Title: Repeat Chlamydia Diagnoses Increase the Hazard of Pelvic Inflammatory Disease among U.S. Army females: A Retrospective Cohort Analysis

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25 **Short Summary**

26 The hazard of PID increased significantly with the number of repeat chlamydia diagnoses among

27 U.S. Army females.

28

Abstract

Background: In the U.S. military, chlamydia is the mostly commonly diagnosed bacterial sexually transmitted infection and the rates of pelvic inflammatory disease (PID) have remained high since the early 2000s.

Methods: The relationship between the number of chlamydia diagnoses and risk of PID was investigated in a retrospective cohort analysis, among chlamydia cases in the U.S. Army during 2006-2012. Cox regression model was used to estimate hazard ratios (HR) for associations between the number of repeat chlamydia diagnoses and PID.

Results: The study population comprised 33,176 females with chlamydia diagnosis. Of these, 25,098 (75%) were diagnosed only once ("non-repeaters"). By comparison, 6,282 (19%), 1,435 (4%), and 361 (1%) women had one, two, and three repeat chlamydia diagnoses, respectively. Among these four groups, 1,111, 325, 72, and 25 PID diagnoses were noted. According to the Cox regression analysis, for every additional diagnosis of chlamydia, the risk of PID increased by 28% (95% CI = 19%, 38%) compared to females with a single diagnosis or "non-repeaters". Moreover, the corresponding adjusted HR of 1.28, 1.35, and 1.97 represented a significantly greater risk for PID among the three "repeater" groups compared to "non-repeaters".

Conclusions: We found an increased risk of PID among U.S. Army females with repeat chlamydia diagnoses and the characterization of a dose-response relationship. These findings reinforce the notion that early diagnosis and treatment of chlamydia is necessary to avoid subsequent PID and associated morbidity.

Keywords: Pelvic inflammatory disease; chlamydia; military; STI; surveillance

Pelvic inflammatory disease (PID) is an infection of the upper female reproductive tract.[1] This serious, but treatable infection, can lead to complications such as ectopic pregnancy, tubal factor infertility, and chronic pelvic pain.[2] Although the rates of PID have decreased over the past decade in the U.S., it still represents a significant health problem among women of reproductive age.[3] Among the U.S. Army females, PID is also commonly diagnosed, with an estimated rate of ~11 PID cases per 1,000 person-years during 2002−2011.[4]

Chlamydia trachomatis infection is an important risk factor for the development of PID, but its risk varies considerably.[5] For example, Hillis et al.[6] reported that the odds ratios of PID were 4.0 and 6.4 among 11,000 Wisconsin women with two versus three or more chlamydia infections, respectively. In contrast, Davies et al.[7] recently reported that among Canadian women the risk of PID after the first and the second repeat chlamydia infections was 1.2 and 1.3 compared to those with one infection, respectively. A plausible explanation for these differences may be due to the clinical diagnosis of PID is imprecise and challenge, with no a gold standard diagnostic test.[1,8]

Since 2000, the U.S. Army has reported that chlamydia is the most commonly diagnosed bacterial sexually transmitted infection [STI].[9] However, the risk of PID after having repeated chlamydia infections has not been studied to date. Therefore, the aim of this study was to determine the relationship between the number of chlamydia diagnoses and hazard of PID among female military personnel. To reach that aim, we conducted a retrospective cohort analysis, using data from the Defense Medical Surveillance System (DMSS).[10]

METHODS

Study Design and Population

Data were extracted from the DMSS, a large relational database that serves as the medical surveillance system for the U.S. Armed Forces since the 1990s.[10] The DMSS collects information on demographics, medical encounters, immunizations, laboratory results, and deployments among personnel from the first day of entry until their departure from service. The study population included all U.S. Army females aged 17−40 years, who were on active duty for any timeframe between 1 January 2006 and 31 December 2012, and who had at least one chlamydia diagnosis while in service. The "chlamydia case" definition was based on the International Classification of Diseases, 9th version (ICD-9) diagnostic codes 099.41 or 099.5x in either the first or second position of an inpatient or outpatient medical record. Women with a diagnosis of chlamydia, PID (614.9), infertility (628.4), pregnancy (V22.2), or gonorrhea (098.0x, 098.1x, or 098.8x) prior to 1 January 2006 were excluded. Pregnant females were excluded to ensure homogeneity in terms of sexual risk in our population as risky behavior would probably be minimized during pregnancy by female service members in accordance with medical advice.

Consequently, medical encounter records from all chlamydia cases were reviewed to determine the number of repeat chlamydia diagnoses (e.g. "repeaters") since the first date of diagnosis until they reached one of the following censoring events: (1) a diagnosis of PID (list of ICD-9 codes is shown as a footnote in Table 2); (2) end of the study period; or (3) termination of service. A repeat chlamydia diagnosis was defined as a diagnosis that occurred $\geq$ 30 days from a prior positive chlamydia test. Although this timeframe varies from study to study, a period of 30 days has been most often used in epidemiologic assessments to avoid duplication of diagnosis for a single episode.[11, 12] Besides, it is based on the evidence that nucleic acid amplification testing

can detect residual *C. trachomatis* nucleic acid for up to three weeks post-treatment[13] and follows the U.S. Centers for Disease Control and Prevention (CDC) guidelines to report a positive chlamydia laboratory as a new infection.[14]

In addition to reviewing medical encounter data, for each chlamydia case detected during the study period, we extracted data from the DMSS, with respect to socio-demographic and military specific variables. These variables included: (1) age (in years), (2) race/ethnicity (white, African-American, Hispanic, and other), (3) education (high school, college or higher), (4) marital status (single, married, and other), and (5) military rank (officer, enlisted).

Statistical Analysis

The demographic characteristics of the study population were summarized using descriptive statistics (e.g., means for continuous data and frequencies for categorical data). We used the Kaplan-Meier estimator to determine the cumulative incidence rate of PID by the end of follow-up after the first, second, and third time "repeaters." To compare Kaplan-Meier cumulative incidence curves of PID diagnosis, the log-rank test was used. We applied a Cox proportional hazards regression model to study differences on the distribution of time to PID diagnosis by the number of repeat chlamydia diagnoses. The proportional-hazards assumption was evaluated using the Schoenfeld's global test.[15] Due to the small number of women with more than three repeat chlamydia diagnoses during the study period, these were excluded from analysis. Data were analyzed using Stata version 12 (Stata Corporation, College Station, TX).

RESULTS

The study population comprised 33,176 female military personnel with a chlamydia diagnosis during 2006-2012. Of these, the mean (standard deviation) age at initial chlamydia diagnosis was 21.9 (standard deviation [SD] = 3.7) years and the median follow-up time was 2.9 years (interquartile range [IQR]: 1.5−4.7 years). As shown in Table 1, 25,098 (75%) women were diagnosed with *C. trachomatis* only once ("non-repeaters"). By comparison, 6,282 (19%), 1,435 (4%), and 361 (1%) women had one, two, and three repeat chlamydia diagnoses, respectively. Among the repeaters, highest burden was in females aged 20-24 years (52-53%), African-Americans (37-44%), those of single marital status (73-78%), those with high school education (93-97%), and those within the enlisted rank (91-95%).

Among the 33,176 females, a total of 1,533 (5%) PID diagnoses were subsequently diagnosed with a median time to diagnosis of 9 months. Results stratified by repeat chlamydia diagnosis (Table 2) indicate that among the 25,098 females without a repeat chlamydia diagnosis, there were 1,111 (4%) PID diagnoses. In this "non-repeater" group, the median time to PID diagnosis was 13 months. For "repeater" females with "one" repeat chlamydia diagnosis, 325 (5%) out 6,282 had a diagnosis of PID, with a median time of 10 months. For females with "two" repeat chlamydia diagnoses, 72 (5%) out 1,435 had a diagnosis of PID, with a median time to diagnosis of eight months. In addition, for 361 females with "three" repeat chlamydia diagnoses, there were 25 (7%) PID diagnoses, with a median time to diagnosis of only one month.

According to the Kaplan-Meier analysis, the cumulative incidence of PID increased in a dose-response relationship among chlamydia "repeaters," from 6% for females with a single chlamydia diagnosis ("non-repeaters") to 9%, 10%, and 11% for females with one, two, and three repeat chlamydia diagnoses, respectively (Table 2). The relationship between the four

study groups and cumulative incidence proportion of PID (1-survival) is depicted in Figure 1. Clearly, we can observe that as the number of chlamydia diagnosis increased, the incidence of PID increased. In addition, the results showed that cumulative risk curves were statistically significantly different by the log-rank test ($P < 0.001$).

According to the Cox regression analysis, for every additional diagnosis of chlamydia, the hazard of PID increased by 28% (95% CI = 19%, 38%, $P < 0.001$) compared to females with a single diagnosis or "non-repeaters". After controlling for age in years, race/ethnicity, marital status, and military rank, the hazard for PID decreased to 22% (95 % CI = 13%, 32%, $P < 0.001$) per additional chlamydia diagnosis, but remained statistically significant. When the number of repeat chlamydia diagnoses was treated as a categorical predictor, as shown in Table 2, analysis revealed that women with one (hazard ratio [HR] = 1.34), two (HR = 1.48), and three (HR = 2.21) repeat chlamydia diagnoses were at a significantly greater hazard of a diagnosis of PID as compared with females with a single diagnosis. Moreover, after controlling for the above potential confounders, the adjusted HRs showed significantly greater risk for PID among the three "repeater" groups compared to "non-repeaters." Based on the Schoenfeld's global test, the proportional-hazards assumption was met in both univariate ($P = 0.338$) and multivariate ($P = 0.176$) analysis.

DISCUSSION

In this retrospective cohort analysis of 33,000 U.S. Army females, we found that the hazard of PID increased in a dose-response relationship with each additional repeat chlamydia diagnosis. This finding highlights the importance of repeat chlamydia as a predisposing factor

for the development of subsequent PID and associated morbidity. Furthermore, our findings are consistent with previous studies conducted among civilian populations.[7,16-18]

The characteristics of the study population were consistent with those reported in other military-based studies in the literature.[9,19] That is, among U.S. Army females, most chlamydia diagnoses occur among those aged 17-24 years, African-Americans, single, and in junior enlisted military ranks. In our study, the percentage of women with repeat chlamydia diagnosis (24%) was slightly higher to that estimated by Owings et al.,[12] who reported that between 2010 and 2014, 20% of 23,482 female service members in the U.S. military were chlamydia "repeaters". Nonetheless, this difference may be due to differences in time periods. According to military surveillance reports, there was a peak in the annual incidence rate for chlamydia in the 2008.[20]

Our results also showed that the median time in months to PID diagnosis decreased by the number of chlamydia diagnoses. Although this finding may have important public health implications in terms of screening, it is worth mentioning that using administrative health data like the DMSS, it is difficult to estimate exactly the time from chlamydia infection to PID diagnosis, because both, the exposure and outcome, are highly asymptomatic. With this caveat in mind, our data suggests that it may be wise to reinforce primary prevention activities as counseling and education to prevent a subsequent PID diagnosis among chlamydia "repeaters" among U.S. Army females.

To the best of our knowledge, this is the first study to report the cumulative effect of repeat chlamydia diagnosis on the risk of PID in the U.S. military, a young, highly mobile, and sexually active population.[21] According to DMSS data, the risk of PID for every one additional diagnosis of chlamydia increased by 28%, although this percentage was attenuated, but remained

high (17%) when adjusted for demographic and military characteristics. Due to the fact that this was an observational study, we are not able to clearly explain the reasons for the increased risk or demonstrate a causal relationship between repeat chlamydia and PID diagnosis. However, we provide additional evidence on the impact of repeat chlamydia diagnoses on the risk of PID to the STI literature.

In addition to the observed cumulative effect of repeat chlamydia on PID, we found that the hazard of PID increases with the number of diagnoses of chlamydia. Although our hazard estimates cannot be directly compared with previous civilian studies because they vary in terms of methodology, study population, and case identification,[6-8,16-18,22] the consensus is the same. In other words, there is an increasing risk of PID after repeat chlamydia diagnoses. Despite these differences, the adjusted HRs of PID were similar to those reported by the largest published cohort of chlamydia and PID conducted to date.[7] In that study, conducted in Canada among 73,883 women aged 12-24 years during 1992-1996, the risk of PID after one and two repeat chlamydia diagnoses was 17% (95% CI = 6%, 30%) and 35% (95% CI = 4%, 75%), respectively.

Limitations

This study has some limitations. First, the DMSS does not collect data on vaginal sanitary practices (e.g., douching), risky sexual behaviors (e.g., multiple risk partners), or use of oral or other forms of contraception. Therefore, unmeasured confounders can affect the accuracy of our estimates. Second, PID and chlamydia are often asymptomatic, therefore, the reported rates of PID might be underestimated and the number of women with repeat chlamydia diagnoses might be higher than analyzed. Besides, a long-term asymptomatic chlamydia

infection increases the likelihood of developing PID compared with those that are diagnosed and treated quickly. Third, the most widely used ICD-9 code (614.9) was applied to identify PID; however, some PID diagnoses might have been missed if a clinical diagnosis of PID was represented by other ICD-9 codes. This may have resulted in an underestimation of the total number of PID cases in our study population. Finally, the length of follow-up in our study (a median of three years) may not have captured all PID cases (e.g., PID could have occurred after the study period ended). Therefore, this could have resulted in an underestimation of the true incidence of PID. However, its effect is difficult to judge because each year ~10,000 new women enter and leave the U.S. Army.[23]

Notwithstanding our limitations, this study included a large number of women, which increases the statistical precision of the parameter estimates. Furthermore, all eligible female service members with a chlamydia diagnosis during the study period were selected, thus, the risk of selection bias was minimized. As well, this is the first investigation reporting the time intervals between repeat chlamydia diagnoses and a subsequent diagnosis of PID, as well as the cumulative effect of repeat chlamydia on PID. With the data presented, we feel that our findings contribute an additional measurable evidence on the role that chlamydia plays as a risk factor for PID.

In conclusion, this study suggests that the hazard of a diagnosis of PID increased significantly with the number of repeat chlamydia diagnoses among U.S. Army females and this relationship was found to be characterized by a dose-response. Additional research is needed to better understand the reasons for this increased risk for subsequent PID diagnosis. These findings reinforce the notion that early diagnosis and treatment of chlamydia is necessary to avoid subsequent PID and associated morbidity. It may be wise for military medical policy

makers to consider developing new preventive strategies that are effective in reducing the risk of PID among chlamydia “repeaters,” a targeted sub-group for further STI research in the U.S. Army.

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Disclaimer:

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318 **TABLE 1.** Characteristics of the study population by the number of repeat chlamydia diagnoses

Feature	Total Study Population N	Women without repeat CT diagnosis n (%)	Women with one repeat CT diagnosis n (%)	Women with two repeat CT diagnoses n (%)	Women with three repeat CT diagnoses n (%)
No. of women	33,176 (100)	25,098 (75)	6,282 (19)	1,435 (4)	361(1)
Age group (years)					
17-19	9,103 (27)	6,144 (24)	2,150 (34)	573 (40)	146 (40)
20-24	18,103 (54)	13,858 (55)	3,311 (53)	742 (52)	192 (53)
25-40	6,060 (19)	5,096 (21)	821 (13)	120 (8)	23 (7)
Race/ethnicity					
White	13,604 (42)	10,733 (43)	2,308 (37)	454 (32)	109 (31)
African-American	10,871 (33)	7,836 (32)	2,281 (37)	597 (42)	157 (44)
Hispanic	4,540 (14)	3,434 (14)	885 (14)	181 (13)	40 (11)
Other[±]	3,616 (11)	2,683 (11)	703 (11)	183 (13)	47 (13)
Marital status					
Single	23,295 (70)	17,311 (69)	4,601 (73)	1,103 (77)	280 (78)
Married	7,942 (24)	6,218 (25)	1,374 (22)	280 (19)	70 (19)
Other[§]	1,925 (6)	1,557 (6)	305 (5)	52 (4)	11 (3)
Education					
High school	29,475 (90)	22,044 (89)	5,721 (93)	1,368 (97)	342 (97)
College or higher	3,108 (10)	2,598 (11)	450 (7)	48 (3)	12 (3)
Military rank					
Enlisted	28,695 (86)	21,318 (85)	5,684 (91)	1,351 (94)	342 (95)
Officers	4,481 (14)	3,780 (15)	598 (9)	84 (6)	19 (5)

319 Note: CT, chlamydia.
320 [±]Other race/ethnicity: American Indian/Alaskan Native, Asian/Pacific Islander, or other.
321 [§]Other marital status: divorced, separated, widowed, or other.
322

TABLE 2. Cumulative incidence rates and hazard ratios for pelvic inflammatory disease by the number of repeat chlamydia diagnoses.

Feature	Women with PID n/N	Median Follow-up time years	Cumulative incidence rate % (95% CI)	Cox Regression analysis	
				Crude HR (95% CI)	Adjusted HR (95% CI)¶
Women without repeat diagnosis	1,111/25,098	3.10	6.54 (6.11, 6.99)	Ref.	Ref.
Women with one repeat diagnosis	325/6,282	2.36	9.35 (7.71, 11.33)	1.34 (1.19, 1.52)[a]	1.28 (1.13, 1.45)[a]
Women with two repeat diagnoses	72/1,435	1.86	10.38 (7.68, 13.96)	1.48 (1.17, 1.88)[b]	1.35 (1.06, 1.72)[b]
Women with three repeat diagnoses	25/361	1.55	11.05 (6.84, 17.58)	2.21 (1.48, 3.28)[a]	1.97 (1.31, 2.96)[b]

Note: PID, pelvic inflammatory disease (ICD-9 codes 098.10, 098.16, 098.17, 098.19, 098.86, 614.0, 614.2, 614.3, 614.5, 614.8, 614.9, 615.0, 615.9); p-y, person-years; CI, confidence interval; HR, hazard ratio; Ref., reference for HR calculation.

¶Adjusted for age (in years), race/ethnicity, marital status, and military rank.

[a]$P < 0.001$

[b]$P < 0.05$

FIGURE 1. Cumulative incidence of pelvic inflammatory disease by the number of repeat chlamydia diagnoses using the Kaplan-Meier method.