ORIGINAL ARTICLE

Prevalence of *Mycoplasma genitalium* and macrolide resistance among asymptomatic people visiting a point of care service for rapid STI screening: a cross-sectional study

Miguel Fernández-Huerta,[1,2] María-Jesús Barberá,[2,3] Juliana Esperalba,[1,2] Candela Fernandez-Naval,[1,2] Marti Vall-Mayans,[2,3] Maider Arando,[2,3] Judit Serra-Pladevall,[1,2] Claudia Broto,[2,3] Francesc Zarzuela,[1,2] Ariadna Rando,[1,2] Tomas Pumarola,[1,2] Mateu Espasa[1,2]

► Additional material is published online only. To view please visit the journal online (http://dx.doi.org/10.1136/sextrans-2019-054124).

[1]Microbiology Department, Vall d'Hebron University Hospital, Barcelona, Spain
[2]Universitat Autònoma de Barcelona, Barcelona, Spain
[3]Sexually Transmitted Infections Unit, Vall d'Hebron University Hospital, Barcelona, Spain

Correspondence to
Dr Mateu Espasa, Microbiology, Vall d' Hebron University Hospital, Barcelona 08035, Spain; mespasa@tauli.cat

Received 11 May 2019
Revised 22 July 2019
Accepted 12 August 2019

To cite: Fernández-Huerta M, Barberá M-J, Esperalba J, *et al*. *Sex Transm Infect* Epub ahead of print: [*please include* Day Month Year]. doi:10.1136/sextrans-2019-054124

ABSTRACT

Objectives Although rapid screening and treatment programmes have been recently implemented to tackle STIs, testing *Mycoplasma genitalium* (MG) among asymptomatic populations is not currently recommended due to the lack of scientific evidence and the emergence of antibiotic resistance. The main objective of this study was to estimate the prevalence of MG and macrolide resistance among asymptomatic people visiting a point of care service for rapid STI screening and to identify risk factors associated with the acquisition of this infection.

Methods Between October 2017 and January 2018, a total of 890 asymptomatic individuals attending to the STI screening service Drassanes Exprés in Barcelona, Spain, were tested for MG and macrolide resistance using the molecular ResistancePlus MG assay (SpeeDx, Australia). Asymptomatically infected individuals were invited to attend the STI Unit for resistance-guided antimicrobial therapy.

Results Overall, the prevalence of MG was 7.4% (66/890; 95% CI 5.8% to 9.3%), being higher among men who have sex with men (MSM) (46/489) compared with heterosexual men and women (20/401; p=0.012). Macrolide resistance was found in 32/46 (69.6%; 95% CI 54.2% to 82.3%) MSM, while only 2/20 (10.0%; 95% CI 1.2% to 31.7%) infections among heterosexuals presented macrolide resistance-mediated mutations (p<0.001). MSM behaviour, receptive anal intercourse, HIV positive status, syphilis history and high-risk sexual activity (more than five sexual partners in the last 3 months) were significantly associated with MG infection. Furthermore, the resistance-guided therapy approach was implemented in 36/66 (54.6%) individuals.

Conclusions The research provides further data regarding the prevalence of MG and macrolide resistance among asymptomatic individuals. It also identifies higher risk subpopulations which might be targets for MG screening. Nevertheless, there is insufficient data to justify MG testing among asymptomatic individuals and current STI guidelines should be followed until evidence shows the cost and effectiveness of screening.

INTRODUCTION

STIs are a major problem worldwide. In fact, the WHO incorporated this issue in the *2030 Sustainable Development Goals* where, for example, an ambitious 90% reduction in the incidence of *Neisseria gonorrhoeae* (NG) globally was proposed as a key target.[1] Despite the efforts executed by the medical community, the number of reported infections caused by *Chlamydia trachomatis* (CT) and NG continues to rise annually; particularly among high-risk subpopulations such as men who have sex with men (MSM).[2]

NG and CT infections were traditionally managed only when patients presented with symptoms. However, rapid testing and treatment STI services have been recently implemented in asymptomatic individuals to putatively reduce complications and control STI transmission and spread since most infections are asymptomatic and may persist in time. Furthermore, current international guidelines already recommend routine screening for NG and CT; especially among sexually active young people and MSM.[3 4] Although not widely evidenced, these screening programmes may, indeed, reduce the prevalence of STIs.[5] In addition, these programmes may also result in selection pressure for antibiotic resistance development not only in NG[6] but also in *Mycoplasma genitalium* (MG) if an undetected co-infection is present and azithromycin is prescribed.[7]

MG is a major cause of urethritis, accounting for 15%–20% of non-gonococcal urethritis (NGU) in men.[8] In women, this STI is associated with cervicitis, pelvic inflammatory disease (PID), preterm birth and spontaneous abortion.[8 9] Since doxycycline demonstrated very poor efficacy eradicating MG infections,[10 11] azithromycin has instead been used and remains the recommended first-line treatment.[12] However, the widespread use of this antibiotic for the syndromic management of STIs may have contributed to the emergence of macrolide resistance (McrR) in MG worldwide.[3 3 4 4–18] Testing for MG among asymptomatic individuals has been suggested[19]; but experts currently plead to avoid

widespread asymptomatic screenings due to the lack of scientific evidence, the disturbing emergence of antibiotic resistance in MG, particularly to azithromycin, and the consequent use of costly antibiotics (that may also cause serious side effects).[7 12 20] In fact, recent studies attempt to evaluate the impact and effectiveness of these screening strategies but ultimately recommend further empirical work to improve understanding on MG and elucidate the real impact of these proposals.[21 22]

In response to this challenge, the primary objective of this study was to estimate the prevalence of MG and McrR among asymptomatic people visiting a point of care (POC) service for rapid STI screening. Second, we aimed to identify factors associated with the acquisition of MG infection since certain subpopulations with large MG endemicity could be potential targets for screening strategies. Also, we evaluated a resistance-guided therapy (RGT) approach for MG treatment.

METHODS

A cross-sectional study was conducted, between October 2017 and January 2018, among asymptomatic people attending a POC STI screening service (Drassanes Exprés-DrasExp) located in Barcelona. A written information report about the study was provided to all attendees and those who accepted signed an informed-consent document. The inclusion criteria were complete STI screening done and acceptance to participate in the study. Thus, in addition to the routine STI screening provided at DrasExp, participants were also tested for MG and McrR.

Drassanes Exprés

DrasExp Programme is a publicly funded POC service for rapid STI screening belonging to Vall d'Hebron University Hospital in Barcelona, Spain. Thus, not only HIV and syphilis but also CT and NG testing is routinely offered to asymptomatic men and women (older than 18 years).

On arrival at DraExp, users complete a short epidemiological questionnaire into the Laboratory Internal Software (LIS) including: age, gender, gender of sex partners, country of origin, sexual practices (oral, vaginal and/or anal sex), number of sexual partners in the last 3 months and HIV and syphilis history. Based on the self-reported answers pointed, the LIS suggests the specimens required for the appropriate screening. Then, a self-sampling collection is performed by the users. Nursing staff also collect blood samples for HIV and syphilis testing if required. Finally, samples are delivered to the laboratory where the microbiological testing takes place. As soon as the results are available and validated (<6 hours), the LIS automatically delivers them to users by SMS/email. Those with positive results are then invited to attend to the STI Unit Vall d'Hebron-Drassanes, also belonging to Vall d'Hebron University Hospital, for the proper management of the infection.

Users excluded of this screening service are: patients reporting symptoms, sexual contacts of known infected partners sent to the STI Unit for specific clinical attendance and those individuals having been screened at DrasExp in the last 3 months as stated by the STI guidelines.[3 4]

Laboratory and clinical procedures

STI screening tests are routinely performed at the laboratory of DraExp. Additionally, attendees enrolled in the study were also tested for MG and McrR genotypic markers using the ResistancePlus MG kit (SpeeDx, Australia) according to the manufacturer's instructions. First-void urines (Vacumed Urine (FL Medical, Italy), 10 mL container), vaginal and rectal flocked swabs (DeltaSwab ViCUM (Deltalab, Spain), 2 mL transport media) were collected and used for this MG screening. The ResistancePlus MG assay is a multiplex real-time PCR for detection of MG and five mutations (A2058G, A2059G, A2058C, A2059C and A2058T; *Escherichia coli* numbering) located in domain V of the 23S ribosomal RNA (rRNA) and associated with phenotypic McrR.[23] Briefly, 200 µL of sample was extracted using the MagNA Pure 96 platform (Roche Diagnostics, USA) and eluted in 100 µL of MagNA Pure elution buffer with the SV Viral NA extraction kit on the Universal Pathogen protocol (Roche Diagnostics). Then, 5 µL of DNA was amplified in a 20 µL reaction volume using a LightCycler 480 Instrument II (Roche Diagnostics). Analysis of the results was performed using the supplied software. McrR-mediating mutations found in MG were retrospectively confirmed by sequencing using a previously described methodology.[17]

Since the MG screening result was delayed by few days, individuals recruited received a second SMS/email regarding, exclusively, the study result. Thus, those patients testing positive for MG were invited to attend the STI Unit for RGT.[12 24 25] Treatment outcomes were retrospectively registered through a comprehensive review of the medical record. Cured was defined as a negative microbiological test of cure (TOC), executed at least 3 weeks after antibiotic therapy.[12] Infected participants who did not return for clinical control were classified as lost to follow-up and no active-recruitment strategies were implemented.

Descriptive and statistical analyses

Characteristics of the study cohort were collected from the LIS questionnaire. Sexual conduct was defined regarding the gender of sexual partners reported by the participant. Statistical analyses were performed using Stata (StataCorp, USA). Univariate analyses were performed to examine the relationships between each variable and MG infection. ORs and their corresponding 95% CIs were calculated. Differences with $p<0.05$ were considered statistically significant.

RESULTS

A total of 1189 users attended to DrasExp during the study period. Of them, 890 (74.9%) accepted to participate in the study. Online supplementary table in supplementary material compares the sociodemographic characteristics of both recruited and non-recruited individuals. There were more MSM among the participants (54.9% vs 39.5%, $p<0.001$), contributing also to a major proportion of high-risk sexual activity and syphilis history among the recruited population. Thus, a total of 1404 specimens were collected and tested for MG and McrR. Samples consisted of 519 rectal swabs, 222 vaginal swabs and 663 first-void urines.

Characteristics of study population

The characteristics of the study population grouped by sexual behaviour are summarised in table 1. The main screened group were MSM, with 489 (54.9%; 95% CI 51.6% to 58.2%) subjects of whom 52 (10.6%; 95% CI, 0.8% to 13.7%) also reported engaging in heterosexual behaviours. The proportion of participants reporting more than five sexual partners in the preceding 3 months was much higher among MSM (38.9%; 95% CI, 34.5% to 43.3%) compared with the heterosexual population (16.0%; 95% CI 12.5% to 19.9%; $p<0.001$). The majority of MSM recruited reported receptive anal intercourse (96.3%; 95% CI 94.2% to 97.8%) while this sexual practice was only occasional among women (26.1%; 95% CI 20.5% to 32.4%).

Table 1 Characteristics of the 890 asymptomatic individuals studied for *Mycoplasma genitalium* and macrolide resistance grouped by sexual behaviour

Characteristics	MSM (n=489) No.; % (95% CI)	MSW (n=179) No.; % (95% CI)	Women (n=222) No.; % (95% CI)	P value*
Median age (IQR)	34.1 (27.7 to 41.8)	30.5 (26.5 to 37.4)	28.9 (24.3 to 36.7)	<0.001
Origin				
Spanish	311; 63.6 (59.2 to 67.9)	99; 55.3 (47.7 to 62.7)	117; 52.7 (45.9 to 59.4)	0.011
Non-Spanish	178; 36.4 (32.1 to 40.8)	80; 44.7 (37.3 to 52.3)	105; 47.3 (40.6 to 54.1)	
Sexual partners (last 3 months)				
1–5	299; 61.1 (56.7 to 65.5)	153; 85.5 (79.4 to 90.3)	184; 82.9 (77.3 to 87.6)	<0.001
6–20	162; 33.1 (29.0 to 37.5)	24; 13.4 (8.8 to 19.3)	27; 12.2 (8.2 to 17.2)	
>20	28; 5.7 (3.8 to 8.2)	2; 1.1 (0.1 to 4.0)	11; 5.0 (2.5 to 8.7)	
HIV positive	84†; 17.2 (13.9 to 20.8)	0; 0.0 (0.0 to 2.0)	0; 0.0 (0.0 to 1.6)	<0.001
Syphilis history	177; 36.2 (31.9 to 40.6)	1; 0.6 (0.0 to 3.1)	1; 0.5 (0.0 to 2.5)	<0.001
Specimens				
Rectal swab	471; 96.3 (94.2 to 97.8)	–	58; 26.1 (20.5 to 32.4)	
Vaginal swab	–	–	222; 100.0 (98.4 to 100.0)	
First void urine	489; 100.0 (99.2 to 100.0)	179; 100.0 (98.0 to 100.0)	–	
CT infection	48‡; 9.8 (7.3 to 12.8)	11; 6.1 (3.1 to 10.7)	18§; 8.1 (4.9 to 12.5)	0.310
NG infection	67; 13.7 (10.8 to 17.1)	0; 0.0 (0.0 to 0.2)	9; 4.1 (1.9 to 7.6)	<0.001

*χ^2 tests for categorical variables and Mann-Whitney U test for quantitative variables.
†Five HIV positive cases were new diagnoses.
‡Eleven MSM individuals had a CT and NG co-infection.
§One woman was co-infected with CT and NG.
MSM, men who have sex with men; MSW, men who have sex with women; CT, *Chlamydia trachomatis*; NG, *Neisseria gonorrhoeae*.

Regarding the routine STI screenings performed at DraExp, five new HIV cases (one co-infected with MG), all in MSM, were diagnosed during the study period. Additionally, overall CT prevalence was 8.7% (95% CI 6.9% to 10.7%) with no statistically significant differences between groups (p=0.310). On the other hand, overall NG infection prevalence was slightly lower (8.5%; 95% CI 6.8% to 10.6%) being more prevalent among MSM compared with MSW and women (p<0.001).

Table 2 Prevalence of *Mycoplasma genitalium* and macrolide resistance among asymptomatic individuals

	Mycoplasma genitalium prevalence		
	Macrolide resistance prevalence		
Population	Resistant MG N.; % (95% CI)	Total MG N.; % (95% CI)	Total individuals tested (N)
Women	1; 10.0 (0.3 to 44.5)	10; 4.5 (2.2 to 8.1)	222
Vagina	1; 14.3 (0.4 to 57.9)	7*; 3.2 (1.3 to 6.4)	222
Rectum	0; 0.0 (0.0 to 60.2)	4; 6.9 (1.9 to 16.7)	58
MSW†	1; 10.0 (0.3 to 44.5)	10; 5.6 (2.7 to 10.0)	179
MSM	32; 69.6 (54.2 to 82.3)	46; 9.4 (7.0 to 12.3)	489
Urethra	8‡; 88.9 (51.8 to 99.7)	9‡; 1.8 (0.8 to 3.5)	489
Rectum	27; 67.5 (50.9 to 81.4)	40; 8.5 (6.1 to 11.4)	471
Total	34; 51.5 (38.9 to 64.0)	66; 7.4 (5.8 to 9.3)	890

Percentages of macrolide resistance are calculated from the MG infections reported.
*One woman had infections in both vagina and rectum.
†All infections in MSW occurred in urethra.
‡Three MSM had infections in both urethra and rectum.
MG, *Mycoplasma genitalium*; MSM, men who have sex with men; MSW, men who have sex with women.

Prevalence of MG and macrolide resistance

The table 2 shows the prevalence of MG and McrR grouped by sexual behaviour. A total of 70 specimens (5.0%; 95% CI 3.9% to 6.3%) from 66 people (7.4%; 95% CI 5.8% to 9.3%) were positive for MG.

The prevalence of rectal MG infections among MSM reporting anal intercourse was 8.5% (95% CI 6.1% to 11.4%), like the one reported in women (6.9%; 95% CI 1.9% to 16.7%, p=0.678). On the other hand, the prevalence of urethral MG infection among MSW was 5.6% (95% CI 2.7% to 10.0%), significantly higher than among MSM (1.9%; 95% CI 1.9% to 3.5%, p=0.011). Furthermore, those MSM engaging also heterosexual behaviours had a significantly higher prevalence of urethral infection (5.8%; 95% CI 1.2% to 15.9%) compared with MSM with no female sexual partners (1.4%; 95% CI 0.5% to 3.0%, p=0.026). In fact, there were no differences in urethral infection prevalence between MSW and bisexual men (p=0.960). Overall, men reporting vaginal sex had higher risk for urethral MG infection (4.28; 95% CI 1.61 to 11.43).

McrR was detected in 37 MG-positive specimens (52.9%; 95% CI 40.6% to 64.9%) from 34 individuals (51.5%; 95% CI 38.9% to 64.0%).

No statistical differences in MG infection (p=0.621) and McrR (p=1.000) were found between women and MSW. However, prevalence of MG infection (p=0.012) and McrR (p<0.001) was markedly higher among MSM compared with heterosexual men and women. Six out of 52 MSM reporting bisexual behaviours (11.5%; 95% CI 4.4 to 23.4) had a MG infection, higher prevalence if compared with MSM with no female sexual partners (9.2%; 95% CI 6.6% to 12.3%, p=0.578).

The sequencing of the 23S rRNA confirmed 29 of the 34 McrR cases. Thus, 18 infections (62.1%; 95% CI 42.3% to 79.3%) harboured SNPs at position A2058G, nine (31.0%; 95% CI 15.3% to 50.8%) at position A2059G and two (6.9%; 95% CI 0.8% to 22.8%) were mixed infections with mutants at both A2058G and

Table 3 Risk factors associated with *Mycoplasma genitalium* infection

Characteristic (N=890)	People with characteristics (N, %)	MG infections (N, %)	Crude OR (95% CI)	P value*
Age (IQR)	32.1 (26.5–40.0)	33.8 (28.2–40.1)	1.01 (0.98 to 1.03)	0.624
Sexual conduct				
Women	222 (24.9)	10 (4.5)	1	
MSW	179 (20.2)	10 (5.6)	1.25 (0.51 to 3.08)	0.621
MSM	489 (54.9)	46 (9.4)	2.20 (1.09 to 4.45)	0.028
Origin				
Spanish	527 (59.2)	33 (6.2)	1	
Non-Spanish	363 (40.8)	33 (9.1)	1.50 (0.90 to 2.47)	0.115
Sexual partners (last 3 months)				
1–5	636 (71.5)	37 (5.8)	1	
6–20	213 (23.9)	22 (10.3)	1.87 (1.07 to 3.24)	0.027
>20	41 (4.6)	7 (17.1)	3.33 (1.38 to 8.02)	0.007
Active oral sex				
No	192 (21.6)	10 (5.2)	1	
Yes	698 (78.4)	56 (10.6)	1.59 (0.79 to 3.17)	0.191
Receptive anal intercourse				
No	361 (40.6)	14 (3.9)	1	
Yes	529 (59.4)	52 (9.8)	2.70 (1.47 to 4.95)	0.001
HIV positive				
No	806 (90.6)	53 (6.6)	1	
Yes	84 (9.4)	13 (15.5)	2.60 (1.35 to 5.00)	0.004
Syphilis history				
No	711 (79.9)	45 (6.3)	1	
Yes	179 (20.1)	21 (11.7)	1.97 (1.14 to 3.40)	0.015
CT co-infection				
No	813 (91.3)	60 (7.4)	1	
Yes	77 (8.7)	6 (7.8)	1.07 (0.45 to 2.58)	0.868
NG co-infection				
No	814 (91.5)	60 (7.4)	1	
Yes	76 (8.5)	6 (7.9)	1.06 (0.44 to 2.54)	0.895

*χ^2 tests for categorical variables and Mann-Whitney U test for quantitative variables.
CT, *Chlamydia trachomatis*;MG, *Mycoplasma genitalium*; MSM, men who have sex with men; MSW, men who have sex with women; NG, *Neisseria gonorrhoeae*.

A2059G positions. The remaining five individuals that tested positive for resistant MG were unable to be sequenced.

Additionally, MG co-infection was found in 8.5% (95% CI 4.5% to 14.4%) of participants testing positive for CT or NG.

Factors associated with MG infection

The univariate analyses described in table 3 suggests that the main risk factors associated with MG infection were: MSM behaviour, receptive anal intercourse, HIV positive status, syphilis history and high-risk sexual behaviours (more than five sexual partners in the last 3 months).

Treatment outcomes

Of the 66 participants infected with MG during the study period, 36 (54.6%) returned for clinical attendance and were treated following the RGT (figure 1). The median time between the DrasExp visit and the beginning of treatment was 15 days (IQR 9–22 days). A total of 24 (66.7%) users completed the proper treatment follow-up with a negative TOC. The median time between treatment and the TOC was 35 days (IQR 32–42).

Only one putative treatment failure case was detected in a wild-type infection treated with extended-dose azithromycin. In this case, not only MG but also a new CT infection was detected in the TOC, 3 months later. Furthermore, the individual reported condomless anal intercourse with multiple partners. Unfortunately, the McrR status was not determined in this second episode. Since the individual had been previously treated with azithromycin, the patient received moxifloxacin and cured.

DISCUSSION

Despite MG being isolated in 1981, nucleic acid amplification tests were not developed until the early 1990s.[8] Henceforth, its pathogenic role has since been established and MG has become a serious concern, especially after the emergence and spread of McrR worldwide.[14–18]

The study aimed to estimate the prevalence of MG and McrR among asymptomatic people visiting a POC service for rapid STI screening. The overall prevalence of MG in our cohort was 7.4%, with higher estimates among MSM (9.4%) compared with heterosexuals (5.0%). There are few investigations on asymptomatically MG-infected people,[26–28] all reporting lower prevalence. To our knowledge, there are no previous estimates of McrR in MG among asymptomatically infected people. Thus, the overall prevalence of McrR was 51.5% in our cohort, being seven times more prevalent among MSM if compared with heterosexuals. Similar resistance rates have been recently reported but in different settings in Spain.[14]

The statistical analyses revealed that MSM behaviour, receptive anal intercourse, HIV positive status, syphilis history and high-risk sexual activity (more than five sexual partners in the

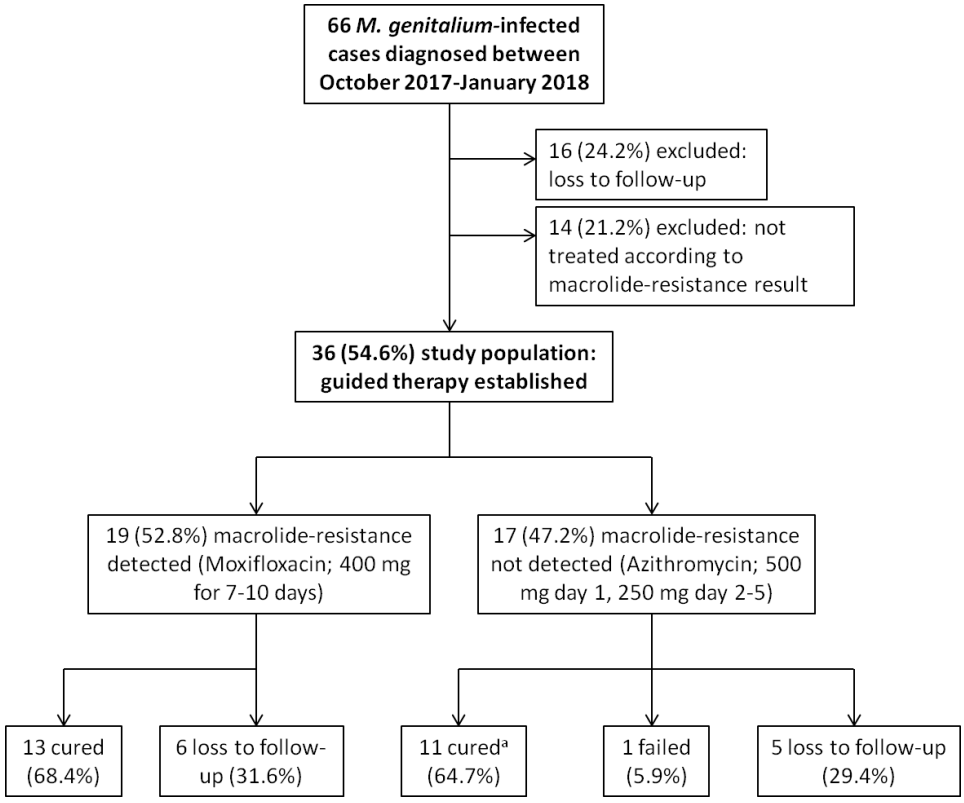

Figure 1 Cases and outcomes of resistance-guided treatment (RGT) for *Mycoplasma genitalium* asymptomatic infections. [a]Two individuals received not the extended but the single-dose 1 g azithromycin regimen due to co-existing *Neisseria gonorrhoeae* and *Chlamydia trachomatis*, respectively.

last 3 months) were significantly associated with MG infection. Furthermore, the study showed that MSW and bisexual men had a similar prevalence of urethral MG infection, much higher when compared with MSM with no female sexual partners. Several studies have reported that urethral MG infection is more associated with unprotected vaginal sex rather than anal sex.[28 29] Behavioural patterns, but also a better environment for MG fitness in vagina, resulting in higher bacterial loads, could explain this fact. Some investigations conclude that vaginal intercourse is a major route of transmission for MG and suggest that strengthening the screening of this bacterium among bisexual men could be an important strategy to control the infection.[28]

Additionally, the study evaluated a RGT approach for MG screening using the workflow of DrasExp. Despite the limited number of MG infected individuals who returned for clinical assistance, no treatment failures were detected among macrolide-resistant infections using moxifloxacin and only one probable azithromycin failure case was reported in a wild-type infection. Similar rates of McrR selection were previously reported using a guided sequential-treatment approach.[25]

Despite the arguments against MG screening, there are some other points that should be considered when discussing this issue. First, MG usually causes urethritis in men but it is also associated with very serious adverse outcomes in women ranging from cervicitis to preterm birth, spontaneous abortion, PID and infertility.[8 9 12] Second, although poorly established, the relation between MG and HIV suggests that the bacterial infection may facilitate HIV acquisition through disruptive and inflammatory processes in the anogenital mucosa during exposure.[30] Last, the present study demonstrates that MG prevalence is similar to the prevalence of CT and NG. Given the similarities, it may be paradoxical why for instance NG screening, which also may select for antimicrobial resistance,[6] is widely recommended while MG is not.[3 12] Additionally, CT and NG screening strategies may facilitate McrR spread in MG if an undetected co-infection (8.5% in our cohort) is present and azithromycin is prescribed.[7]

Considering that a sample of subjects was selected, some limitations to our report must be addressed. First of all, there were more MSM among the recruited population, contributing also to a higher proportion of high-risk sexual activity and syphilis history. This may have biased the results overestimating the prevalence of MG and McrR among the study population. Additionally, we should note that the study population, as a group attending sexual health testing, is at higher risk STIs than the general population. Second, the epidemiological information was collected from a computer-based questionnaire that users had to complete, always assisted by nursing staff. Consequently, the information may be susceptible to misreporting. Third, no active-recruitment strategies were implemented for the MG-positive individuals to be treated; so, only the information report given at DrasExp and the automatically delivered SMS accounted for the low adherence of participants to the RGT protocol. Of note, likely asymptomatic people are not fully aware and worried about the impact of MG infection on health. In addition, registrations of treatment outcomes were collected retrospectively and there were many tracking losses that could have also limited the strength of the results. However, our results showed that individuals who followed the RGT had a high cure rate, in accordance with results by Read *et al*.[25] Regarding the laboratory techniques, 200 µL of neat urine was extracted without any concentration procedure. This may have underestimated MG infection in urethra although the positivity rate in this location is the expected one compared with previous investigations.[28] Finally, the cross-sectional design of the study cannot provide key evidence about the possible impact of MG screening on public health.

In conclusion, the research provides additional data regarding the prevalence of MG and McrR among asymptomatic people at risk to acquire STIs. Furthermore, the results of the investigation also point subpopulations and sexual practices at higher risk for MG infection, which could be potential targets for MG screening in terms of infection control. Lastly, the research successfully engages MG testing to a POC service for rapid STI screening, establishing the basics for future screening strategies. Nevertheless, current available evidence is insufficient to justify MG screening among any defined asymptomatic population. So, current guidelines should be followed until more empirical studies improve the understanding of MG natural history and assess the cost and effectiveness of screening.

Key messages

- The prevalence of *Mycoplasma genitalium* (MG) asymptomatically infected individuals among people visiting a point of care service for rapid STI screening was 7.4%.
- The prevalence of MG and macrolide resistance was much higher among men who have sex with men compared with heterosexual men and women.
- The resistance-guided therapy protocol demonstrated high efficacy eradicating MG infections and preventing new MG macrolide-resistant cases.
- The current limited understanding in MG infection natural history is insufficient to justify MG testing and treatment among asymptomatic individuals.

Handling editor Catherine A Ison

Acknowledgements We thank all the staff of the Drassanes Exprés Programme, especially the nursing and laboratory teams for their essential work at the STI screening service. We also thank people at the Microbiology Department of the Vall d'Hebron University Hospital, particularly Dr Andrés Antón and Cristina Pinatar. Finally, we would like to thank SpeeDx Pty Ltd for their support.

Contributors ME led the design and conduct of the study. MF-H and CFN contributed to design the study and performed the experimental work. MF-H, AR and ME analysed the results. MF-H wrote the initial draft with ME. MJ-B, JE, CF-N, MV-M, MA, JS-P, CB, FZ, AR and TP reviewed and commented on the manuscript.

Funding SpeeDx Pty Ltd supplied all the reagents for molecular testing of Mycoplasma genitalium.

Disclaimer The conclusions in this report are those of the authors and do not represent the official position of any national/international institution. The funders had no role in study design, data collection and analyses, decision to publish or preparation of the manuscript.

Competing interests CFN is a researcher who has received partial salary support from SpeeDx Pty Ltd. MF-H and ME have participated in symposiums organised by SpeeDx Pty Ltd.

Patient consent for publication Not required.

Ethics approval Ethical approval for the study was obtained from the Vall d'Hebron University Hospital Ethics Committee (209/2017).

Provenance and peer review Not commissioned; externally peer reviewed.

Data availability statement All data relevant to the study are included in the article or uploaded as supplementary information.

REFERENCES

1 World Health Organization. Global health sector strategy on sexually transmitted infections, 2016-2017. Available: www.who.int [Accessed June 2016].
2 Yarbrough ML, Burnham C-AD. The ABCs of STIs: an update on sexually transmitted infections. *Clin Chem* 2016;62:811–23.
3 Bignell C, Unemo M, European STI Guidelines Editorial Board. 2012 European guideline on the diagnosis and treatment of gonorrhoea in adults. *Int J STD AIDS* 2013;24:85–92.
4 Lanjouw E, Ouburg S, de Vries HJ, *et al*. 2015 European guideline on the management of *Chlamydia trachomatis* infections. *Int J STD AIDS* 2016;27:333–48.
5 Tsoumanis A, Hens N, Kenyon CR. Is screening for *Chlamydia* and gonorrhea in men who have sex with men associated with reduction of the prevalence of these infections? A systematic review of observational studies. *Sex Transm Dis* 2018;45:615–22.
6 Fingerhuth SM, Bonhoeffer S, Low N, *et al*. Antibiotic-resistant *Neisseria gonorrhoeae* spread faster with more treatment, not more sexual partners. *PLoS Pathog* 2016;12:e1005611.
7 Bradshaw CS, Horner PJ, Jensen JS, *et al*. Syndromic management of STIs and the threat of untreatable *Mycoplasma genitalium*. *Lancet Infect Dis* 2018;18:251–2.
8 Taylor-Robinson D, Jensen JS. *Mycoplasma genitalium*: from Chrysalis to multicolored butterfly. *Clin Microbiol Rev* 2011;24:498–514.
9 Lis R, Rowhani-Rahbar A, Manhart LE. *Mycoplasma genitalium* infection and female reproductive tract disease: a meta-analysis. *Clin Infect Dis.* 2015;61:418–26.
10 Mena LA, Mroczkowski TF, Nsuami M, *et al*. A randomized comparison of azithromycin and doxycycline for the treatment of *Mycoplasma genitalium* – positive urethritis in men. *Clin Infect Dis* 2009;48:1649–54.
11 Manhart LE, Gillespie CW, Lowens MS, *et al*. Standard treatment regimens for nongonococcal urethritis have similar but declining cure rates: a randomized controlled trial. *Clin Infect Dis* 2013;56:934–42.
12 Jensen JS, Cusini M, Gomberg M, *et al*. European guideline on *Mycoplasma genitalium* infections. *J Eur Acad Dermatol Venereol* 2016;2016:1686–93.
13 Lau A, Bradshaw CS, Lewis D, *et al*. The efficacy of azithromycin for the treatment of genital *Mycoplasma genitalium*: a systematic review and meta-analysis. *Clin Infect Dis.* 2015;61:1389–99.
14 Barberá MJ, Fernández-Huerta M, Jensen JS, *et al*. *Mycoplasma genitalium* macrolide and fluoroquinolone resistance: prevalence and risk factors among a 2013-2014 cohort of patients in Barcelona, Spain. *Sex Transm Dis* 2017;44:457–62.
15 Getman D, Jiang A, O'Donnell M, *et al*. *Mycoplasma genitalium* prevalence, coinfection, and macrolide antibiotic resistance frequency in a multicenter clinical study cohort in the United States. *J Clin Microbiol* 2016;54:2278–83.
16 Deguchi T, Yasuda M, Horie K, *et al*. Drug resistance-associated mutations in *Mycoplasma genitalium* in female sex workers, Japan. *Emerg Infect Dis* 2015;21:1062–4.
17 Jensen JS, Bradshaw CS, Tabrizi SN, *et al*. Azithromycin treatment failure in *Mycoplasma genitalium* – positive patients with nongonococcal urethritis is associated with induced macrolide resistance. *Clin Infect Dis* 2008;47:1546–53.
18 Ito S, Shimada Y, Yamaguchi Y, *et al*. Selection of *Mycoplasma genitalium* strains harbouring macrolide resistance-associated 23S rRNA mutations by treatment with a single 1 G dose of azithromycin. *Sex Transm Infect* 2011;87:412–4.
19 Manhart LE. Has the time come to systematically test for *Mycoplasma genitalium*? *Sex Transm Dis* 2009;36:607–8.
20 Golden MR, Workowski KA, Bolan G. Developing a public health response to *Mycoplasma genitalium*. *J Infect Dis* 2017;216(suppl_2):S420–S426.
21 Birger R, Saunders J, Estcourt C, *et al*. Should we screen for the sexually-transmitted infection *Mycoplasma genitalium*? Evidence synthesis using a transmission-dynamic model. *Sci Rep* 2017;7:16162.
22 Baumann L, Cina M, Egli-Gany D, *et al*. Prevalence of *Mycoplasma genitalium* in different population groups: systematic review andmeta-analysis. *Sex Transm Infect* 2018;94:255–62.
23 Tabrizi SN, Tan LY, Walker S, *et al*. Multiplex assay for simultaneous detection of *Mycoplasma genitalium* and macrolide resistance using PlexZyme and PlexPrime technology. *PLoS One* 2016;11:e0156740.
24 Unemo M, Jensen JS. Antimicrobial-resistant sexually transmitted infections: gonorrhea and *Mycoplasma genitalium*. *Nat Rev Urol* 2017;93:S65–8.
25 Read TRH, Fairley CK, Murray GL, *et al*. Outcomes of resistance-guided sequential treatment of *Mycoplasma genitalium* infections: a prospective evaluation. *Clin Infect Dis* 2019;68:554–60.
26 Sonnenberg P, Ison CA, Clifton S, *et al*. Epidemiology of *Mycoplasma genitalium* in British men and women aged 16–44 years: evidence from the third National Survey of Sexual Attitudes and Lifestyles (Natsal-3). *Int J Epidemiol* 2015;44:1982–94.
27 Seña AC, Lee JY, Schwebke J, *et al*. A silent epidemic: the prevalence, incidence and persistence of *Mycoplasma genitalium* among young, asymptomatic high-risk women in the United States. *Clin Infect Dis* 2018;67:73–9.
28 Zheng B-jie, Yin Y-ping, Han Y, *et al*. The prevalence of urethral and rectal *Mycoplasma genitalium* among men who have sex with men in China, a cross-sectional study. *BMC Public Health* 2014;14:195.
29 Slifirski JB, Vodstrcil LA, Fairley CK, *et al*. *Mycoplasma genitalium* infection in adults reporting sexual contact with infected partners, Australia, 2008–2016. *Emerg Infect Dis* 2017;23:1826–33.
30 Napierala Mavedzenge S, Weiss HA. Association of *Mycoplasma genitalium* and HIV infection: a systematic review and meta-analysis. *AIDS* 2009;23:611–20.