.

Workowski, et. al. has demonstrated that none of twenty chlamydia-infected women were culture positive immediately after therapy, but ten had cervical specimens positive by PCR and two had PCR positive urethral specimens (1). In addition, three women had detectable DNA by PCR one week after therapy. After two weeks all specimens became negative and remained negative by both culture and PCR for 5 months of follow-up, except for one patient, who became reinfected.

In a study by Gaydos, et. al. urine PCR and LCR results for 33 chlamydia infected high school females were followed every other day after therapy with single dose azithromycin (26 students) or doxycycline (7 students) (11). Of the 33 infected students who presented follow-up urine specimens, urine PCR was positive for 33 (100%) at baseline, while urine LCR was positive for 29 (87.9%). Several specimens negative at baseline by urine LCR subsequently became positive on follow-up specimens. At 1-3 days after therapy, PCR of urine specimens was positive for 40% and LCR was positive for 73.3%. By days 4-6 post-therapy, PCR was positive for 21.1% and LCR was positive for 36.8%. From 8 students providing urine specimens at days 7-9, PCR was positive for 25% and LCR was positive for 12.5%. No specimens were PCR positive from 10 students providing specimens at days 10-12, but one (10%) were LCR positive. However, at days 13-15 after therapy, one of 8 (14.3%) urines was positive by PCR, while none were positive by LCR. None of 5 specimens collected 16-21 days after therapy and none of 7 urines collected >21 days after therapy were positive by PCR or LCR.

## Recommendation

Because of the persistence of chlamydial DNA and chlamydial antigens in cervical and urine specimens after successful therapy to eradicate viable chlamydial organisms, these assays should not be used for test-of-cure or testing of follow-up specimens within the first 3 weeks following antibiotic therapy.

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