Adjusting for differential testing: Estimating the incidence of *Chlamydia trachomatis* infections identified through Public Health Ontario laboratories in Peel region, Ontario, 2010–2018

Lindsay Obress MSc[1], Olaf Berke PhD[1], David N. Fisman MD MPH[2], Shilpa Raju MPH[3], Ashleigh R. Tuite PhD MPH[2], Monali Varia MHSc[3], Amy L. Greer PhD[1,2]

[1] Department of Population Medicine, University of Guelph, Guelph, Canada

[2] Dalla Lana School of Public Health, University of Toronto, Toronto, Canada

[3] Peel Public Health, Peel region, Canada

Corresponding Author: Amy L. Greer, PhD. Department of Population Medicine, University of Guelph. 50 Stone Rd E, Guelph, ON N1G 2W1. Email: agreer@uoguelph.ca

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Abstract

Background: Public health guidelines in Canada recommend annual chlamydia testing in sexually active individuals under 25 years of age and/or those with additional risk factors, such as pregnancy or history of sexually transmitted infections. However, the frequency of testing is variable across practitioners and clinic settings. Testing in females is emphasized as they carry the largest reported burden of chlamydia and the largest risk of chlamydia associated sequelae. While lower rates of chlamydia infections have been observed in males, it has been hypothesized that males are hidden reservoirs for *Chlamydia trachomatis* and differential testing may explain the difference in rates observed across the sexes.

Methods: This study was based on diagnostic test results recorded by Public Health Ontario laboratories for individuals living in Peel region, Ontario. The goal of the study was to compare and interpret results from age and sex standardized testing rates.

Results: Observed incidence and testing was highest in females aged 20–29, while males had the highest standardized test positivity across all age groups. After adjusting for testing, males in the 15–19 and 30–39 age groups had 60.2% and 9.7% increase in incidence compared to the observed incidence.

Interpretation: It was found that infections in males are likely being missed due to differential testing and this may be contributing to the persistent increase in reported cases in Canada. Public health programming that targets males, especially in high-risk settings and communities, and use of innovative partner notification methods, could be critical to curbing overall rates of chlamydia.

Introduction

Public health guidelines for sexually transmitted infection (STI) testing and screening make recommendations regarding the groups of individuals to whom to target testing and screening resources (1). In the case of chlamydia, the Public Health Agency of Canada recommends annual screening for sexually active people under the age of 25; and a recent report from the Canadian Task Force on Preventive Care recommends annual screening for sexually active people under the age of 30 (1,2). Despite these guidelines, many factors determine if, and how often, individuals get tested. In Canada, 60% of individuals surveyed reported they had never been screened for STIs, suggesting that even with current guidelines, many patients are not screened routinely for STIs (3). There is significant emphasis placed on testing females under the age of 25 due to the often asymptomatic nature of chlamydia infections and the possibility of long-term health complications including pelvic inflammatory disease (PID), infertility, and ectopic pregnancy (4–6). Screening males however, has been the subject of significant debate and various organizations in the United States, including the Centers for Disease Control and Prevention (CDC) and the U.S. Preventive Services Task Force (USPSTF), claim there is insufficient evidence to support regular screening of young men (7,8). Despite noting asymptomatic infections are common in both males and females, the CDC does not advise general screening of males for chlamydia and only states it should be considered in high-risk populations such as men who have sex with men (7). In Canada, while guidelines are not sex-specific, screening efforts have primarily been focused on women under the age of 25 and rates of chlamydia in this group continue to increase (9). This leads to questions regarding the factors that may be contributing to this observed increase. Are increased rates simply a function of increased testing and therefore, improved case finding? Or, is there a group of individuals (e.g. males or another age group) being missed by current screening

guidelines that are contributing to the transmission dynamics but not being identified and receiving treatment to cure their undiagnosed infection. For example, infections in males going undiagnosed can result in an ongoing chain of transmission, where in heterosexual relationships, female partners are at risk of infection and/or reinfection.

To better understand the influence of testing rates on case detection rates in populations where not all subgroups are tested at the same intensity, standardization can be used to adjust for testing rates among different subgroups of interest to provide an adjusted incidence estimate based on the assumption that all groups were tested at the same rate as the observed maximally tested group (10). This study will focus on chlamydia testing done by Public Health Ontario Laboratories for individuals residing in Peel, Ontario, a municipality in the Greater Toronto Area (GTA). The objectives of this study were to 1) describe the trends in incidence, tests, and test positivity of chlamydia across subgroups over the study period, 2010–2018, in Peel region, Ontario; 2) to determine subgroups that were at highest risk of infection and had the highest testing and test positivity rates; and 3) to estimate the test-adjusted incidence of chlamydia in subgroups assuming they were tested at the same rate as the maximally tested group.

Materials and Methods

Dataset and study population

The dataset included results from all chlamydia tests submitted to the Public Health Ontario (PHO) Laboratory in Ontario, Canada between 2010 and 2018. All individuals were 15 years of age (or older) at the time of testing and had a postal address within the region of Peel, Ontario. Incident case reports were obtained from the integrated Public Health Information System (iPHIS) and the number of tests completed was obtained from the Ontario Laboratories Information System

(OLIS). Testing data from OLIS were aggregated into 10-year age bands, except for ages 15–19 years. To calculate rates, population estimates were obtained from Statistics Canada 2006, 2011, and 2016 census profiles (11–13). Linear interpolation and extrapolation were used to estimate non-census year population sizes. The 2006 and 2011 census population were used to estimate the 2010 population, the 2011 and 2016 census populations were used to estimate the 2012–2015 populations, and the 2017–2018 populations. For the purpose of the study, the dataset was divided into age-sex subgroups as follows: 15–19 years, 20–29 years, 30–39 years, age 40 and over, for males, females, and the overall population. Due to low testing rates and case counts in individuals aged 40+, all age groups above 40 years were aggregated into a single group.

Standardized morbidity ratio, testing ratio, and test positivity

Standardized morbidity ratio, testing ratio, and test positivity (SMR, STR, and STP) were estimated for monthly and cumulative data to explore infections, testing, and test positivity. Where a ratio above 1 indicate infections, testing, or test positivity are higher than expected, and below 1 indicates they are lower than expected. First, average annual incidence of infection, testing, and positivity per test, was calculated for the population as a whole and for each age-sex subgroup. The ratios were then estimated by dividing subgroup specific estimates by population estimates. Confidence intervals were calculated using standard error estimates as follows (14):

$$\text{SE (ln(SMR, STR or STP))} = \sqrt{\left(\frac{1}{A}\right) - \left(\frac{1}{A+C}\right) + \left(\frac{1}{B}\right) - \left(\frac{1}{B+D}\right)}$$

Where A is the case or test count in a subgroup, B is the case or test count not in the subgroup, C is the subgroup population or test count subtract A, and D is the population or test count not in the subgroup subtract B.

The differences in ratios across subgroups and time were explored by constructing meta-regression models weighted by standard error estimates (as described above):

$$\ln(\text{SMR, STR, or STP}) = \alpha + \beta_i X_i + \beta_j X_{j+} \beta_k X_k$$

where α is the model intercept and each β represents a coefficient for the i^{th} age group, j^{th} sex, and k^{th} year (15).

Adjusting for testing

To account for the differential testing across age and sex subgroups, a test-adjusted incidence using standardization was applied. First, the "test-adjusted" SMR was estimated for each subgroup, age- and sex-specific meta-regression models, with standard errors, described above, were constructed. Within-subgroup standard errors were estimated by the summed square root of variance of both SMR and STR. The model follow the form of:

$$\ln(\text{SMR}) = \alpha + \beta_1 \ln(\text{STR})$$

When STR is equal to 1, where testing in a given subgroup is equivalent to the overall population test rate, ln(STR) is zero and therefore, the SMR is equal to the intercept, α, exponentiated (e^{α}). This can be interpreted as the test-frequency-adjusted SMR expected when all age-sex subgroups were tested at the same rate as the population overall. To investigate this further, the expected test-frequency-adjusted incidence of each subgroup (if tested at the same rate as the maximally tested subgroup) was calculated, as follows:

$$I_{iTmax} = SMR_i * I_0$$

Hence, the observed average annual incidence in the maximally tested i^{th} age-sex subgroup, I_{iTmax}, was divided by the test-adjusted SMR_i to find I_0, the test-adjusted expected incidence in the population overall. For all other subgroups, the test-adjusted incidence when tested at the rate of the maximally tested subgroup, was the test-adjusted SMR_i multiplied by I_0. This calculation was conducted for each age-sex subgroup, where females aged 20–29 were the maximally tested subgroup. To determine if there was a change in incidence after test-frequency-adjustment, test-adjusted incidence was compared to observed incidence. If the observed incidence was within the 95% confidence interval of the test-adjusted incidence, we assumed the test adjustment did not change the incidence.

Data Analysis

All analyses were conducted using StataIC 16 (16). Figures were created using the *ggplot2* package in R-4.0.2 and RStudio (17–19). A significance level of 5% was used for all tests and confidence intervals. Programming code is available upon request from corresponding author.

Ethics Approval

This study was approved by the University of Guelph Research Ethics Board (REB# 18-11-001).

Results

The dataset included 10,298 cases and 186,567 tests reported by Public Health Ontario Laboratories in Peel, Ontario between January 1st, 2010 and December 31st, 2018. Individuals with

unknown age (0 cases, 16 tests), and with unknown sex (21 cases, 3210 tests) were removed resulting in a final dataset of 10,277 cases and 183,341 tests.

Incidence, testing rate, and test positivity ranged across age and sex groups over time (Figure 1). Case incidence in the 15–19 age group was highest among females from 2010–2014 but decreased from 481 to 357 cases per 100,000 over the study period. In 2014, the female age group with the highest incidence was 20–29 years old and increased from 331 cases per 100,000 in 2014 to 435 cases per 100,000 in 2018. In males, the 20–29 age group had consistently higher incidence compared to all other male age groups, ranging from 291 cases per 100,000 to 408 cases per 100,000. The lowest case incidence was in the 40 and over age group across both sexes. In regards to testing, the 20–29 age group was the most tested per 100,000 population in both sexes, however, the rate of testing was much higher in females at approximately 7,000 tests per 100,000 population throughout the study period while in males this ranged from approximately 3,000 tests per 100,000 in 2010 to 4,500 tests per 100,000 population in 2018. Percent test positivity was highest among the 15–19 age group in both sexes throughout the study period, except in 2017 among males where the 20–29 year age group had slightly higher test positivity. Percent positivity was generally higher in males, when comparing the same age groups.

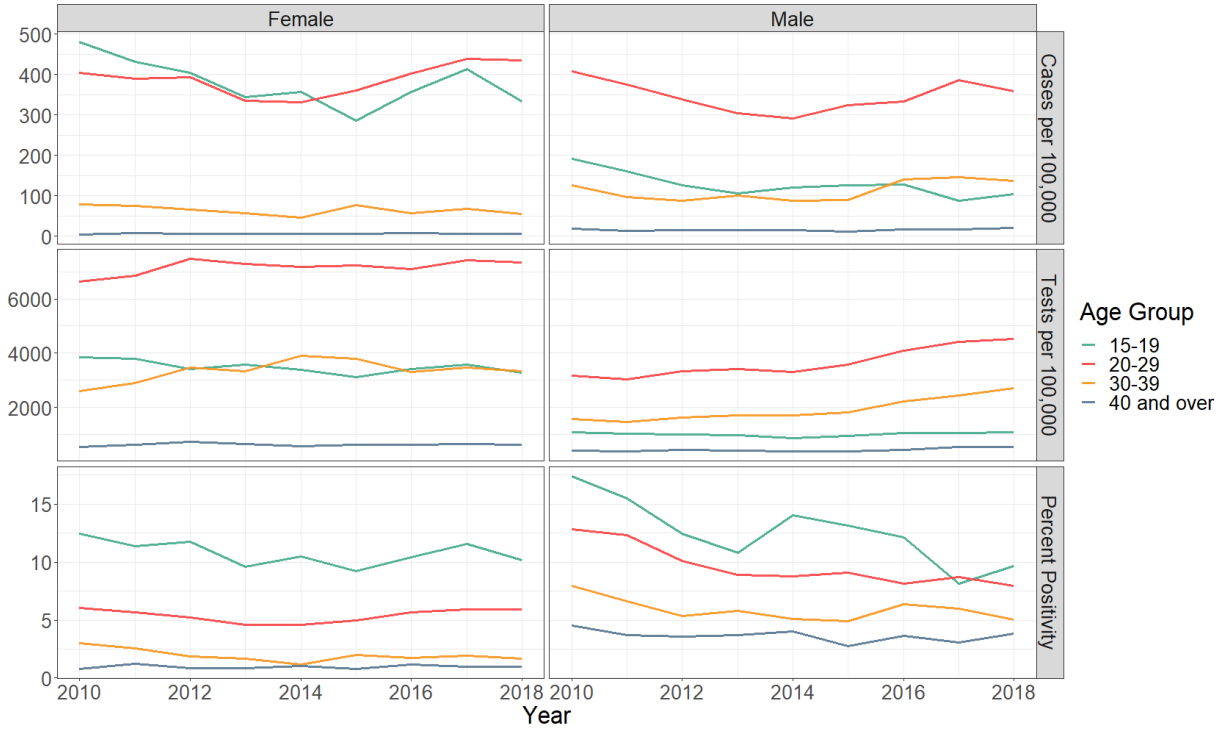

Figure 1: Line graphs of annual chlamydia cases and tests per 100,000 population (top 2 panels in each column) and test positivity (bottom panel in each column), grouped by sex and age, in Peel, Ontario identified through Public Health Ontario laboratories, 2010–2018.

The average annual incidence was examined across the age groups by sex (Figure 2). In the 15–19 and 20–29-year age groups, females had higher incidence than males, however this result is reversed in the 30–39 and 40 and over age groups where males had higher average annual incidence. Overall average annual incidence was highest among the 20–29 year age group, with the 15–19 age group also having high rates (Figure 2).

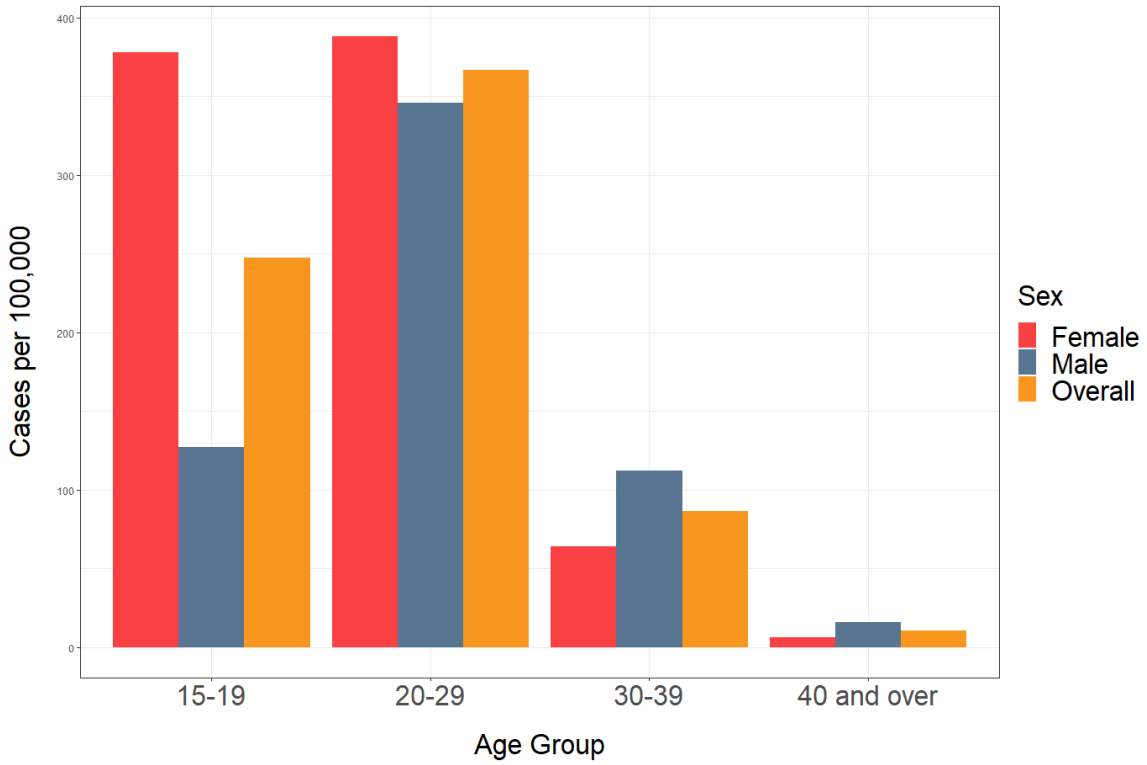

Figure 1: Bar graph of average annual incidence of chlamydia, grouped by age and sex, in Peel, Ontario identified through Public Health Ontario laboratories, 2010–2018.

The standardized morbidity ratio (SMR) was above 1 in females and males aged 15–19 and 20–29, and in 30–39-year-old males (Figure 3; Table 1). The SMR was below 1 for females 30 and over, and in males 40 and over (Figure 3; Table 1). The standardized testing ratio (STR) was above 1 for females under 40 and males 20–29 and 30–39 (Figure 3; Table 1). The STR was below 1 for females and males 40 and over, and in males 15–19 years old (Figure 3; Table 1). The standardized test positivity (STP) was above 1 in females and males 15–19 years, and in males 20–29 and 30–39 (Figure 3; Table 1).

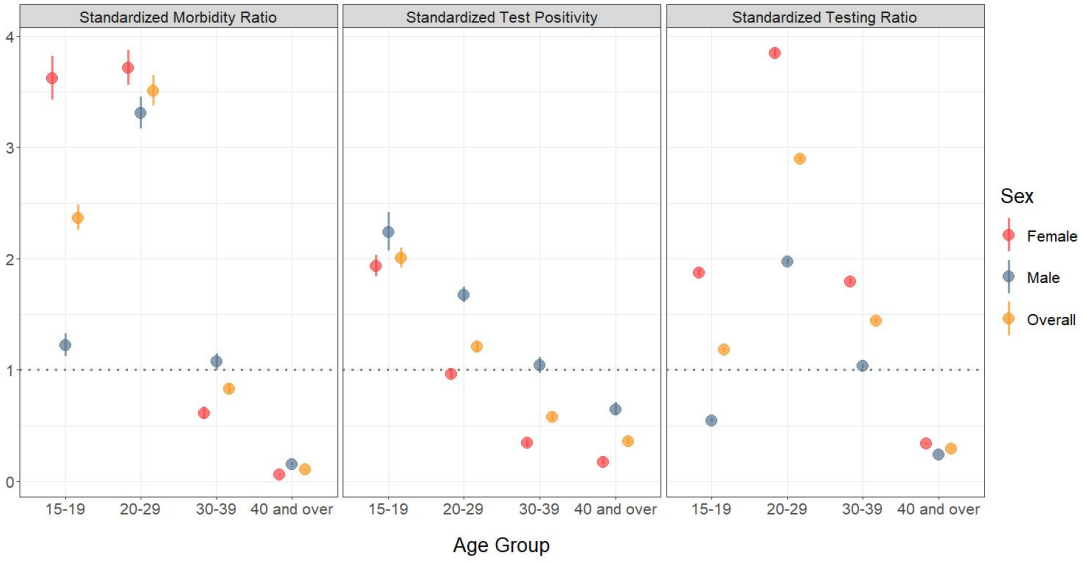

Figure 2: Standardized morbidity ratio (SMR), test positivity (STP), and testing ratio (STR) of chlamydia infections in Peel, Ontario identified through Public Health Ontario laboratories, 2010–2018, by sex and age subgroups. The circle indicates the point estimate of SMR, STP, or STR and line indicates 95% confidence interval.

Table 1: Standardized morbidity ratio (SMR), testing ratio (STR), and test positivity (STP) of chlamydia infections, by age and sex groups, in Peel, Ontario, identified through Public Health Ontario laboratories, 2010–2018.

Demographic		Standardized Ratios		
Sex	**Age Group**	**SMR (95% CI)**	**STR (95% CI)**	**STP (95% CI)**
Female	**15-19**	3.62 (3.43–3.82)	1.87 (1.84–1.90)	1.93 (1.83–2.04)
	20-29	3.71 (3.56–3.87)	3.85 (3.81–3.89)	0.97 (0.92–1.01)
	30-39	0.61 (0.56–0.67)	1.79 (1.77–1.82)	0.34 (0.32–0.37)
	40 and over	0.06 (0.05–0.07)	0.34 (0.33–0.34)	0.17 (0.15–0.20)
Male	**15-19**	1.22 (1.12–1.32)	0.54 (0.53–0.56)	2.24 (2.05–2.45)
	20-29	3.31 (3.17–3.45)	1.97 (1.95–2.00)	1.68 (1.60–1.75)
	30-39	1.07 (1.01–1.15)	1.03 (1.01–1.05)	1.04 (0.97–1.12)
	40 and over	0.15 (0.14–0.17)	0.24 (0.23–0.24)	0.65 (0.59–0.71)

Meta-regression models including the predictors age group, sex, and year were applied to estimate investigate the effects on SMR, STR, and STP values. Year was found to have little effect and was removed from the final models (Table 2). Test-adjusted SMR was determined for each age and sex subgroup (Figure 4). Test-adjusted SMR was above 1 in males aged 15–19, 20–29, and 30–39. In females and the overall population, test-adjusted SMR was above 1 in ages 15–19 and 20–29. Additionally, the 30–39 age group overall had an SMR above 1 but contained 1 in the confidence interval. All other subgroups, females 30–29, and all sexes 40 and over, had test-adjusted SMR below 1.

Table 2: Meta-regression models of sex and age on standardized morbidity ratios, standardized testing ratios, and standardized test positivity, in Peel, Ontario, identified through Public Health Ontario laboratories, 2010–2018. Females aged 20-29 were used as the referent group in meta-regression models.

Covariate	Standardized Morbidity Ratio		Standardized Testing Ratio		Standardized Test Positivity	
	SMR (95% CI)	P-value	STR (95% CI)	P-value	STP (95% CI)	P-value
Sex						
Male	0.872 (0.826–0.920)	<0.001	0.504 (0.490–0.518)	<0.001	1.837 (1.753–1.901)	<0.001
Female	1 (referent)		1 (referent)		1 (referent)	
Age						
15-19	0.754 (0.706–0.805)	<0.001	0.387 (0.372–0.403)	<0.001	1.842 (1.759–1.953)	<0.001
20-29	1 (referent)		1 (referent)		1 (referent)	
30-39	0.264 (0.245–0.283)	<0.001	0.490 (0.472–0.509)	<0.001	0.521 (0.498–0.562)	<0.001
40 and over	0.041 (0.037–0.045)	<0.001	0.101 (0.098–0.105)	<0.001	0.366 (0.343–0.406)	<0.001

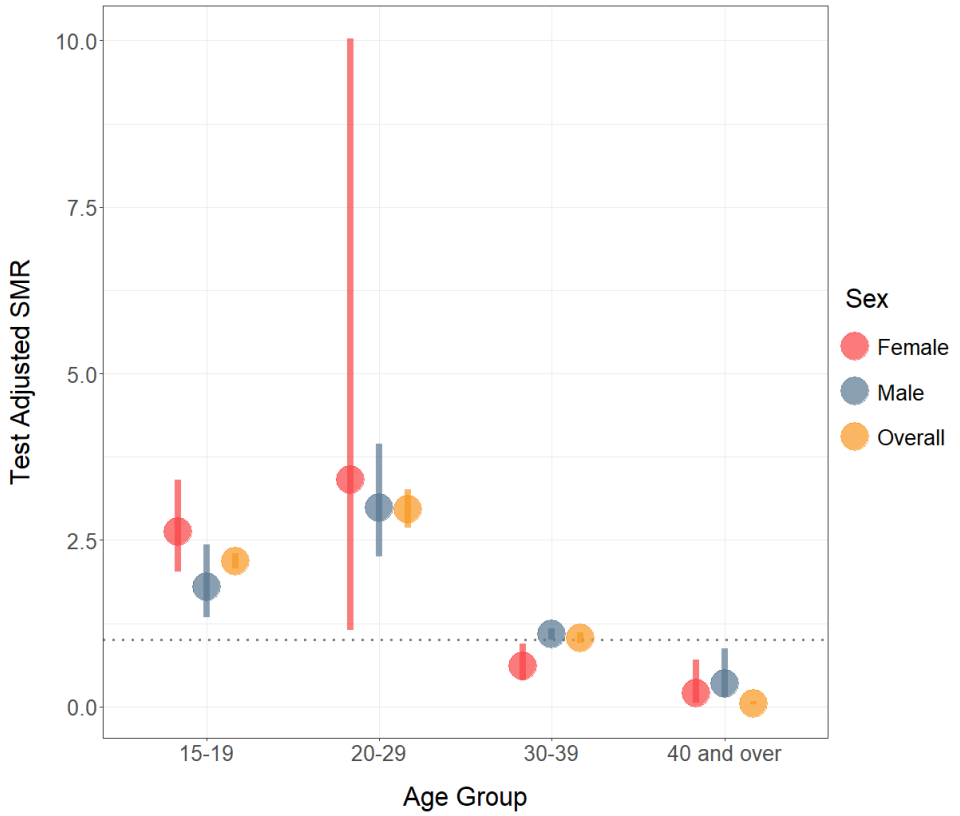

Figure 3: Test-adjusted standardized morbidity ratio (SMR) of chlamydia infections, by age and sex subgroups, in Peel, Ontario, identified through Public Health Ontario laboratories, 2010–2018. The centre of circle indicates the point estimate of test-adjusted SMR and the line indicates the 95% confidence interval.

The most frequently tested age-sex group was women aged 20–29 (STR = 3.85) and therefore the average annual incidence of this group was used to derive test-adjusted incidence for all other subgroups (Figure 5; Table 3). The estimated test-adjusted incidence in the population overall, I_0, was 114 cases per 100,000 population. This is an 8.5% increase compared to the observed average annual incidence of 105 cases per 100,000 population. Test-adjusted incidence was higher than observed incidence in males aged 15–19 and 30–39 and in the 30–39 age group overall. In males aged 15–19, the test-adjusted incidence was 60.2% higher than the observed

incidence (205 vs. 128 cases per 100,000). In males aged 30-39, the test-adjusted incidence was 9.7% higher than the observed incidence (124 vs. 113 cases per 100,000). The overall incidence in the 30–39 age group was 35.6% higher after adjusting for testing than the observed incidence (118 vs. 87 cases per 100,000). The overall incidence in the 40 and over age group showed a decrease from 11 cases per 100,000 to 6 cases per 100,000 after adjusting for testing. The test-adjusted incidence in 15-19-year-old females, 20–29-year-old males and 20–29-year-old age groups overall showed decreases, however, the observed incidence was within the 95% confidence interval of the test-adjusted incidence and deemed to be not different.

Table 3: Observed and test-adjusted incidence of chlamydia infections, by age and sex subgroups, in Peel, Ontario, identified through Public Health Ontario laboratories, 2010–2018.

Age Group	Female		Male		Overall	
	Obs.	Test-adj. (95% CI)	Obs.	Test-adj. (95% CI)	Obs.	Test-adj (95% CI)
15–19	379	299 (231–388)	128	205 (152–277)	248	249 (236–263)
20–29	388	388 (132–1143)	346	340 (257–449)	367	338 (306–372)
30–39	65	69 (45–108)	113	124 (115–134)	87	118 (109–127)
40 and over	7	23 (6–81)	16	40 (16–99)	11	5 (3–9)

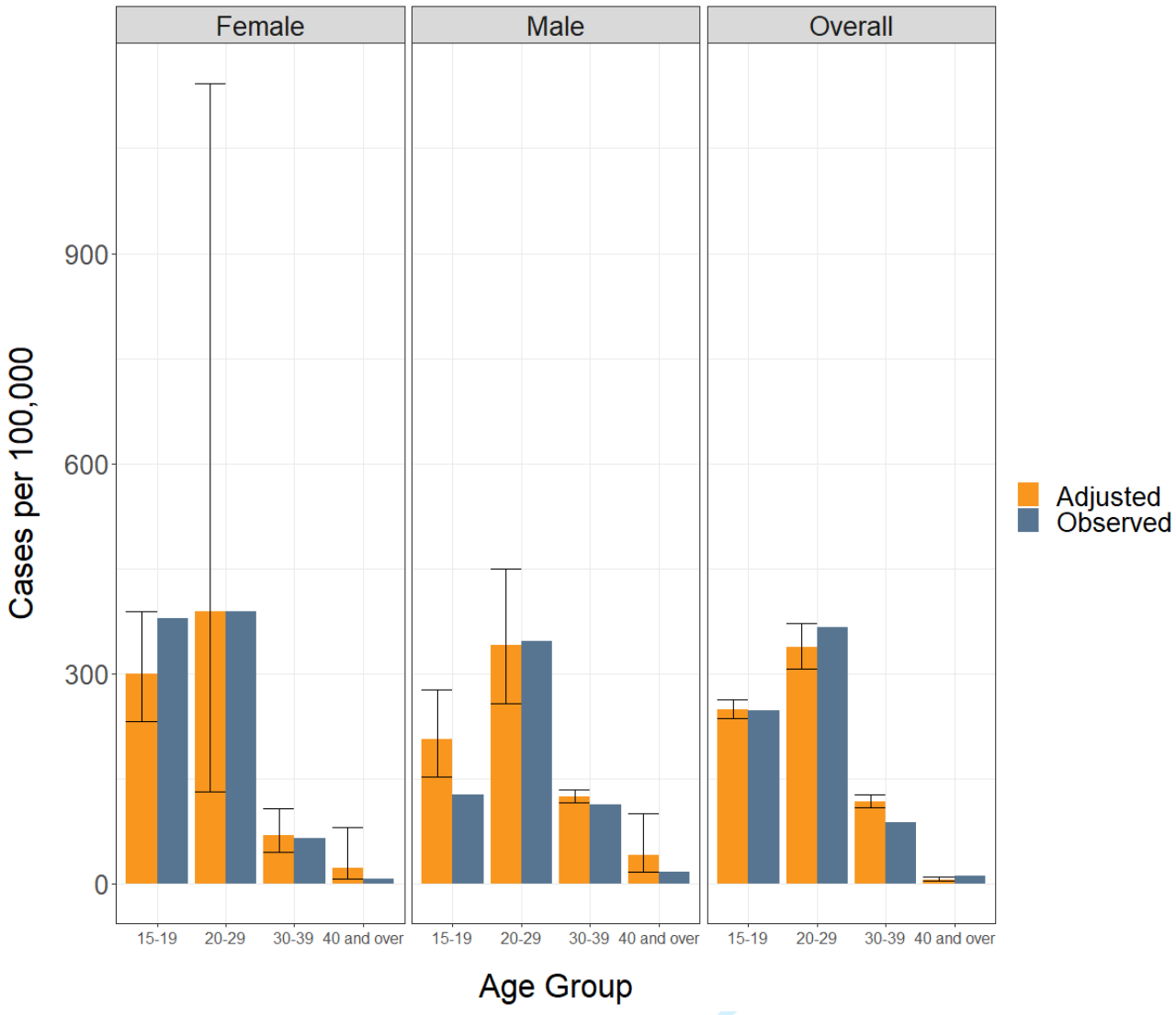

Figure 4: Bar graph of observed and test-adjusted incidence of chlamydia infections, by age and sex subgroups, in Peel, Ontario, identified through Public Health Ontario laboratories, 2010–2018. Error bars indicate 95% confidence interval of test-adjusted incidence estimates.

Interpretation

The findings of this study emphasize the need to further investigate the role of males in the dynamics of chlamydia infections in Ontario. Males were most likely to test positive for chlamydia but had the lowest testing rate, demonstrated by the highest STP but the lowest STR across all age groups (Figure 3). After adjusting for testing frequency, males in the 15–19 and 30–39 age groups

showed a 60.2% and 9.7% increase in average annual incidence of chlamydia when compared to the observed rates (Figure 5; Table 3). The 30–39 age group showed a 35.6% increase in average annual incidence after adjusting for testing compared to observed rates when both sexes were examined together (Figure 5; Table 3). These increases, after adjusting for testing frequency, suggest that these groups may be under-tested and that they may play a larger role in the transmission dynamics of chlamydia infections than previously considered.

In this study, year did not have an effect in meta-regression model despite changes to public health policy over time. This could be partially due to the study data source. This study used public health laboratory testing data only, which focuses primarily on tests performed at public health clinics. In Ontario, a large proportion of STI testing is completed at private laboratories and these data were not accessible for the study. Individuals tested through private laboratories may represent a population with different risk factors and may be screened differently due to variability in STI screening practices across primary care physicians and nurses (20–22). In specialized public health clinics, individuals are often seeking STI testing as the reason for their visit and may also have longer consults with care providers (23,24). This may allow for increased opportunistic screening, or a lower threshold for testing in public health clinics compared to primary care due to the dynamics across these settings.

The hypothesis that males would have higher observed incidence rates if tested more often is supported by literature that finds males less likely to seek health care and be screened for STIs during healthcare visits (25–28). Males are less likely to be tested than females for chlamydia during routine medical exams despite current testing guidelines indicating anyone under age 25 is at risk (3,22,29). Sex is also related to consultation length where females have longer consultations with their primary care physician than males, providing more time to enquire about sexual health

related risks and to discuss STI screening (27). Teenaged males, ages 13–18, are also less likely to attend sexual health clinics compared to teenaged females (24,28,30). This difference can in part be attributed to teenaged females seeking access to contraceptives, however, once an individual attends a sexual health clinic they are likely to return for future sexual health services providing more opportunities for STI screening and consultations (24,28,30).

In this study, it was found that there would be more cases identified in males if testing was increased in this group. Modelling has shown that screening males may be cost-effective and help prevent new cases of chlamydia and pelvic inflammatory disease in females (31–33). Modelling by Qu *et al*. showed that for each male screened, 0.062 cases in males and 0.204 cases in females were prevented (32). Modelling also suggests that screening males should target high-risk individuals specifically (31,32). This could include settings where chlamydia rates are known to be high (such as in secondary and post-secondary schools), males who attend sexual health clinics, or within geographic areas with known clusters of cases (31).

Age is also associated with health care-seeking behaviour where younger people, those who would be most at risk for STIs, are less likely to seek healthcare (25). This could explain the persistence of chlamydia in the younger (under 30) population. It also indicates that more innovative solutions may be needed to curb infections if high-risk individuals are not seeking out testing and treatment. Increased communication around the nature of infections, risk of long-term sequelae, and recommended testing intervals could help younger individuals make more informed choices regarding STI testing. Innovative methods of outreach such as at-home test kits via an Internet and postal mail service, and expedited partner therapy, could help reach these groups (34–38). Studies have found that individuals who use internet-based STI testing have a higher rate of

repeat testing compared to individuals using clinic-based services (36). This could help increase testing rates in those less likely to seek out healthcare, including young males.

Limitations

There are several limitations to consider when making conclusions from this study. The largest limitation is that only testing data from provincial public health laboratories were included. When STI screening is completed through primary care physicians, testing is usually completed at a private laboratory such as LifeLabs or Dynacare, in Ontario, Canada (39). Tests performed through public health laboratories may be biased towards individuals screened at public health clinics, where individuals are often seeking STI testing. Additionally, focusing on Peel, Ontario as a subset of the Ontario population may not be representative of chlamydia dynamics in other health units. For these reasons, generalizability outside of the community that uses public health clinics should be cautioned.

Conclusion

In conclusion, the role of males in transmission dynamics of chlamydia requires further investigation. This study found that males are under-tested and if tested at the same rate as 20–29-year-old females, the observed maximally tested group for chlamydia, teenaged males (ages 15-19) and males 30–39 years would likely have higher observed average annual rates of chlamydia than what is identified through current testing. Programs that target hard-to-reach, high-risk males, specifically those under 30 years old could be critical for reducing the overall burden of chlamydia in this health unit.

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Appendix

Table A1: Summary of monthly chlamydia incidence and testing rates and percentage positivity by age group and sex in Peel, Ontario, identified through Public Health Ontario laboratories, 2010–2018.

Demographic		Monthly Median Value (Range)		
Sex	**Age Group**	**Cases**	**Tests**	**Positivity (%)**
Female	**15-19**	14 (4–26)	134 (96–194)	11.1 (3.8–18.5)
	20-29	29 (15–47)	552 (407–668)	5.3 (2.5–8.9)
	30-39	5 (1–14)	269 (166–365)	1.7 (0.3–6.3)
	40 and over	2 (0–6)	174 (90–268)	1.0 (0–3.3)
Male	**15-19**	5 (0–14)	43 (23–71)	12.5 (0–27.0)
	20-29	27 (14–42)	270 (182–443)	9.1 (4.4–16.2)
	30-39	7 (1–17)	132 (82–219)	5.6 (0.8–12.6)
	40 and over	4 (0–9)	105 (67–182)	3.4 (0–11.8)