

## RESEARCH LETTER

**TITLE:** Association of past *Chlamydia trachomatis* infection with miscarriage

**SUBTITLE:** *Chlamydia trachomatis* infection and miscarriage

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**Tweet:**

UK study shows that there is no significant association of past chlamydial exposure with spontaneous first trimester miscarriage - but observed past infection rates of >25% suggest that prevalence of Ct in young women remains underestimated *@horne\_research*

**Introduction:**

First trimester miscarriages are commonly associated with chromosomal abnormality of the embryo (~50% of cases)<sup>1</sup>. However, 15% of first trimester, and 66% of second trimester miscarriages, are attributed to reproductive tract infections<sup>2</sup>. It has been suggested that *Chlamydia trachomatis* (Ct) is a causative organism but its association with miscarriage is inconsistent<sup>2-4</sup>. This difference of opinion likely reflects the poor performance of major outer membrane protein (MOMP) peptide-based serology assays and the inability of nucleic acid amplification tests (that detect current infection) to detect prior exposure<sup>5</sup>. It is now possible to accurately measure lifetime exposure to Ct using an enzyme-linked immunosorbent assay (ELISA) that detects antibodies to the chlamydial plasmid-encoded protein Pgp3<sup>5</sup>. This ELISA is more sensitive (73.8%) and specific (97.6%) than commercial ELISAs, including the Medac MOMP-peptide ELISA, or previous serological antibody tests<sup>5</sup>. Pgp3 is unique to Ct, eliminating cross-reactivity with antibodies to *C. pneumoniae* infection (a common respiratory pathogen), a major weakness of previous serological tests. The aim of this study was to provide an estimation of the risk of prior Ct infection on spontaneous first trimester miscarriage.

**Methods:**

We performed a case-control study recruiting 251 women with an ultrasound confirming absence of a fetal heart in the first trimester of pregnancy ('miscarriage group') and 118 women with normal pregnancies that had progressed into the third trimester from the same catchment population ('controls'). Women with a past history of miscarriage were excluded from the controls. We anticipated a Ct seroprevalence of 15% in women with miscarriage and 7% in the controls, based on literature review<sup>3</sup> and pilot work, and our proposed sample size (200 cases, 100 controls) had >95% power, with a level of significance (alpha) of 0.05, to

estimate a doubling of the *Ct*-Population Attributable Risk for miscarriage. We collected serum samples and self-taken vulvo-vaginal swabs taken from 2-3 inches within the vagina for *Ct* nucleic acid amplification testing to detect current infection. The Scotland A Research Ethics Committee approved the study (12/SS/0098). Written informed consent was obtained from all participants. Participants were identified from the Pregnancy Support Unit and Delivery Suite at the Royal Infirmary of Edinburgh (a large UK NHS teaching hospital). The first study participant was recruited on 22 January 2013 and the last participant recruited on 26 September 2019. Analysis was by two-tailed Fisher's exact test.

### **Results:**

The groups were well balanced for all characteristics measured at baseline (see Table 1). 26% (CI:20.6-31.4) of the miscarriage group and 28% (CI:19.9-36.1) of the control group were Pgp3 antibody positive, suggesting prior infection with *Ct* ( $P=0.71$ ). There was no evidence of active *Ct* infection in either group. More women in the miscarriage group (13.5%; CI:11.3-15.7) than the control group (1.7%; CI:0.5-2.9) self-reported past *Ct* infection ( $P=0.0001$ ).

### **Discussion:**

Contrary to the study by Baud et al<sup>3</sup>, which was conducted on a similar size dataset using a MOMP-peptide ELISA, our study, using the more sensitive Pgp3 ELISA, demonstrates that there is no significant association of past *Ct* exposure with spontaneous first trimester miscarriage. The lack of genetic analysis of the miscarriage and inability to match for past obstetric history are limitations of the study. It is unclear why more women in the miscarriage group self-reported *Ct* infection as recall bias is unlikely to explain such a difference. One possibility is that this may reflect that they were more likely to have had symptomatic *Ct*

infection and therefore seek testing. However, the seroprevalence rates of over 25% observed in both cohorts suggests that the prevalence of *Ct* infection in young women, and potential clinical impact on other reproductive disorders, such as female infertility and ectopic pregnancy, remains underestimated.

**Table 1. Summary data**

Characteristic		Controls (n=118)	Miscarriage group (n=251)	Significance
Median age in years		34 (CI: 32-35)	33 (CI: 32-35)	<i>P</i> =0.72
Mean BMI in kg/m <sup>2</sup>		26.0 (CI: 25.0-27.0)	25.6 (CI: 24.9-26.3)	<i>P</i> =0.55
Self-reported past <i>Ct</i> infection		2 (1.7%)	34 (13.5%)	<i>P</i> =0.0001 ****
<i>Ct</i> seropositivity		33 (28.0%)	65 (25.9%)	<i>P</i> =0.71
Prior miscarriage		-	106 (41.4%)	-
Prior live births		85 (72.0%)	127 (50.6%)	<i>P</i> =0.0001 ****
History of smoking	Never	75 <sup>a</sup> (66.4%)	165 <sup>b</sup> (66.8%)	<i>P</i> =1.00
	Ex-smoker	27 <sup>a</sup> (23.9%)	60 <sup>b</sup> (24.3%)	<i>P</i> =1.00
	Smoker	11 <sup>a</sup> (9.7%)	22 <sup>b</sup> (8.9%)	<i>P</i> =0.84

<sup>a</sup>113 and <sup>b</sup>247 responses

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