Title: Prevalence and factors associated with multi-drug resistant *Neisseria gonorrhoeae* in England and Wales between 2004 and 2015: analysis of annual cross-sectional surveillance surveys

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Synopsis

Objectives: To describe trends in prevalence, susceptibility profile and risk factors for multi-drug resistant *Neisseria gonorrhoeae* (MDR-NG) in England and Wales.

Methods: Isolates from 16,242 gonorrhoea episodes at sexual health clinics within the Gonococcal Resistance to Antimicrobials Surveillance Programme (GRASP) underwent antimicrobial susceptibility testing. MDR-NG was defined as resistance to ceftriaxone, cefixime, or azithromycin, *PLUS* ≥2 of: penicillin, ciprofloxacin, spectinomycin. Trends in resistance are presented for 2004-2015; prevalence and logistic regression analyses for MDR-NG cover the period of the most recent treatment guideline (ceftriaxone plus azithromycin), 2011-15.

Results: Between 2004-2015, the proportion of NG isolates fully susceptible to all antimicrobial classes fell from 80% to 46%, with the proportion resistant to multiple (two or more) classes increasing from 7.3% to 17.5%. In 2011-2015, 3.5% of isolates were MDR-NG, most of which were resistant to cefixime (100% in 2011, decreasing to 36.9% in 2015) and/or azithromycin (4.2% in 2011, increasing to 84.3% in 2015). After excluding azithromycin-resistant isolates, modal azithromycin MICs were higher in MDR versus non-MDR isolates (0.5 mg/L versus 0.125 mg/L), with similar results for ceftriaxone (modal MICs 0.03 mg/L versus ≤0.002 mg/L). After adjustment for confounders, MDR-NG was more common among isolates from heterosexual men, although absolute differences in prevalence were small (4.6% versus 3.3% (MSM) and 2.5% (women)).

Conclusions: NG is becoming less susceptible to available antimicrobials. Since 2011, a minority of isolates were MDR-NG, however MICs of azithromycin or ceftriaxone (first line therapies) for many of these were elevated. These findings highlight the importance of continued antimicrobial stewardship for gonorrhoea.

Introduction

Gonorrhoea, caused by the bacterium *Neisseria gonorrhoeae* (NG), is the second most commonly diagnosed bacterial Sexually Transmitted Infection (STI) in England.[1] Although estimated prevalence in community settings in Britain is low (<0.1%),[2] recent years have seen worrying increases in diagnoses among men who have sex with men (MSM).[1,3] The history of rapid development of resistance to antimicrobials used for treatment has made antimicrobial resistant (AMR) NG a major global health concern and a priority area for Public Health England (PHE).[4–9] Extended-spectrum cephalosporins (ESCs) are the most recent class of antimicrobial introduced to treat gonorrhoea, and represent a 'last-line' treatment option, with no new antimicrobials available and few in development.[4,5,9,10] Since 2011, recommended first-line treatment in the UK has been dual therapy with 500 mg intramuscularly-injectable ceftriaxone (an ESC) plus 1 g oral azithromycin, in an attempt to prevent sustained resistance to ESCs becoming established,[10,11] an approach which has been implemented in many other regions globally[12]. Although resistance to ceftriaxone is rare, recent increases in azithromycin resistance have been described, including high-level resistance.[13,14] There is also evidence of *N. gonorrhoeae* becoming gradually less susceptible to ceftriaxone,[15] and the first reported global case of dual treatment failure was also detected in the UK.[16] Many have now highlighted that multi-drug resistant *N. gonorrhoeae* (MDR-NG) poses a serious threat, and untreatable gonorrhoea is an increasingly plausible prospect.[5,7,9,10]

It is critical to understand the epidemiology of AMR NG to inform treatment guidelines and prevention and control measures, in addition to wider gonorrhoea prevention efforts.[4,7,17] The Gonococcal Resistance to Antimicrobials Surveillance Project (GRASP) was established in 2000 to monitor trends and provide information on determinants of AMR NG in England and Wales,[18] and combines susceptibility data with demographic, behavioural, and clinical information from those diagnosed with gonorrhoea at selected specialist sexual health services. GRASP has provided

detailed insight into patterns of AMR in NG and informed changes to treatment guidelines.[10,19] To date, GRASP data on trends and risk factors for individual antimicrobials have been reported,[13,15,20,21] but detailed analyses of MDR-NG have not. This project seeks to fill this knowledge gap by investigating the prevalence, resistance profile, and risk factors for MDR-NG in England and Wales in order to understand implications for the future treatment of gonorrhoea, and contribute to the scant epidemiological literature on MDR-NG.[7,22–27]

Methods

Ethics and governance

PHE has permission to handle data obtained by GRASP under section 251 of the UK National Health Service Act of 2006, which was renewed annually by the ethics and confidentiality committee of the National Information Governance Board until 2013. Since then, the power of approval of public health surveillance activity has been granted directly to PHE.

Data sources

Detailed descriptions of GRASP methodology have been published previously.[13] Briefly, isolates from individuals with gonorrhoea attending 27 specialist sexual health clinics in England and Wales during a three month period each summer between 2004 and 2015 were cultured and submitted to PHE for susceptibility testing. An agar dilution method was used to determine MICs for ceftriaxone, cefixime, spectinomycin, azithromycin, penicillin, and ciprofloxacin. Susceptibility data were matched to demographic, clinical, and behavioural data submitted by the clinics. All patients diagnosed with gonorrhoea during this period were eligible for inclusion, however approximately half of episodes did not have susceptibility data due to culture not being attempted or successful.[28]

Azithromycin MIC data

The Diagnostic Sensitivity Test (DST) medium used for susceptibility testing for GRASP isolates was changed in 2015, whereupon MICs of azithromycin, and subsequently the proportion of resistant isolates, increased.[13] A validation study compared the MICs determined by the new and old DST agars and found MICs of azithromycin were higher by approximately one dilution using the new DST medium. The new DST medium provided better pH and physiological conditions for growth of fastidious strains of *N. gonorrhoeae* which subsequently resulted in more reliable azithromycin MIC determination; this was also confirmed by local quality assurance data.[13] Azithromycin MIC data for 2013 and 2014 (the years in which the problems with growth on the old DST medium were seen) were therefore adjusted upwards by a factor of one dilution to enable more accurate description of trends over time.

Definition of MDR-NG

Defining MDR-NG is challenging due to differences in first-line treatments over time and internationally. We therefore present analyses of i) patterns of AMR-NG over time, including resistance or decreased susceptibility to multiple classes of antimicrobials, to facilitate international and longer-term comparisons; ii) MDR-NG defined as relevant to current clinical practice in the UK. For the latter, we adapted the working definition proposed by Tapsall *et al* (2009): resistance or decreased susceptibility to **one or more** antimicrobial in widespread use to treat gonorrhoea (category 1), and resistance to **two or more** antimicrobials in less frequent use/little use but proposed for more frequent use (category 2).[7] The original Tapsall definition included spectinomycin in category 1, and azithromycin in category 2; we updated this based on currently recommended therapies in the UK (table 1). Although cefixime has not been recommended for treatment of NG since 2011, we retained it in category 1 as it is an ESC.

Descriptive and statistical analyses

Descriptive analyses were used to examine trends in AMR NG for 2004-2015. Analysis of MDR-NG was restricted to 2011 onwards, to reflect prevalence and risk factors relevant to current treatment

guidelines. Modal ceftriaxone and azithromycin MICs were compared between MDR and non-MDR isolates to assess whether there was a drift towards resistance to first-line therapies in MDR-NG among those not yet resistant to these antimicrobials. As resistance to ceftriaxone or azithromycin is included in the definition of MDR-NG, these sub-analyses excluded all isolates which already had decreased susceptibility to ceftriaxone (for comparison of ceftriaxone MICs) or resistance to azithromycin (for comparison of azithromycin MICs). For the risk factor analysis, logistic regression was used to generate Odds Ratios (ORs) initially adjusted for year, age (as a quadratic term, to account for the non-linear relationship) and gender/sexual orientation (groupings: MSM, heterosexual men, all women). Associations with the following variables were examined: ethnicity, residential neighbourhood-level Index of Multiple Deprivation (IMD), number of recent sexual partners (past 3 months), recent sex abroad (past 3 months), previous gonorrhoea infection, symptomatic infection, clinician-coded site of infection, concurrent STIs, and HIV status. Multivariable logistic regression analyses were used to determine independent associations, using a forwards model-building approach, with variables retained in the final model based on a p-value ≤0.10. Exploratory analysis showed evidence of within-clinic (but not within-patient) clustering of MDR-NG, i.e. there was greater similarity in MDR-NG between isolates collected within the same clinic than at different clinics. To avoid this resulting in underestimated standard errors, clustering was accounted for in the risk factor analysis using Generalised Estimating Equations under the assumption of an exchangeable correlation matrix.[29]

Results

Sample characteristics

Between 2004 and 2015, 16,242 isolates from 15,781 patients underwent susceptibility testing, with 47.4% of isolates from MSM, 28.2% from heterosexual men, and 20.6% from women (supplementary table 1).

Patterns of antimicrobial resistance in GRASP 2004-2015

Resistance to azithromycin was generally low until 2012 (<5%), then increased to 9.8% in 2015 (figure 1). Decreased susceptibility to ceftriaxone was rare (0.1%), whereas levels of decreased susceptibility to cefixime increased rapidly between 2008 and 2010 (from 3% to 17%), then decreased to 1.1% in 2015 (figure 1). Resistance to penicillin and ciprofloxacin was widespread (>10%) throughout the study period.

Between 2004 and 2015, the proportion of NG isolates fully susceptible to all classes of antimicrobials fell from 80% to 46%, with the proportion resistant to multiple (two or more) classes increasing from 7.3% to 17.5% (figure 2). Resistance to three classes increased between 2008 and 2010 then declined, mirroring trends in cefixime resistance (figure 1, figure 2). A small number of isolates were resistant to all four classes tested (n=49; 0.3% overall).

Despite high levels of susceptibility to prevailing first-line therapies immediately following revisions to treatment guidelines in 2005 (from ciprofloxacin to cefixime)[30] and 2011 (to ceftriaxone and azithromycin dual therapy)[11], the proportion of isolates fully susceptible to first-line therapies declined in the following years, from 99.9% in 2005 to 82.9% in 2010, and from 99.5% in 2011 to 90.2% in 2015 (figure 3).

Prevalence and profile of MDR-NG

Between 2011 and 2015, 3.5% (n=266) of isolates were MDR-NG, with small increases in prevalence between 2011 and 2013 (from 3.7% to 4.5%) followed by a subsequent decrease to 2.2% in 2015

(chi-squared test p=0.004). The profile of resistance to category 1 antimicrobials among MDR-NG isolates changed over time: 100% were resistant to cefixime in 2011, reducing to 36.9% in 2015, with increases in the proportion resistant to azithromycin over this time frame from 4.2% to 84.3% (p<0.01) (figure 4). All MDR-NG isolates were resistant to both ciprofloxacin and penicillin, but not spectinomycin, in category 2. Over time, the proportion of MDR-NG resistant to four antimicrobials (cefixime, azithromycin, penicillin, and ciprofloxacin) increased from 4.2% in 2011 to 21.1% in 2015. One isolate had resistance/decreased susceptibility to five antimicrobials: ceftriaxone (MIC 0.125 mg/L), cefixime (MIC 0.25 mg/L), azithromycin (MIC 1.0 mg/L), ciprofloxacin (MIC ≥16 mg/L), and penicillin (MIC 1 mg/L).

In a sub-analysis excluding all isolates with resistance to azithromycin, the MICs of azithromycin for MDR isolates were elevated (modal MIC 0.5 mg/L) compared with those for non-MDR isolates (modal MIC 0.125 mg/L) (figure 5a). Similarly, after excluding isolates with ceftriaxone decreased susceptibility, modal ceftriaxone MICs for MDR isolates were higher (0.03 mg/L) than those for non-MDR isolates (≤0.002 mg/L) (figure 5b). The elevated ceftriaxone MICs among MDR isolates were observed in a further sub-analysis stratified by year (grouped as 2011-13 (n=176 MDR isolates) and 2014-15 (n=87 MDR isolates)); ceftriaxone modal MIC of 0.03 mg/L among MDR isolates versus 0.004 mg/L among non-MDR isolates in both time periods.

Risk factors for MDR-NG

MDR-NG was more common among heterosexual men than MSM or women (4.6% versus 3.3% and 2.5%, respectively); these differences remained significant after adjustment for other factors (Table 2). Multivariable analysis also found MDR-NG to be more common among isolates from older patients (Adjusted Odds Ratio for those aged ≥45 years: 1.89 [95% Confidence Interval: 1.25-2.86], compared with those aged ≤24 years) and those reporting recent sex abroad (AOR 1.38 [95% CI: 1.02-1.87]), and less common among isolates from patients who were of black Caribbean ethnicity

(AOR 0.26 [95% CI 0.47-0.87]), were HIV positive (AOR 0.64 [0.47-0.87]), or had a concurrent STI (AOR 0.77 [0.61-0.97]). Neighbourhood deprivation (IMD) data were not available in GRASP for 2011 therefore IMD was not included in the multivariable model, however a separate multivariable model run on 2012-2015 data found no association between MDR-NG and IMD after adjustment for other factors.

Discussion

Our data demonstrate increases in antimicrobial resistance of *N. gonorrhoeae* over the past decade in England and Wales, with the proportion of NG isolates fully susceptible to all classes of antimicrobials falling from 80% in 2004 to 46% in 2015, and the proportion resistant to two or more classes increasing from 7.3% to 17.5% over this period. Despite high levels of susceptibility to recommended treatments immediately following changes to treatment guidelines in 2005 and 2011, the proportion fully susceptible to these decreased in a matter of years. In 2011-2015, 3.5% of isolates were classed as MDR-NG, meaning they were resistant to either azithromycin or an extended spectrum cephalosporin, as well as to penicillin and ciprofloxacin. In 2011, MDR-NG isolates were predominantly resistant to cefixime, however, by 2015 the majority were resistant to azithromycin, with around one in five resistant to both. We also found evidence of drift towards azithromycin resistance among MDR-NG isolates that were not resistant to azithromycin, and similar drift towards ceftriaxone resistance. The fact that these elevated ceftriaxone MICs in the MDR isolates were observed in 2014-15 as well as 2011-13 suggests this finding is unlikely to be solely due to the cefixime-resistant ST1407 strains which also have elevated ceftriaxone MICs and were prevalent during 2011-2013.[25] Although the risk factor analysis found MDR-NG was more common among isolates from some groups, including heterosexual men and older patients, absolute differences in prevalence were small, providing little basis for targeted treatment strategies. These

findings highlight the importance of continued surveillance and prevention efforts, given the severely limited treatment options available to those with MDR-NG.

Advantages to using the GRASP surveillance data include the large sample size, ability to look at trends over time, and to combine MIC data with demographic, clinical, and behavioural information. GRASP is reasonably representative of gonorrhoea cases across England, although MSM are somewhat overrepresented.[31] It is likely that risk characteristics of MSM and heterosexual sexual networks differ, but small numbers of MDR-NG prohibited stratification of risk factor analysis by gender/sexual orientation. Another limitation is the potential for missing data to bias results of the risk factor analysis; for example completion of behavioural data varied by clinic resulting in more missing data for MSM. Azithromycin MIC data from 2013-14 were adjusted to account for poor organism growth using DST media which underestimated MICs. This adjustment enabled comparisons of AMR over time, but is a crude correction and some misclassification is likely.

In line with low levels of resistance to ceftriaxone generally,[13] few MDR-NG cases had decreased susceptibility to ceftriaxone. However, MICs of ceftriaxone have been drifting towards resistance in recent years,[15] leaving no room for complacency about the long-term effectiveness of current dual therapy. Our analysis found that the ceftriaxone MICs for MDR-NG isolates were higher than for non-MDR isolates, after excluding those with decreased susceptibility to ceftriaxone, demonstrating how easily MDR-NG with resistance to ceftriaxone could emerge and lead to infections which would be difficult to treat. Although all isolates were susceptible to spectinomycin, this is not considered a viable first-line treatment as resistance has historically developed rapidly when it has been used, it is not effective in treating pharyngeal gonorrhoea, and it is currently unavailable in many countries.[5] It should be borne in mind that 96.5% of isolates were not MDR, which is promising in light of efforts to develop point-of-care AMR testing to guide treatment choices.[32] If successful, such tests could

identify infections for which previously-used therapies would be effective, reducing ceftriaxone use and the selection pressure for resistance.

Despite commentaries on the emerging threat of MDR-NG,[5,6,33] the international literature on prevalence and risk factors is scant, perhaps due to lack of consensus on an MDR definition, preventing comparison of prevalence and risk factors.[22,23,26] In general the risk factors for MDR-NG identified here are consistent with those associated with resistance to some individual antimicrobials in previous studies, including being a heterosexual man (ciprofloxacin, azithromycin, cefixime),[34] the inverse relationships with black Caribbean ethnicity (ciprofloxacin, cefixime),[20,21] and concurrent STI infection (ciprofloxacin, cefixime).[20,21] However, previous analyses of GRASP data have found a positive association between HIV and NG that was resistant to cefixime, ciprofloxacin, penicillin,[20,21] in contrast with the inverse relationship with MDR-NG found here. Continued monitoring of the epidemiology of MDR-NG alongside resistance to individual antibiotics is therefore needed.

These data emphasise the loss of susceptibility of NG to sequential antimicrobial classes used for treatment, and provide the first prevalence estimates of MDR-NG in England and Wales. Although prevalence of MDR-NG was relatively low, many of these isolates were resistant to azithromycin and had ceftriaxone MICs that were higher than for non-MDR isolates. In the context of limited new treatment options, the emergence of ceftriaxone resistance in MDR-NG could herald the prospect of untreatable NG. MDR-NG was generally homogenously distributed by demographic and behavioural characteristics, with no groups identified as being at especially high risk, highlighting the need for continued culture and susceptibility testing and test of cure among all patients with gonorrhoea. Our findings underline the essential role of timely surveillance to identify and respond to clinically significant changes in AMR NG in the coming years.

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Transparency declarations

HF is a member of the Scientific Advisory Board for Discuva Ltd. SC, HM, HB, KT, MF, MC, OC, GH have all declared no conflict of interest.

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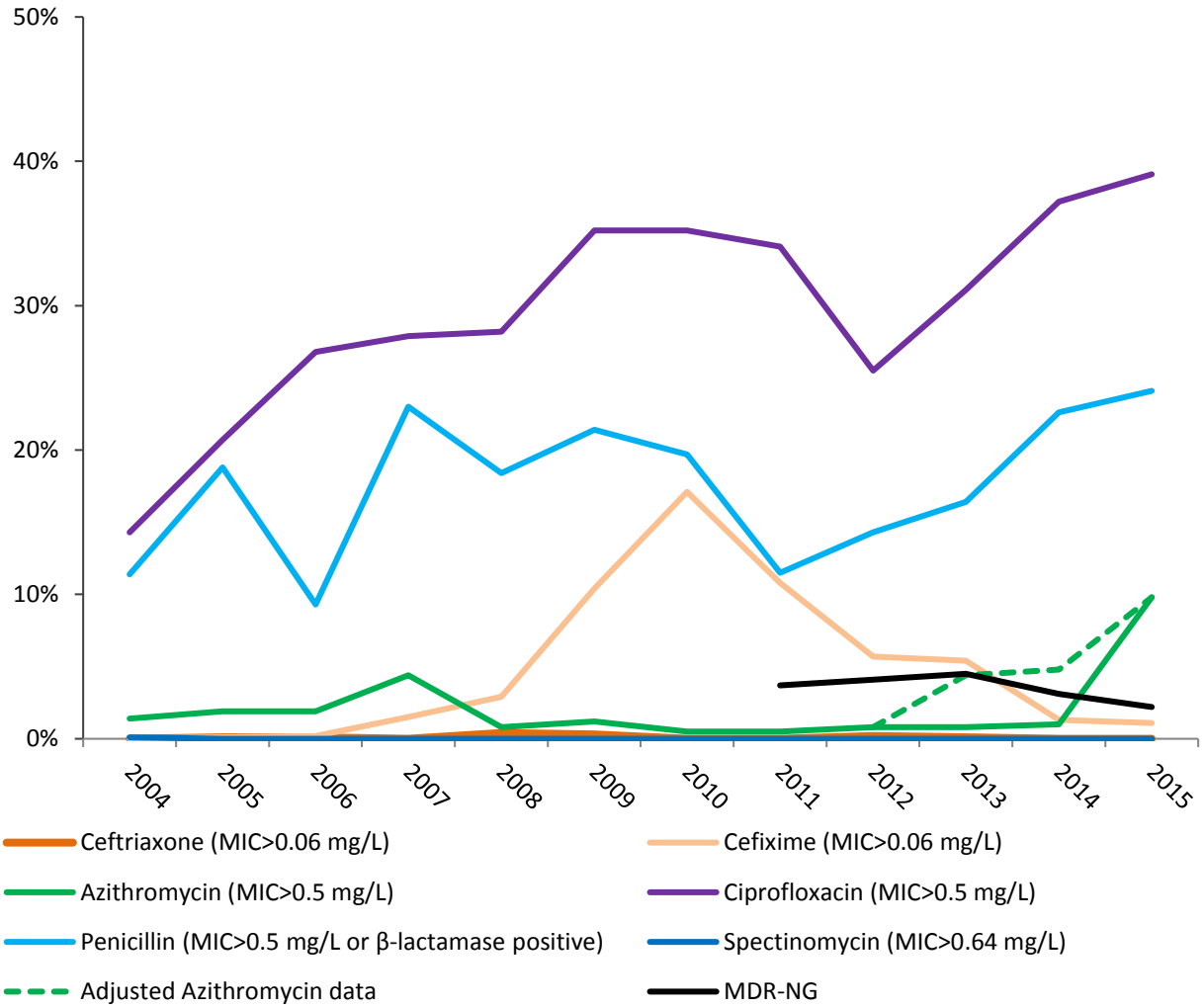
Tables and figures

Table 1: Category of antibiotic and MIC thresholds used to define resistance or decreased susceptibility in GRASP

Category 1 antibiotics:	**MIC breakpoint for resistance^{R} or decreased susceptibility^{DS}**
Ceftriaxone^{DS}	>0.06 mg/L
Cefixime^{DS}	>0.06 mg/L
Azithromycin^{R}	>0.5 mg/L
Category 2 antibiotics:	
Ciprofloxacin^{R}	>0.5 mg/L
Penicillin^{R}	>0.5 mg/L or β-lactamase positive
Spectinomycin^{R}	>64 mg/L

GRASP standard breakpoints.[13] Decreased susceptibility, rather than 'resistance' is used in GRASP for ESCs as treatment failures have been observed across a range of MIC values in some patients but not others.

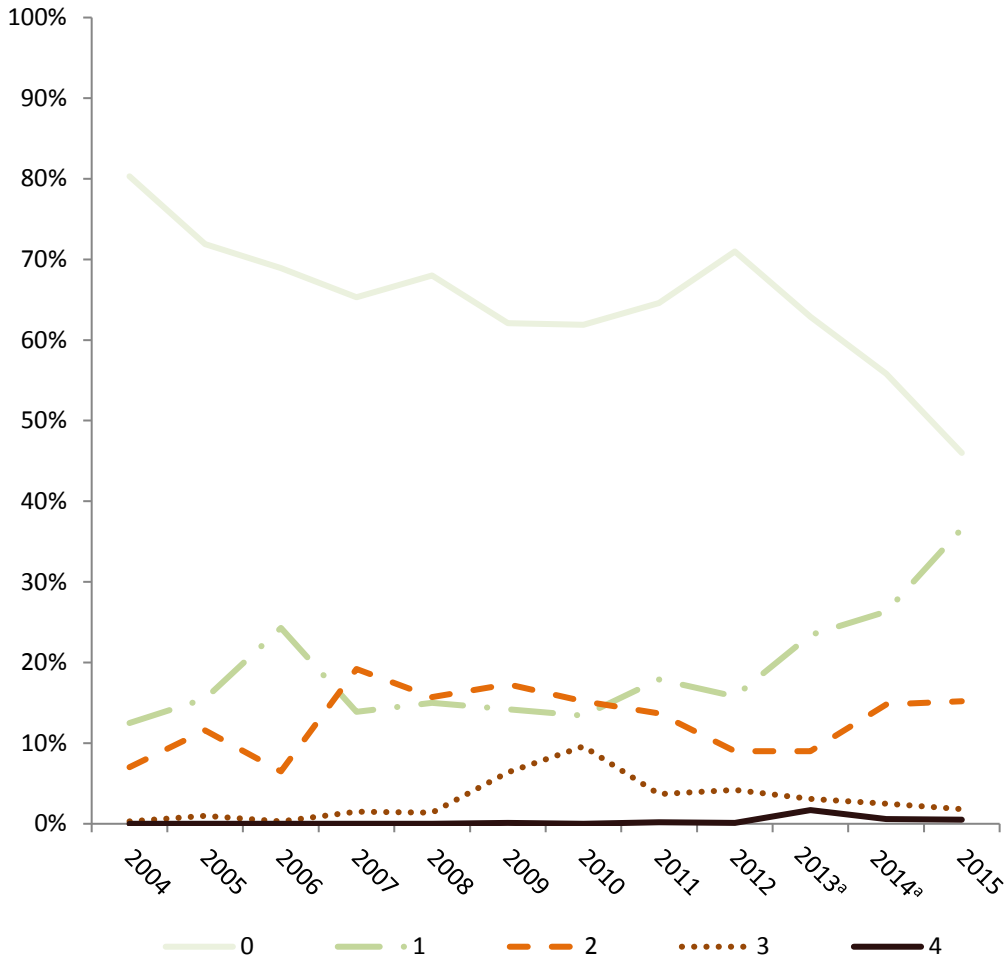
Figure 1: Percentage of *Neisseria gonorrhoeae* isolates with antimicrobial resistance/decreased susceptibility, by year, England and Wales, GRASP 2004–2015 (n=16242)



Note: Azithromycin data for 2013-14 adjusted to account for poor growth on the DST medium leading to underestimation of azithromycin MIC in those years (See Methods for further details).

This figure appears in colour in the online version of JAC and in black and white in the print version of JAC.

Figure 2: Percentage of *Neisseria gonorrhoeae* isolates resistant to 0, 1, 2, 3, or 4 classes of antimicrobials, by year, England and Wales, GRASP 2004–2015 (n=16242)



Classes of antimicrobials: a) extended-spectrum cephalosporin (cefixime & ceftriaxone) b) macrolide (azithromycin) c) fluoroquinolone (ciprofloxacin) d) penicillin e) aminoglycoside (spectinomycin).
[a] 2013-14 estimates calculated using adjusted azithromycin data.

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Figure 3: Percentage of isolates fully susceptible to recommended first-line therapy [a] England and Wales, GRASP 2004–2015 (n=16242)

[a] Recommended first-line therapies: ciprofloxacin (2004), cefixime (2005-2010), ceftriaxone and azithromycin (2011-2015).
[b] Azithromycin data for 2013-14 adjusted to account for poor growth on the DST medium leading to underestimation of azithromycin MIC in those years (see Methods for further details).

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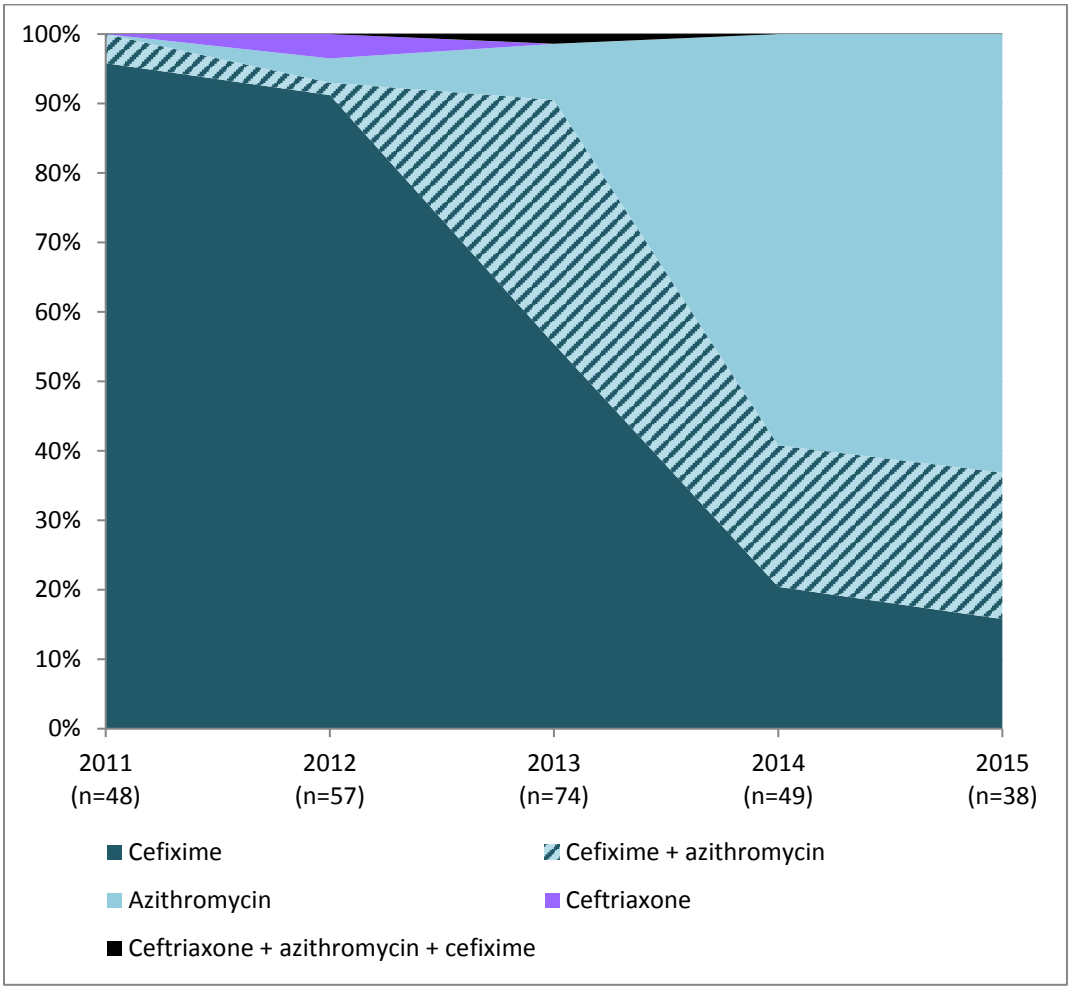
Figure 4: Resistance/decreased susceptibility to category 1[a] antimicrobials among MDR-NG isolates over time, England and Wales, GRASP 2011–2015 (n=266)



[a] Category 1 antimicrobials: Ceftriaxone, Cefixime, Azithromycin; category 2 antimicrobials: ciprofloxacin, penicillin, spectinomycin. See methods for further details.
All MDR-NG isolates were resistant to ciprofloxacin and penicillin in category 2.

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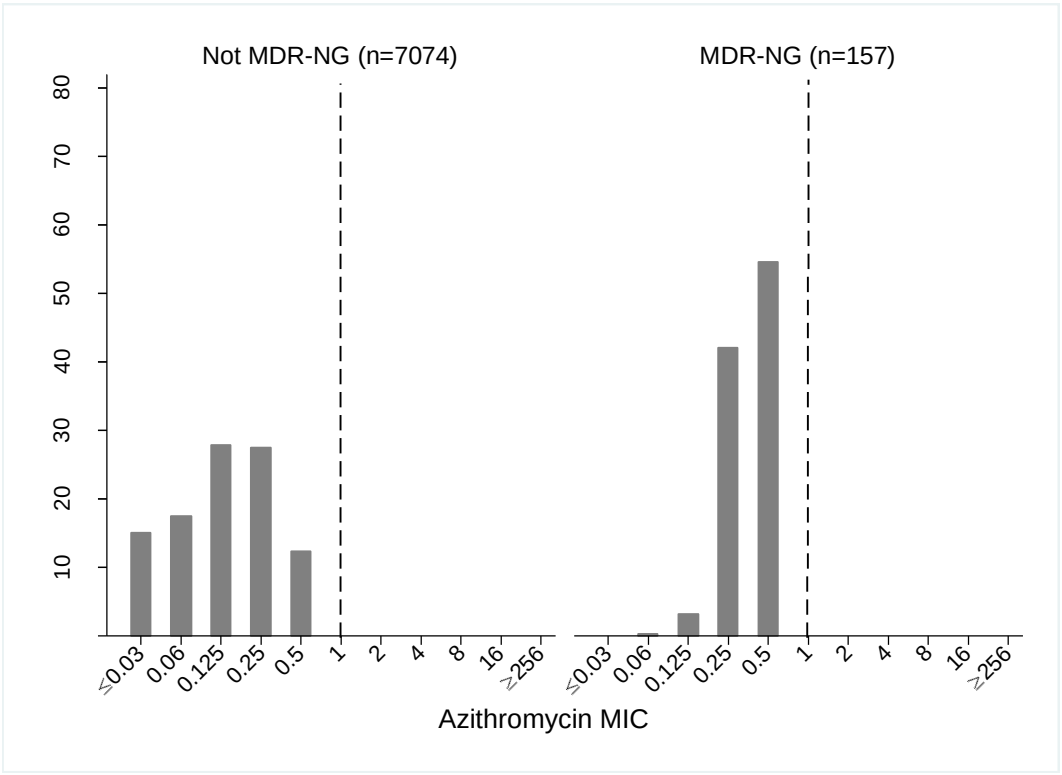
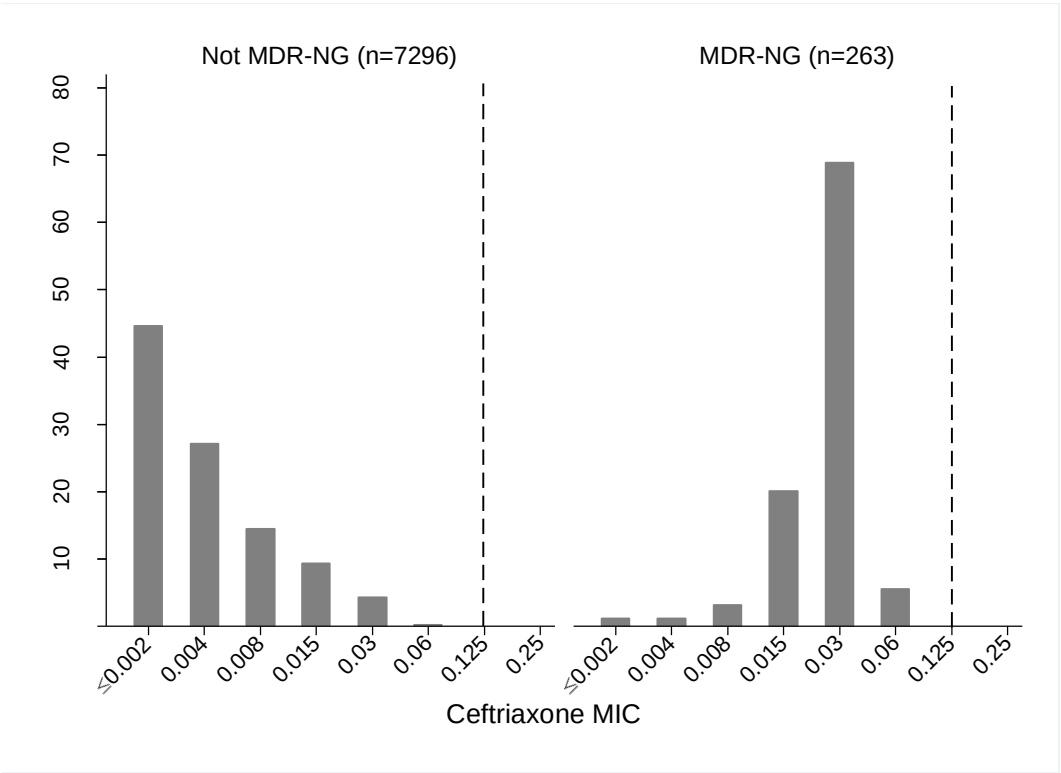
Figure 5: Distribution of MICs for a) azithromycin and b) ceftriaxone among MDR-NG and non-MDR-NG isolates not meeting the resistance/decreased susceptibility thresholds for ceftriaxone/azithromycin respectively. England and Wales, 2011–2015

a) Azithromycin



b) Ceftriaxone



Threshold for resistance / decreased susceptibility

Table 2: Risk factors for MDR-NG among *Neisseria gonorrhoeae* isolates, England and Wales, GRASP 2011-2015

	Total N	n	%	Adjusted for age, year, and gender/sexual orientation AOR1	(95% CI)	Multivariable model (n=4027) AOR2	(95% CI)
All	*7562*	266	3.5%				
Year					p=0.04		p<0.001
2011	*1288*	48	3.7%	*ref*		*ref*	
2012	*1375*	57	4.1%	1.16	(0.76 - 1.66)	1.16	(0.72 - 1.88)
2013	*1636*	74	4.5%	1.26	(0.77 - 2.08)	3.86	(2.51 - 5.94)
2014	*1564*	49	3.1%	0.84	(0.62 - 1.14)	3.61	(2.43 - 5.37)
2015	*1699*	38	2.2%	0.64	(0.36 - 1.11)	0.60	(0.35 - 1.03)
Gender/sexual orientation					p=0.02		p=0.05
Heterosexual men	*1571*	72	4.6%	*ref*		*ref*	
MSM	*4683*	156	3.3%	0.67	(0.45 - 1.01)	0.76	(0.57 - 0.99)
Women	*1123*	28	2.5%	0.62	(0.42 - 0.90)	0.66	(0.44 - 0.99)
Age group					p=0.13		p=0.01
≤24	*2337*	64	2.7%	*ref*		*ref*	
25-44	*4507*	173	3.8%	1.42	(0.98 - 2.03)	1.35	(1.01 - 1.81)
≥45	*714*	29	4.1%	1.49	(0.96 - 2.29)	1.89	(1.25 - 2.86)
Ethnicity[a]					p=0.01		p=0.004
White	*5187*	197	3.8%	*ref*		*ref*	
Black Caribbean	*580*	12	2.1%	0.42	(0.21 - 0.81)	0.26	(0.47 - 0.87)
Black African	*281*	7	2.5%	0.55	(0.25 - 1.19)	0.87	(0.43 - 1.76)
Black Other	*180*	5	2.8%	0.63	(0.24 - 1.66)	0.27	(0.04 - 1.78)
Asian (including Chinese)	*360*	15	4.2%	1.04	(0.53 - 2.06)	1.58	(0.79 - 3.14)
Mixed Ethnic Group	*500*	13	2.6%	0.63	(0.30 - 1.32)	0.75	(0.35 - 1.62)
Other Ethnic Group	*181*	5	2.8%	0.64	(0.25 - 1.63)	1.00	(1.02 - 1.87)
Patient's area-level deprivation (IMD) quintile					p=0.09		
1 or 2 (least deprived)	*698*	34	4.9%	*ref*			
3	*835*	26	3.1%	0.63	(0.42 - 0.93)		
4	*1974*	70	3.5%	0.71	(0.49 - 1.04)		
5 (most deprived)	*2341*	73	3.1%	0.59	(0.37 - 0.96)		
Number of sexual partners (past 3 months)					p>0.99		
0-1	*1974*	71	3.6%	*ref*			
2-5	*3021*	112	3.7%	0.99	(0.72 - 1.38)		
≥6	*845*	30	3.6%	1.00	(0.60 - 1.69)		
Sex while abroad (past 3 months)					p=0.03		p=0.04
No	*5170*	175	3.4%	*ref*		*ref*	
Yes	*670*	38	5.7%	1.55	(1.04 - 2.30)	1.38	(1.02 - 1.87)
Previous gonorrhoea infection					p=0.69		
No	*4440*	159	3.6%	*ref*			
Yes	*2325*	77	3.3%	0.95	(0.72 - 1.25)		
Symptoms of gonorrhoea[b]					p=0.68		
No	*1810*	59	3.3%	*ref*			
Yes	*4241*	162	3.8%	1.07	(0.78 - 1.46)		
Clinicial-coded site of infection[c]							
Genital					p=0.87		
No	*1848*	69	3.7%	*ref*			
Yes	*4644*	167	3.6%	0.88	(0.62 - 1.25)		
Rectal					p=0.49		
No	*4027*	155	3.8%	*ref*			
Yes	*2480*	81	3.3%	0.90	(0.68 - 1.20)		
Throat					p=0.06		
No	*4517*	154	3.4%	*ref*			
Yes	*1990*	82	4.1%	1.43	(0.98 - 2.09)		

Other					p=0.67		
No	*6338*	229	3.6%	*ref*			
Yes	*169*	7	4.1%	1.31	(0.73 - 2.34)		
Multiple sites of infection					*p=0.76*		
No	*4353*	*160*	*3.7%*	*ref*			
Yes	*2154*	*76*	*3.5%*	*1.06*	*(0.72 - 1.57)*		
Concurrent STI (excl HIV)					p=0.01		p=0.03
No	*5639*	219	3.9%	*ref*		*ref*	
Yes	*1923*	47	2.4%	0.66	(0.48 - 0.90)	0.77	(0.61 - 0.97)
HIV status					p=0.002		p=0.005
Negative	*3592*	141	3.9%	*ref*		*ref*	
Positive	*1264*	31	2.5%	0.52	(0.34 - 0.78)	0.64	(0.47 - 0.87)

AOR1=Adjusted Odds Ratio, adjusted for year, age (quadratic), gender/sexual orientation. AOR2=Multivariable model, adjusted for other variables shown. Age adjusted for as a quadratic variable, but AORs and p-value presented for age group for ease of interpretation.

[a] IMD was not available for 2011 in GRASP; [b] Discharge/painful urination (dysuria); [c] Sites not mutually-exclusive. Where patient had multiple infection sites, this site may not correspond to the site that underwent susceptibility testing as only one isolate per patient was included in GRASP. Isolates were prioritised as follows: 1) Male Rectal 2) Male Urethral 3) Female Cervical 4) Any other site.