Population-level antimicrobial consumption is associated with decreased antimicrobial susceptibility in *Neisseria gonorrhoeae* in 24 European countries: an ecological analysis

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Summary of article: Differences in population level consumption of particular antimicrobials may contribute to the variations in the level of antimicrobial resistance in *N. gonorrhoeae*.

Objectives: There are substantial variations in *Neisseria gonorrhoeae* susceptibility to antimicrobials between different populations, and the reasons for this are largely unexplored. We aimed to assess if the population level consumption of antimicrobials is a contributory factor.

Methods: Using antimicrobial susceptibility data from 24 countries in the European Gonococcal Antimicrobial Surveillance Programme and antimicrobial consumption data from the IQVIA MIDAS database, we built mixed effects linear/logistic regression models with country-level cephalosporin, fluoroquinolone and macrolide consumption (standard doses/1000 population/year) as the explanatory variables (from 2009 to 2015) and 1-year lagged ceftriaxone, cefixime, azithromycin and
ciprofloxacin geometric mean minimum inhibitory concentrations (MIC) as the outcome variables (2010 to 2016).

**Results:** Positive correlations were found between the consumption of cephalosporins and geometric mean MIC of ceftriaxone and cefixime (both P's <0.05). Fluoroquinolone consumption was positively associated with the prevalence of resistance to ciprofloxacin (P<0.05).

**Conclusions:** Differences in population level consumption of particular antimicrobials may contribute to the variations in the level of antimicrobial resistance in *N. gonorrhoeae* in different settings. Further interventions to reduce misuse and overuse of antimicrobials in high-consumption populations and core-groups are required.
Introduction

*Neisseria gonorrhoeae* has developed resistance to all antimicrobials used to treat gonorrhoea.[1] The first gonococcal isolates with combined resistance to ceftriaxone and high-level resistance to azithromycin were recently reported in the United Kingdom (UK) and Australia.[2, 3] Various *in vitro* and individual-level studies have established a link between antimicrobial exposure and the development of antimicrobial resistance (AMR) in Ng.[1, 4-10] As a result, resistance to an antimicrobial has typically emerged in Ng a few years or decades after this agent was introduced as a treatment for gonorrhoea.[1, 4, 11] This has led to the hypothesis that the antimicrobials used to treat gonorrhoea (and other sexually transmitted infections - STIs) are the predominant drivers of AMR development in Ng.[12]

A complementary and underexplored potential driver of AMR in *N. gonorrhoeae* is the consumption of antimicrobials in the general population.[13] For a wide range of bacterial species, strong population-level correlations have been found between the level of antimicrobial consumption and resistance to that antimicrobial or similar antimicrobials.[14-21] Part of this effect can be explained by bystander
selection, the inadvertent selection of AMR in microbes not targeted by antimicrobial therapy.[22] One analysis of AMR and prescription data from the United States of America (USA) in 2010/2011, estimated that this selection effect was moderate to considerable for *N. gonorrhoeae* - 97.5% of all gonococcal fluoroquinolone and 14.6% of all cephalosporin exposures were of a bystander nature.[22] Of note, the more prevalent a species was, the more likely it was to be exposed to antibiotics used for any indication.

As a result, because most humans are colonized by at least one of 20 *Neisseria* non-gonococcal species at any point in time, these species may be particularly susceptible to the bystander effects of high antimicrobial exposure.[23] This is illustrated in Figure 1, where high antimicrobial exposure is followed by an increased prevalence/abundance of non-susceptible *Neisseria* non-gonococcal species and an increased probability of AMR developing directly in circulating *N. gonorrhoeae*. In addition, the pathway to AMR in *N. gonorrhoeae* may be indirect: gonococci circulating in the high antimicrobial consumption population may acquire the resistance-conferring-DNA from the commensal *Neisseria* spp. via horizontal gene transfer. Various studies have provided suggestive evidence that *N.
*N. gonorrhoeae* has acquired penicillin, cephalosporin and macrolide resistance via this type of mechanism.[24, 25]

Previous studies have reached different conclusions regarding the association between general antimicrobial consumption in the emergence or persistence of AMR in *N. gonorrhoeae*. In an ecological analysis of county level data from the USA for the years 2005-2013, Kirkcaldy et al. did not find an association between antimicrobial consumption and antimicrobial susceptibility or AMR in *N. gonorrhoeae*.[13] However, a study of all countries contributing data to the World Health Organization’s Global Gonococcal Antimicrobial Surveillance Programme, found positive associations between the antimicrobial consumption and resistance to cephalosporins, macrolides and fluoroquinolones.[26] Both these studies had limitations: the Kirkcaldy study[13] took place in a single country with low heterogeneity in AMR and antimicrobial consumption whereas the global study[26] relied on limited AMR estimates generated by different non-standardized surveillance methods examining significantly different sample sizes in different countries and the fact that the analysis was limited to simple linear regression at two time points - 2009 and 2013.
In the present study, we assessed the association between antimicrobial consumption and antimicrobial susceptibility and AMR in all European Union (EU) and European Economic Area (EEA) countries providing isolates to the European Gonococcal Antimicrobial Surveillance Programme (Euro-GASP).[27] This approach offers considerable advantages over the previous studies as there are large differences in both antimicrobial consumption and *N. gonorrhoeae* AMR between the Euro-GASP countries.[27] Furthermore, Euro-GASP uses a standardized, quality-controlled methodology to evaluate AMR and has data for six contiguous years, which allowed us to use mixed effects linear/logistic regression to assess the association between AMR and consumption.

For each antimicrobial, we assessed the correlation between antimicrobial consumption and three measures of susceptibility by country and year: 1) Geometric mean MIC of all isolates, (2) the percentage of isolates with resistance, and (3) geometric mean MIC of all isolates excluding those defined as resistant for that antimicrobial (i.e. including isolates with susceptibility and intermediate susceptibility - henceforth referred to as the geometric mean MIC of susceptible isolates).
Our rationale for this third analysis was to assess if consumption was correlated with only an increase in the percent of isolates that were resistant (such as the "Bimodal" population in Figure 2), or with a right shift of the entire population MIC distribution (such as that of Greece in Figure 2). We reasoned that in the case of the latter but not the former there should be a correlation between antimicrobial consumption and geometric mean MIC of susceptible isolates. If antimicrobial consumption was found to be associated with the percent of isolates that were resistant but not with the geometric mean of susceptible isolates, then this would suggest that antimicrobial consumption has little influence on the MICs of the general population of circulating gonococci but only an effect on a resistant subpopulation. This finding would support AMR prevention approaches to identify the risk factors that were responsible for translating antimicrobial exposure into AMR in this subpopulation. Conversely, if consumption was also associated with geometric mean of susceptible isolates then this would favor AMR prevention approaches that place greater emphasis on overall reduction in antimicrobial consumption.
Material and Methods

Our data came from two sources.

Antimicrobial susceptibility and resistance

Euro-GASP includes a sentinel AMR surveillance programme that tests a representative number of isolates from EU/EEA Member States every year for a range of antimicrobials through a hybrid centralised/decentralised system, see Table S1 for the 24 included Euro-GASP countries. We used the Euro-GASP MIC data for ceftriaxone, cefixime, azithromycin, and ciprofloxacin from 2010 to 2016. The full Euro-GASP methodology, including recommended sampling strategy, laboratory techniques, external quality assurance and internal quality control mechanisms, has been published elsewhere.[27][28] The following EUCAST MIC resistance breakpoints (www.eucast.org) were used to define AMR in Euro-GASP between 2010 and 2016: Azithromycin resistance: (R)>0.5 mg/L, cefixime: R>0.12 mg/L, ceftriaxone: R>0.12 mg/L, and ciprofloxacin: R>0.06 mg/L (www.eucast.org).[29, 30]

Antimicrobial consumption
Data from MIDAS Quantum data base of the marketing research company IQVIA (IQVIA, Danbury, CT, USA) were used as a measure of national antimicrobial drug consumption. IQVIA uses national sample surveys that are performed by pharmaceutical sales distribution channels to estimate antimicrobial consumption from the volume of antibiotics sold in retail and hospital pharmacies.[19] The sales estimates from this sample are projected with use of an algorithm developed by IQVIA to approximate total volumes for sales and consumption. Antimicrobial consumption estimates are reported as the number of standard doses (a dose is classified as a pill, capsule, or ampoule) per 1000 population per year.[19] Data on consumption of cephalosporins, macrolides and fluoroquinolones (Table S2) were available for 34 WHO European Region countries. We used data for the 24 countries that were also represented in the Euro-GASP dataset. We compared susceptibility with consumption from the previous year and as a result we used consumption data from the years 2009 to 2015.

Data analysis
For each antimicrobial, we calculated the following susceptibility variables by country and year: 1) Geometric mean MIC of all isolates, (2) the percentage of isolates with resistance, and (3) geometric mean MIC of all isolates excluding those defined as resistant for that antimicrobial (i.e. including isolates with susceptibility and intermediate susceptibility - henceforth referred to as the geometric mean MIC of susceptible isolates). For each antimicrobial consumption and susceptibility variable, we then calculated the 2009-2015/2010-2016 geometric mean by individual country. We described how these varied between countries. In addition, we used these 2009-2015/2010-2016 geometric means to categorize countries as being in the top or bottom quartile for antimicrobial consumption and geometric-mean-MIC-of-all-isolates.

**Correlation between antimicrobial susceptibility and consumption**

In separate analyses for each antimicrobial, mixed effects linear regression was used to assess the correlation between antimicrobial consumption and 1) the geometric mean of all isolates and 2) the geometric mean of susceptible isolates. Because AMR is reported as a proportion, mixed effects logistic regression was
used to assess the correlation between consumption and AMR. The following mixed effects linear/logistic regression model was used:

\[(\text{MIC/resistance in year Y and country C}) \sim (\text{antimicrobial consumption in year Y-1 and country C}) + (\text{random intercept for country C}) + \text{intercept} + \text{error}.\]

**Association between AMR and geometric mean MIC of susceptible isolates**

Mixed effects linear regression was used to assess the associations between the prevalence of resistance in a country and the homologous geometric mean MIC of susceptible isolates for ceftriaxone, cefixime, azithromycin, and ciprofloxacin in the same year. This analysis was performed to further assess if the prevalence of AMR was associated with a shift in the population MIC distribution - as opposed to a bimodal distribution.

All mixed effects analyses were weighted by the number of isolates by country and year used in the analysis. The MIC values were log transformed to create more normal distributions for these regression analyses. When MIC values were reported as less/more than the lowest/highest concentration tested, these were replaced with the lowest or highest concentrations tested.
Spearman’s correlation was also used to summarize the association between countries’ consumption of cephalosporins, macrolides and fluoroquinolones and *N. gonorrhoeae* geometric mean MIC of cefixime, ceftriaxone, azithromycin and ciprofloxacin. In these analyses each country was represented by a single data point which was the geometric mean value from the years 2009 to 2015 for antimicrobial consumption and from 2010 to 2016 for MIC values.

We assumed that all missing data was missing completely at random and therefore all missing data points were dropped from the analyses. All statistical analyses were performed in R V.3.3.2 and Stata 13.0.

**Results**

The number of countries with antimicrobial consumption data available increased from 19 countries (2009 to 2012), to 20 in 2013, 22 in 2014, and 24 in 2015. The completeness of data for AMR improved by year for all antimicrobials. The number of countries with MIC data increased between 2010 and 2016; from 18 to 24 for azithromycin and ceftriaxone, 18 to 23 for cefixime, and 9 to 20 countries for ciprofloxacin.
Antimicrobial susceptibility

The 2010-2016 geometric mean MIC of ceftriaxone varied 5-fold: from 0.004 mg/L in the Czech Republic to 0.02 mg/L in Luxembourg (Table 1, Figure 3). The 2010-2016 geometric mean MIC of susceptible isolates varied 8-fold and the prevalence of AMR varied between 0.0% and 5.0% (Table 1, Figure S2).

For cefixime, there was a 4-fold variation in the 2010-2016 geometric mean MIC between countries, ranging from 0.01 mg/L in the UK to 0.04 mg/L in Greece (Table 1, Figures 3 & S1). The 2010-2016 geometric mean MIC of susceptible isolates varied 3-fold whereas the prevalence of AMR ranged from 0.0% to 12.7% (Table 1, Figure S1).

The azithromycin 2010-2016 geometric mean MICs varied from 0.13 mg/L in the Netherlands to 0.63 mg/L in Norway (5-fold variation; Table 1, Figures 3 & S1). The 2010-2016 geometric mean MIC of susceptible isolates varied 3-fold and the prevalence of AMR varied between 0.0% and 13.5% (Table 1, Figure S2).

For ciprofloxacin, there was considerable (226-fold) variation in the 2010-2016 geometric mean MIC between countries, with a maximum of 1.64 mg/L in Greece.
and minimum of 0.007 mg/L in Estonia (Table 1, Figures 3 & S1). The 2010-2016 geometric mean MIC of susceptible isolates varied 35-fold (range 0.003 to 0.11 mg/L) whilst the prevalence of AMR differed 3-fold (range from 20.7% to 70.0%; Table 1, Figure S1).

**Association between AMR and geometric mean of susceptible isolates**

Mixed effects linear regression revealed positive correlations between the prevalence of resistance in a country and the homologous geometric mean MIC of susceptible isolates for ceftriaxone (coef. 51, P<0.001), cefixime (coef. 539; P<0.001), and azithromycin (coef. 19; P<0.001), but not ciprofloxacin (coef. 63; P=0.797).

**Antimicrobial consumption**

There were large inter-country variations in the consumption of cephalosporins, macrolides and fluoroquinolones (Table 2, Figures 3 & S1). As has been detailed in other analyses, the rates of consumption of these three classes of antimicrobials tended to be lower in northern than southern European countries.[20]
The geometric mean consumption of cephalosporins in 2010-2016 varied from 76 doses per 1000 population per year in the Netherlands to 3376 in Greece (a 44-fold difference; median 925, IQR 219-1219). From 2010 to 2016, consumption increased by ≥25% (from a low baseline) in Denmark and decreased by ≥25% in the UK, Sweden, Slovenia and France (Table 2, Figures 3 & S1).

Macrolide consumption varied by a factor of 11 between countries; from 296 in Sweden to 3300 in Greece (median 1037, IQR 670-1343). Consumption increased by ≥25% in Latvia and decreased by ≥25% in Denmark, Greece, Portugal, France, Austria, Norway, Slovenia and Sweden (Table 2, Figure 3).

Consumption of fluoroquinolones varied by a factor of 6; from 266 in the UK to 1501 in Italy (median 649, IQR 383-1028). Consumption increased by ≥25% in Hungary and decreased by ≥25% in Portugal and France (Table 2, Figure 3).

**Correlation between antimicrobial susceptibility and consumption**

Mixed effects linear regression analyses, with a random intercept for country, revealed positive correlations between geometric mean MIC of the four antimicrobials and homologous antimicrobial class consumption level. However, this
correlation was only statistically significant for ceftriaxone (coefficient 0.003; Standard Error [SE] 0.0008; P=0.003) and cefixime (coefficient 0.004; SE 0.0017; P=0.017). This means that for every unit increase in cephalosporin consumption, the geometric mean MIC for ceftriaxone and cefixime increased by 0.003 and 0.004 mg/L on average, respectively (Table 3). Repeating the analyses using geometric mean MIC of susceptible isolates as the dependent variable, cephalosporin consumption remained significantly correlated with ceftriaxone and cefixime MIC (both P's < 0.05), but no significant correlations were found for macrolides and fluoroquinolones (Table 3). On re-running the models using mixed effects logistic regression and AMR as the independent variable, only fluoroquinolone consumption was significantly correlated with homologous (ciprofloxacin) resistance (P<0.01; Table 3). Spearman’s correlation between countries consumption of cephalosporins, macrolides and fluoroquinolones versus N. gonorrhoeae geometric mean MIC of cefixime, ceftriaxone, azithromycin and ciprofloxacin revealed significant associations for cephalosporins-cefixime (ρ=0.43; 95% CI 0.06-0.81; P=0.024), cephalosporins-ceftriaxone (ρ=0.41; 95% CI 0.01-0.82;
Discussion

We found large variations in the consumption of cephalosporins, macrolides and fluoroquinolones between 24 EU/EEA countries. Cephalosporin consumption was correlated with ceftriaxone and cefixime geometric mean MICs as well as the geometric mean MIC of susceptible isolates, but not with cephalosporin resistance (possibly related to the low number of resistant isolates). Fluoroquinolone consumption was associated with ciprofloxacin resistance. These findings are commensurate with those from other bacteria where similar associations have been found.[14-20] No significant associations were found for macrolides.

Another finding was that the proportion of resistance to ceftriaxone, cefixime and azithromycin was positively associated with shifts in the whole MIC distribution. We also found that the amount of cephalosporins consumed in a country was positively associated with geometric mean cefixime and ceftriaxone MIC of susceptible isolates. These findings suggest that the intensity of
consumption of antimicrobials in the general population, including an exposure of

*Neisseria gonorrhoeae* and other bacterial species to sub-inhibitory antimicrobial
concentrations, might play a role in increasing the cephalosporin MICs in *N.
gonorrhoeae* by right shifting the MIC distribution.

The magnitudes of the associations between antimicrobial consumption and
decreased susceptibility were generally small but in the same range as those
found in analogous studies [21]. There are a number of other possible
explanations for our findings. The prevalence of antimicrobials consumed by the
general population may be correlated with the antimicrobials used to treat *N.
gonorrhoeae*-associated STI syndromes and particularly for the cephalosporins and
macrolides, this may be an important driver of AMR in *N. gonorrhoeae*. It is likely
that a complex interplay between a large number of factors underpin the
emergence and differential spread of AMR in *N. gonorrhoeae*. [12] These factors
include: variations in national STI screening practices,[13, 31] differences in the
sexual network structure,[32-34] differences in the prevalence of *N. gonorrhoeae*
overall and particular *N. gonorrhoeae* strains,[8, 35, 36] variations in the
prevalence of sexual practices (such as oral and anal sex),[32, 37] differences in
rates of non-prescription antimicrobial consumption,[38] and travel/sexual tourism (i.e. importation of AMR *N. gonorrhoeae* strains).[39] We did not control for any of these in our study. A further study limitation is the fact that most *N. gonorrhoeae* infections occur in younger adults whereas our antimicrobial consumption data were not limited to this age band. Our analysis was ecological and is thus susceptible to the ecological inference fallacy. Finally, we had incomplete data and/or small isolates for a number of countries. We assumed that missing data was missing completely at random but if this was not the case then our analytical approach of dropping missing data may have biased our results.

We did not evaluate the accuracy of the antimicrobial consumption data. The consumption estimates from IQVIA for 2015 have, however, been found to correlate closely with those from other estimates such as those from the European Surveillance of Antimicrobial Consumption Network data (correlation of 93% in 2015).[19, 40] We used a one-year time lag between antimicrobial consumption and AMR as this has been previously shown to provide the best fit to the emergence of penicillin and macrolide resistance in *Streptococcus pneumoniae*.[18]
In sensitivity analyses, using no-lag period made little difference to the results (Table S3).

Our results should not be viewed in isolation. A number of studies have found strong associations at various levels between antimicrobial consumption and AMR in other bacterial species.[14-20] In Europe, strong associations have been found between higher consumption of various antimicrobials in Southern European countries and higher rates of AMR.[15-17, 20] These associations have been found for an array of antimicrobials (including cephalosporins, penicillin, macrolides, fluoroquinolones and cotrimoxazole) and different bacterial species.[17, 18, 20] Our findings suggest that the problem of declining antimicrobial susceptibility in *N. gonorrhoeae* can be productively viewed as being part of this broader problem of AMR induced by higher antimicrobial consumption.[41]

This interpretation supports the results of our previous global study which found country and regional level associations between consumption and resistance for cephalosporins, fluoroquinolones and macrolides (the latter association could not be confirmed in the present study).[26] This was particularly striking for cephalosporins in East Asian countries. Strains resistant to extended-spectrum...
cephalosporins such as cefixime, have been documented in Japan as early as 1995.\textsuperscript{[42]} By 2001, a third of Japanese \textit{N. gonorrhoeae} isolates exhibited decreased susceptibility to cefixime.\textsuperscript{[43]} A plausible explanation for this explosion of cefixime resistance was the intensity of cephalosporin (particularly inappropriate oral cephalosporin) consumption in the general population. In 2000, Japan's consumption of cephalosporins was nearly twice that of the next highest consumer globally and 72 times that of the country with the lowest consumption.\textsuperscript{[26]}

The heterogeneity in the 2009-2015 geometric mean consumption of macrolides, fluoroquinolones and cephalosporins between the 24 countries in this study was greater than that from the 23 counties (2005-2013) included in the analogous Kirkcaldy et al. study from the USA (44-, 6- and 11-fold difference in Europe vs. 4-, 3.9-, and 3.6-fold difference in the United States, respectively; USA data from Table S8 [13]). This greater variation in Europe is one possible explanation for why we found evidence of a positive association between antimicrobial consumption and decreased susceptibility whereas no such association was found in the USA study.
In the present study, we found similar positive associations between antimicrobial consumption and increased MICs and/or AMR that have been found for a range of other bacteria in Europe and elsewhere.[14-16, 18, 20] Thus, countries with high rates of cephalosporin, macrolide and fluoroquinolone consumption frequently have lower susceptibility to these antimicrobial classes in not only *N. gonorrhoeae* but also a wide range of other bacteria.[44] These results suggest that, as has been established for other bacteria,[45, 46] slowing the emergence of AMR in *N. gonorrhoeae* may be assisted by interventions that reduce antimicrobial prescription and consumption in the general population, locally, nationally and internationally. Given the higher prevalence of *N. gonorrhoeae* in groups with higher rates of partner change such as MSM and the frequent emergence and/or spread of AMR in this group[33] particular attention should be placed on also evaluating strategies (such as vaccinations, condom usage and oral mouthwashes[47]) that could reduce antimicrobial consumption in this and other high-risk groups.

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Data availability


Funding

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Transparency declarations

None to declare. All the authors declare that they have no conflicts of interest.
References

23. Weyand NJ. Neisseria models of infection and persistence in the upper respiratory tract. Pathog Dis 2017; 75.
33. Lewis DA. The role of core groups in the emergence and dissemination of antimicrobial-resistant N gonorrhoeae. Sex Transm Infect 2013; 89 Suppl 4:iv47-51.
### Table 1. Prevalence of antimicrobial resistance and 2010-2016 geometric mean of *Neisseria gonorrhoeae* MIC in 24 Euro-GASP countries

<table>
<thead>
<tr>
<th>Antimicrobial</th>
<th>2010-2016 geometric mean MIC</th>
<th>2010-2016 geometric mean MIC of susceptible isolates</th>
<th>2010-2016 geometric mean resistance (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>2010-2016 geometric mean MIC of susceptible isolates</td>
<td>2010-2016 geometric mean resistance (%)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>(country)</td>
<td>Minimum (country)</td>
<td>fd</td>
</tr>
<tr>
<td>Cefixime</td>
<td>0.04 (EL)</td>
<td>0.01 (UK)</td>
<td>4</td>
</tr>
<tr>
<td>Ceftriaxone</td>
<td>0.02 (LU)</td>
<td>0.004 (CZ)</td>
<td>5</td>
</tr>
<tr>
<td>Ciprofloxacin</td>
<td>1.64 (EL)</td>
<td>0.007 (EE)</td>
<td>226</td>
</tr>
<tr>
<td>Azithromycin</td>
<td>0.63 (NO)</td>
<td>0.13 (NL)</td>
<td>5</td>
</tr>
</tbody>
</table>

Euro-GASP, European Gonococcal Antimicrobial Surveillance Programme

*aCountry designations: AT, Austria; BE, Belgium; CZ, Czech Republic; DE, Germany; DK, Denmark; EL, Greece; ES, Spain; FI, Finland; FR, France; HR, Croatia; HU, Hungary; IE, Ireland; IT, Italy; LU, Luxembourg; NL, the Netherlands; PT, Portugal; SE, Sweden; SI, Slovenia; SK, Slovakia; UK, United Kingdom.

*bCountries in bold text are also represented in the high antimicrobial consumption quartile.
Table 2. Antimicrobial consumption and changes in consumption in 24 Euro-GASP countries (the geometric mean for the years 2009-2015 of standard doses/1000 population/year)

<table>
<thead>
<tr>
<th>Antimicrobial</th>
<th>Maximum (country)</th>
<th>Minimum (country)</th>
<th>fd</th>
<th>Top Quartile&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Bottom Quartile&lt;sup&gt;b&lt;/sup&gt;</th>
<th>≥25% increase&lt;sup&gt;b&lt;/sup&gt;</th>
<th>≥25% decrease&lt;sup&gt;b&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cephalosporins</td>
<td>3376 (EL)</td>
<td>76 (NL)</td>
<td>44</td>
<td>EL, SK, DE, LU, HR, PL</td>
<td>NL, DK, SE, NO, UK, LV</td>
<td>UK, SE, SI, FR</td>
<td></td>
</tr>
<tr>
<td>Fluoroquinolones</td>
<td>1501 (IT)</td>
<td>266 (UK)</td>
<td>6</td>
<td>IT, BE, ES, EL, LU, PT</td>
<td>UK, NO, DK, EE, LV, SE</td>
<td>HU</td>
<td>PT, FR</td>
</tr>
<tr>
<td>Macrolides</td>
<td>3300 (EL)</td>
<td>296 (SE)</td>
<td>11</td>
<td>EL, SK, IE, IT, FR, PL</td>
<td>SE, FI, NL, LV, DK, SI</td>
<td>LV</td>
<td>DK, EL, PT, FR</td>
</tr>
</tbody>
</table>

Euro-GASP, European Gonococcal Antimicrobial Surveillance Programme

<sup>a</sup>Top quartile refers to the 6 countries (in descending order of consumption) with the highest geometric mean consumption of the class of antimicrobial for the period 2009 to 2015. 'Bottom quintile' refers to the 6 countries with the lowest consumption and is arranged in ascending order of consumption.

<sup>b</sup>≥25% Increase/decrease in consumption' is defined as countries with ≥25% more/less consumption in 2015 compared to 2009 (or the earliest year with available data). Countries are arranged in descending order of magnitude of change in consumption.

<sup>c</sup>Country designations: AT, Austria; BE, Belgium; CZ, Czech Republic; DE, Germany; DK, Denmark; EL, Greece; ES, Spain; FI, Finland; FR, France; HR, Croatia; HU, Hungary; IE, Ireland; IT, Italy; LU, Luxembourg; LV, Latvia; NL, the Netherlands; PT, Portugal; SE, Sweden; SI, Slovenia; SK, Slovakia; UK, United Kingdom.
Table 3. Mixed effects linear/logistic regression analyses of relationship between antimicrobial consumption (doses/1000 population/year) and geometric mean MIC, geometric mean MIC of susceptible strains and antimicrobial resistance (AMR)

<table>
<thead>
<tr>
<th>Dependent variable</th>
<th>Independent variable</th>
<th>Geometric mean MIC</th>
<th>Geometric mean MIC of susceptible isolates</th>
<th>Antimicrobial resistance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Coefficient</td>
<td>SE</td>
<td>P-value</td>
</tr>
<tr>
<td>AMR/MIC&lt;sub&gt;CRO&lt;/sub&gt;</td>
<td>C&lt;sub&gt;Cephalosporin&lt;/sub&gt;</td>
<td>0.003</td>
<td>0.0008</td>
<td>0.003</td>
</tr>
<tr>
<td>AMR/MIC&lt;sub&gt;CFM&lt;/sub&gt;</td>
<td>C&lt;sub&gt;Cephalosporin&lt;/sub&gt;</td>
<td>0.004</td>
<td>0.0017</td>
<td>0.017</td>
</tr>
<tr>
<td>AMR/MIC&lt;sub&gt;AZM&lt;/sub&gt;</td>
<td>C&lt;sub&gt;Macrolide&lt;/sub&gt;</td>
<td>0.039</td>
<td>0.024</td>
<td>0.115</td>
</tr>
<tr>
<td>AMR/MIC&lt;sub&gt;CIP&lt;/sub&gt;</td>
<td>C&lt;sub&gt;Fluoroquinolone&lt;/sub&gt;</td>
<td>0.495</td>
<td>0.263</td>
<td>0.063</td>
</tr>
</tbody>
</table>

* P<0.05; ** P<0.005; *** P<0.0005; AMR - Antimicrobial resistance, CRO - ceftriaxone, CFM - cefixime, AZM - azithromycin, CIP - Ciprofloxacin; SE - Standard Error
Figure 1. A schematic representation of how differential antimicrobial consumption in the general population (red=antimicrobial exposed individuals) can result in the differential emergence and/or spread of antimicrobial resistance (AMR) in Neisseria gonorrhoeae. After the commencement of high antimicrobial consumption (black arrow), the prevalence/abundance of antimicrobial-susceptible Neisseria commensals (green) declines. This could create a selection pressure for the emergence of AMR strains (red) that could return to equilibrium prevalence/abundance.[34]

Because N. gonorrhoeae is frequently asymptomatic (particularly in women and for rectal and pharyngeal infections in both sexes), N. gonorrhoeae may also be frequently exposed to antimicrobials used for other indications in this population. High antimicrobial exposure may thus place a similar direct AMR selection pressure on N. gonorrhoeae. In addition, the pathway to AMR in N. gonorrhoeae may be indirect: gonococci circulating in the high antimicrobial consumption population may acquire the antimicrobial-resistance-conferring-DNA from the commensal Neisseria spp. via horizontal gene transfer (bidirectional gray arrows).
Figure 2. Kernel density plots of azithromycin MIC in Greece, the Netherlands (2010 to 2016) and a hypothetical population with a bimodal distribution (band width 0.1). Vertical black line depicts breakpoint for antimicrobial resistance.
Figure 3. Scatter plot of country-level consumption of cephalosporins, macrolides and fluoroquinolones (doses/1000 population/year) versus *Neisseria gonorrhoeae* geometric mean MIC of cefixime, ceftriaxone, azithromycin and ciprofloxacin (mg/L) from 2009 to 2016 in 24 Euro-GASP countries. All values are geometric means of values from the years 2009 to 2015 for antimicrobial consumption and 2010 to 2016 for MIC values.
Figure 1

High antimicrobial consumption

Time

Neisseria spp.

High consumption

Low antimicrobial consumption

Time

N. gonorrhoeae

Genetic exchange

Low consumption