The role of "spillover" in antibiotic resistance

Short title: Antibiotic resistance "spillover"

Scott W. Olesen[1], Marc Lipsitch[1,2], Yonatan H. Grad[1,3]

1 Department of Immunology and Infectious Diseases, Harvard T. H. Chan School of Public Health, Boston, Massachusetts 02115

2 Center for Communicable Disease Dynamics, Harvard T. H. Chan School of Public Health, Boston, Massachusetts 02115

3 Division of Infectious Diseases, Department of Medicine, Brigham and Women's Hospital, Harvard Medical School, Boston, Massachusetts 02115

Correspondence:
Yonatan Grad
Harvard T. H. Chan School of Public Health
665 Huntington Ave, Building 1, Room 715, Boston, Massachusetts 02115
617 432 2275
ygrad@hsph.harvard.edu

ABSTRACT (240 words)

Antibiotic use is a key driver of antibiotic resistance. Understanding the quantitative association between antibiotic use and resulting resistance is important for predicting future rates of antibiotic resistance and for designing antibiotic stewardship policy. However, the use-resistance association is complicated by “spillover”, in which one population’s level of antibiotic use affects another population’s level of resistance via the transmission of bacteria between those populations. Spillover is known to have effects at the level of families and hospitals, but it is unclear if spillover is relevant at larger scales. We used mathematical modeling and analysis of observational data to address this question. First, we used dynamical models of antibiotic resistance to predict the effects of spillover. Whereas populations completely isolated from one another do not experience any spillover, we found that if even 1% of interactions are between populations, then spillover may have large consequences: the effect of a change in antibiotic use in one population on antibiotic resistance in that population could be reduced by as much as 50%. Then, we quantified spillover in observational antibiotic use and resistance data from US states and European countries for 3 pathogen-antibiotic combinations, finding that increased interactions between populations were associated with smaller differences in antibiotic resistance between those populations. Thus, spillover may have an important impact at the level of states and countries, which has ramifications for predicting the future of antibiotic resistance, designing antibiotic resistance stewardship policy, and interpreting stewardship interventions.

INTRODUCTION

Antibiotic resistance is a major threat to public health (1). Outpatient antibiotic use, which accounts for approximately 80% of human antibiotic use (2,3), is considered a principal driver of antibiotic resistance in the community (4). Understanding the relationship between use and resistance is important because it allows accurate predictions of the future of antibiotic resistance and goal-oriented antibiotic stewardship policy. The use-resistance association has been previously characterized in many ecological studies at the level of US states (5–7) and European countries (8,9). However, antibiotic resistance is a complex, temporally dynamic phenomenon (10–13), and many factors complicate the use-resistance association, making what should be an "obvious" connection sometimes difficult to identify and quantify (11). Even when detected, observed use-resistance associations are sometimes weaker than might be expected (14). One factor that could account for the difficulty in detecting use-resistance associations in ecological studies and for the apparent weakness of such associations is "spillover".

"Spillover" is a consequence of the fact that antibiotic-resistant and -susceptible bacteria can be transmitted from person to person. Thus, one person's risk of an antibiotic resistant infection depends on their own antibiotic use (15,16) as well as the rates of antibiotic use among their contacts (17). For example, one person's use of antibiotics increases the risk of an antibiotic resistant infection among their family members (18–21). As another example, hospitalized patients with no recent antibiotic use can have a higher risk of resistance than people in the community with high antibiotic use (22)

because antibiotic use and resistance in other hospitalized patients are high.

Spillover is important for three reasons. First, it means antibiotic resistance is not merely a localized problem. It is well-understood that new resistance determinants can emerge in one geography and spread globally (23,24), but the role of spillover in determining the levels of resistance in a given locale is not well-quantified. To what degree, for example, can one US state expect that its antibiotic resistance levels are due to antibiotic use within its borders, rather than in surrounding states? Second, spillover makes it difficult to design antibiotic stewardship interventions and understand their results. For example, if antibiotic use in one hospital changes, resistance might not change as expected because of spillover, from the community or other hospitals into that hospital's patients. Finally, spillover makes it difficult to interpret the results of controlled antibiotic interventions, such as the effect of mass drug administration on antibiotic resistance (25,26), when the intervention and control populations are not wholly epidemiologically separate.

The effect of spillover should scale with the amount of interaction between populations. If two populations do not interact at all, then antibiotic use in one population cannot affect resistance in the other. However, if two populations liberally exchange bacteria, then the rates of antibiotic resistance in the two populations will be very similar, regardless of whether their rates of antibiotic use differ greatly.

The effects of spillover also depend on population sizes. For example, two large

69 populations will have most interactions within themselves, rather than between each
70 other. Spillover should therefore be most pronounced when considering small
71 populations and become less important for large populations. As mentioned above, a
72 single individual's risk of resistance is modulated by antibiotic use in their family or in
73 their healthcare facility. Spillover is also observed at the level of hospitals, as the level
74 of resistance in one hospital appears to be affected by resistance levels in nearby
75 hospitals as well as by antibiotic use rates in the surrounding communities (27–29).
76 Presumably, when examining ever larger populations, such as US Census tracts (30),
77 US states, or European countries, the effect of spillover will become less important.
78 However, the relationship between population size and spillover effects is not well
79 understood.
80
81 We hypothesized that US states and European countries, which are large populations
82 with relatively independent public health policies, may be subject to substantially lower
83 levels of antibiotic resistance spillover than family- or hospital-sized populations. This
84 hypothesis, if true, would mean that individual states or countries could act as
85 independent "laboratories" of antibiotic use and resistance. If not, it means that
86 outpatient antibiotic resistance policy must be national or international in order to
87 achieve its full effect. To evaluate this hypothesis, we first use mathematical models of
88 antibiotic use and resistance to make quantitative predictions about the effect of
89 spillover between populations as a function of their amount of mutual interaction. Then,
90 we search for signals of spillover in observational data of antibiotic use and resistance
91 in US states and European countries.

METHODS

Dynamical model of antibiotic resistance

To examine how interactions between populations could theoretically affect the association between antibiotic use and resistance, we used the within-host neutrality (WHN) mathematical model presented by Davies *et al*. (31) and described in the Supplemental Methods. Briefly, the model predicts the prevalence ρ of antibiotic resistance that results from an antibiotic use rate τ in a single, well-mixed population. To verify that conclusions drawn from the WHN model are not specific to the model structure, we also repeated all analyses with the "D-types" model of use and resistance (32). We selected these two models because they demonstrate coexistence between sensitive and resistant strains at equilibrium over a wide parameter space. Parameter values and simulation methodology for both models are in the Supplemental Methods. In the simulations, antibiotic use is measured as monthly treatments per capita and resistance as the proportion of colonized hosts carrying resistant strains.

To conceptually frame and clarify the question of spillover, we simulated an antibiotic stewardship intervention experiment using a structured host population approach inspired by Blanquart *et al.* (33). We considered pairs of an intervention population with antibiotic use rate τ_{int} and a control population with use rate τ_{cont}. To determine how spillover affects the intervention's measured outcome, we modulated the proportion ε of each population's contacts that are in the other population. For ε = 0%, the populations

115 are completely separate. For ε = 50%, contacts across populations are just as likely as
116 contacts within populations (Supplemental Methods). We varied ε between 0% and
117 50%, and we varied the difference in use $\Delta\tau = \tau_{cont} - \tau_{int}$ between 0 and 0.15 treatments
118 per person per month while keeping the average use $0.5 \times (\tau_{cont} + \tau_{int})$ fixed at 0.125,
119 reflecting the range of antibiotic use rates in the original model presentations.
120
121 *Observational data*
122 We examined antibiotic use and resistance for 3 pathogen-antibiotic combinations: *S.*
123 *pneumoniae* and macrolides, *S. pneumoniae* and β-lactams, and *Escherichia coli* and
124 quinolones. We considered these 3 combinations because they are the subject of many
125 modeling (31,32) and empirical studies (5,15).
126
127 Observational data were drawn from 3 sources. First, we used MarketScan (34) and
128 ResistanceOpen (35) as previously described (7). The MarketScan data includes
129 outpatient pharmacy antibiotic prescription claims for 62 million unique people during
130 2011-2014. ResistanceOpen includes antibiotic resistance data collected during 2012-
131 2015 from 230 hospitals, laboratories, and surveillance units in 44 states. Second, we
132 used the QuintilesIMS Xponent database (36) and the US Centers for Disease Control
133 and Prevention's (CDC) National Healthcare Safety Network (NHSN) (37). The Xponent
134 data includes state-level data on US quinolone use during 2011-2014. NHSN includes
135 state-level data on quinolone resistance among *E. coli* catheter-associated urinary tract
136 infections during 2011-2014. Third, we used the European Center for Disease
137 Prevention and Control's (ECDC) ESAC-Net antimicrobial consumption database (38)

138 and EARS-Net Surveillance Atlas of Infectious Disease (39) for 2011-2015. The ESAC-
139 Net data includes country-level outpatient antibiotic use data provided by WHO and
140 Ministries of Health from member countries. The EARS-Net data includes country-level
141 resistance data. In the observational data, we quantified antibiotic use as yearly
142 treatments per capita and resistance as the proportion of collected isolates that were
143 non-susceptible. Further details about preparation of these data sources and their
144 availability are in the Supplemental Methods.
145
146 We excluded the *S. pneumoniae* resistance to β-lactams in US states from the analysis
147 because, in previous work using the same primary datasets, the point estimate for the
148 use-resistance relationship was negative (14).
149
150 *Use-resistance relationships by populations' adjacency*
151 To test the theoretical prediction that the same difference in antibiotic use will be
152 associated with smaller differences in antibiotic resistance when two populations (US
153 states or European countries) have stronger interactions, we tested whether the use-
154 resistance association is weaker in adjacent pairs of populations, which presumably
155 have more cross-population contacts, compared to non-adjacent populations. Two
156 populations were considered adjacent if they share a land or river border (40–42).
157
158 We quantified the use-resistance association as the percentage point difference (i.e.,
159 absolute risk difference) in resistance (proportion of non-susceptible isolates) divided by
160 the difference in antibiotic use. We summarized use-resistance associations among

161 adjacent pairs and non-adjacent pairs of populations using the median value. Because
162 use-resistance associations between pairs of populations are correlated, we used the
163 jackknife method to compute confidence intervals on the difference in medians between
164 groups. We used the Mann-Whitney *U* test to compute statistical significance. Because
165 our theoretical results suggested the ratio of percentage points difference in resistance
166 divided by difference in antibiotic use rates was a predictable function of the degree of
167 population mixing, we considered only this functional form for the use-resistance
168 association.
169
170 *Use-resistance associations by interactions*
171 Because adjacency might be too coarse measure of populations' interactions to detect
172 spillover, we performed a similar analysis as above, but predicting the use-resistance
173 association between populations using transportation data. For US states, we used
174 inter-county commuting statistics from the US Census (43). For European countries, we
175 used inter-country passenger flight data from Eurostat (44). Rather than trying to infer a
176 precise mathematical relationship between transportation statistics and epidemiological
177 contacts, we used a nonparametric approach: we assumed that pairs of populations
178 with relatively little inter-population transportation also have relatively few inter-
179 population contacts, but we infer only the rank ordering of inter-population contacts, not
180 their magnitudes. Specifically, we first converted the matrix of the number of counts
181 (workers in the commuting data and passengers in the flight data) from each population
182 to every other population into the proportion of counts moving from one population to
183 another (i.e., divided each row by its sum), then symmetrized the resulting matrix (taking

the elementwise average of the matrix and its transpose), and finally converted the resulting values into ranks. We assumed that intra-population interactions outnumber inter-population interactions and so set diagonal entries, which represent within-population interactions, to the highest rank. We measured the association between ranked interactions and use-resistance associations using the nonparametric Spearman's correlation, computed confidence intervals using the jackknife method, and tested for statistical significance using the Mantel test with 999 permutations. To quantify the effect of spillover, we compared the median use-resistance associations among the top and bottom decile of ranked interactions.

Simulations and observational analyses were made using R (version 3.6.0) (45). The Mantel test used the *vegan* package (46). Multiple hypotheses were accounted for using Benjamini-Hochberg false discovery rate.

RESULTS

In simulations of two populations, representing an intervention and control group, interactions between the two populations attenuated the effect of the intervention (Figure 1). With increasing interaction strength, the same intervention, that is, the same difference in antibiotic use between the populations, was associated with a smaller difference in antibiotic resistance. The difference in resistance between populations increases with the difference in antibiotic use (Figure 1d), but the use-resistance association, measured as the ratio of the difference in resistance to the difference in

use, depends strongly on the interaction strength (Figure 1e). Thus, spillover between populations attenuates the measured use-resistance association.

The precise relationship between ε, the proportion of each population's contacts that are in the other population, and the attenuation of the use-resistance association depended on the choice of mathematical model (Supplemental Table 1, Supplemental Figure 1). For ε = 1%, the use-resistance declined by approximately 30% in the WHN model and more than 60% in the "D-types" model. In other words, the models predict that as few as 1% of contacts need to be across populations, rather than within populations, to cause the observed effect of an antibiotic stewardship intervention to shrink by one-third, or even half.

To test whether spillover is important at the scale of US states or European countries, we measured use-resistance associations between pairs of populations in 6 combinations of pathogen species, antibiotic class, and data source (Figure 2). We reasoned that if spillover is relevant at these scales, then pairs of states or countries with stronger interactions would have detectably weaker use-resistance associations.

We first tested whether pairs of physically adjacent populations (e.g., Massachusetts and Connecticut) had weaker use-resistance associations than non-adjacent populations (e.g., Massachusetts and Alaska). In 5 of 6 pathogen/antibiotic/dataset combinations, the median use-resistance association was smaller among adjacent populations than among non-adjacent populations (Figure 3), but in no case was the

difference statistically significant after multiple hypothesis correction (Supplemental Table 2). Point estimates of the relative difference in median use-resistance associations between adjacent populations were 18% to 50% weaker than between non-adjacent populations (excepting *S. pneumoniae* with β-lactams, which was an outlier), consistent with the theoretical modeling results showing a 50% reduction in the use-resistance association for populations with approximately 1% of interactions across populations (Supplemental Table 1).

Next, to account for the possibility that adjacency was too coarse a measure for interactions between populations, we instead used a rank-ordered estimate of interactions using US commuting and European airline passenger flows (Supplemental Figure 2). In 4 of 6 dataset/pathogen/antibiotic combinations, the nonparametric association between increased inter-population interactions and decreased use-resistance associations was statistically significant, thus confirming the general trend observed in the adjacency analysis (Figure 4, Supplemental Table 3). The weakest significant result was for *S. pneumoniae* and macrolides in the MarketScan/ResistanceOpen dataset (Spearman's ρ 0.07, 95% jackknife confidence interval -0.03 to 0.18; $p = 0.028$, Mantel test), and the strongest was for *E. coli* and quinolones in the Xponent/NHSN dataset ($\rho = 0.13$, 95% jackknife confidence interval 0.007 to 0.25; $p = 0.001$). The correlation for *S. pneumoniae* and macrolides in the ECDC data has a point estimate suggesting spillover but was not statistically significant, while the correlation for *S. pneumoniae* and β-lactams in the ECDC data had the opposite point estimate, consistent with the adjacency analysis (Figure 3).

Finally, to quantify the effect of increased interactions on the observed use-resistance associations, we compared the use-resistance associations in pairs of populations within the lowest decile of interactions against those in the highest decile, using the same approach as for the adjacency analysis above (Supplemental Table 4). In 5 of 6 dataset/pathogen/antibiotic combinations, the point estimate for the different in use-resistance associations was consistent with spillover, with a weaker association among pairs of populations with greater interactions. In those 5 cases, the point estimates ranged from a 18% reduction up to a 75% reduction in use-resistance associations among the highest-interacting pairs of populations, compared to the lowest-interacting populations.

DISCUSSION

We used theoretical models to show that interactions between two populations can attenuate the observed use-resistance association. In simulations, the quantitative relationship between inter-population interactions and the attenuation of the use-resistance association was dependent on the theoretical model used. However, we found that, in two models of the use-resistance association, having on the order of 1% of interactions between a control and intervention population was sufficient to attenuate the observed effect of theoretical stewardship intervention by 50%, relative to a situation where the two populations were completely isolated. These theoretical results suggest that even small numbers of interactions could lead to substantial spillover.

When examining observational antibiotic use and resistance data from US states and European countries, we did not detect a robust signal of spillover among pairs of adjacent populations, as opposed to non-adjacent pairs, even across 3 pathogen-antibiotic combinations in 3 separate datasets. However, when using more fine-grained transportation data to estimate the relative ranking of epidemiological contacts between those populations, we found a correlation between increased interactions and attenuated use-resistance associations. Pairs of populations in the highest decile of inter-population interactions, that is, those most subject to spillover, had use-resistance associations on the order of 50% weaker than pairs in the lowest decile of interactions. The 2 pathogen/antibiotic dataset combinations with data not indicative of spillover, namely *S. pneumoniae* and β-lactams and macrolides in the ECDC data, may have not shown the same signal as other cases because of the smaller number of populations in those cases (27, versus 28 to 50 in the other cases) led to insufficient statistical power or potentially because the biology or epidemiology of *S. pneumoniae* resistance in these cases is somehow different and does not exhibit spillover.

These theoretical and empirical results suggest that spillover is relevant at the level of US states and European countries. This finding has important ramifications. First, attempts to attribute changes in a population's level of antibiotic resistance to changes in that population's rates of antibiotic use may lead to inaccurate conclusions unless use and resistance in surrounding populations is accounted for. Second, state- or country-level antibiotic stewardship pilot studies may substantially underestimate the potential

299 reduction in antibiotic resistance that would follow from a reduction in antibiotic use if
300 that reduction were implemented at a larger scale. Third, mass drug administration trials
301 may lead to elevated levels of antibiotic resistance in the control populations if those
302 populations are not entirely separated from the intervention population. Finally, spillover
303 can at least partly explain why use-resistance associations at the level of US states or
304 European countries are sometimes difficult to detect and, when they are detected, are
305 sometimes weaker than expected (5,11,14). Furthermore, spillover means that
306 theoretical models of antibiotic use and resistance that treat US states or European
307 countries as epidemiologically independent populations will not accurately represent the
308 dynamics of resistance (33).
309
310 Our study has several limitations. First, we interpreted the theoretical results and
311 ecological data as if the association between antibiotic use and resistance were causal
312 and deterministic. However, decreases in the use of an antibiotic may not necessarily
313 lead to declines in resistance to that antibiotic in a target pathogen (12,47–49). We do
314 not address co-resistance and cross-selection (50,51), and we assumed that resistance
315 equilibrates on a timescale comparable to an intervention. Previous research has shown
316 that resistance among *E. coli*, *S. pneumoniae*, *N. gonorrhoeae* and other organisms can
317 respond to changes in antibiotic use on the timescale of months (52–55), but the
318 expected delay between a perturbation to antibiotic use and the resulting change in
319 resistance remains a subject of active study (13,52,56,57). Nevertheless, the use of
320 ecological data was essential to addressing our hypothesis, as data from multiple
321 controlled, state- or country-wide experiments are not available.

Second, our analyses attributed all differences in antibiotic resistance between populations to differences in use across those populations and to interactions between them. In fact, antibiotic resistance is associated with factors beyond antibiotic use (6,58), and those factor are likely spatially correlated. In other words, closely interacting populations might have more similar use-resistance associations because they tend to be more similar with respect to other determinants of antibiotic use. Our estimates of the correlation between inter-population interactions and the attenuation of use-resistance relationships may therefore be overestimates. A more careful quantification of the relative roles of spillover versus other spatially-correlated determinants of resistance is required.

Third, our analysis only considered pairs of populations, when in fact spillover is happening between all pairs of populations in our analysis simultaneously. We used the pairs approach because it allowed for a simple theoretical model and a straightforward comparison of theory with the observational data. However, more sophisticated approaches that account for the network of spillover interactions will likely lead to more refined characterizations of spillover.

Finally, analyses based on administrative entities like US states or European countries, although logistically attractive “laboratories” of antibiotic stewardship, will always be difficult to interpret because administrative entities average over important dimensions of population structure like age (59), sexual networks (60), and race/ethnicity (61). Thus,

use-resistance associations measured across states and countries may be different from those that appear among geographically-proximate populations with dissimilar antibiotic use rates, such as the sexes (62) and racial/ethnic groups (63).

We suggest 3 lines of investigation that could refine our understanding about the role of spillover at levels of US states and European countries. First, further mathematical modeling studies with more realistic structuring of the host population might articulate more detailed theoretical expectations about the relationship between intervention scale and spillover. For example, models could be parameterized with epidemiological information about individuals' contacts and travel patterns, as has been done for other infectious diseases (64). Second, meta-analysis of existing studies of use-resistance relationships (5,65,66), both experimental and observational, could potentially determine the empirical relationship between intervention population size and the importance of spillover. This kind of meta-analysis might reveal that populations other than US states are feasible "laboratories" for resistance: it may be that cities, daycares, schools, workplaces, or even families represent the optimal trade-off between maximizing logistical feasibility and minimizing spillover. Finally, future experimental outpatient antibiotic stewardship interventions should make careful and deliberate decisions about the sizes and interconnectedness of the populations they target. We hope that a better understanding of spillover will improve predictions about the future of antibiotic resistance, the formulation of stewardship policy, the design of stewardship interventions and antibiotic administration trials, and theoretical models of resistance.

368 **References**

369 1. Review on Antimicrobial Resistance. Tackling drug-resistant infections globally:
370 final report and recommendations. 2016.

371 2. Public Health England. English Surveillance Programme for Antimicrobial
372 Utilisation and Resistance (ESPAUR). 2014.

373 3. Public Health Agency of Sweden, National Veterinary Institute. Consumption of
374 antibiotics and occurrence of antibiotic resistance in Sweden.

375 4. US Centers for Disease Control and Prevention. Antibiotic Resistance Threats in
376 the United Staes, 2013. 2013.

377 5. Bell BG, Schellevis F, Stobberingh E, Goossens H, Pringle M. A systematic review
378 and meta-analysis of the effects of antibiotic consumption on antibiotic resistance.
379 BMC Infect Dis. 2014 Jan 9;14:13.

380 6. MacFadden DR, McGough SF, Fisman D, Santillana M, Brownstein JS. Antibiotic
381 resistance increases with local temperature. Nat Clim Change. 2018 Jun;8(6):510–
382 4.

383 7. Olesen SW, Barnett ML, MacFadden DR, Brownstein JS, Hernández-Diaz S,
384 Lipsitch M, et al. The distribution of antibiotic use and its association with antibiotic
385 resistance. eLife [Internet]. 2018 Dec 18 [cited 2018 Dec 19];7. Available from:
386 https://elifesciences.org/articles/39435

387 8. Goossens H, Ferech M, Vander Stichele R, Elseviers M. Outpatient antibiotic use
388 in Europe and association with resistance: a cross-national database study. The
389 Lancet. 2005 Feb 12;365(9459):579–87.

390 9. van de Sande-Bruinsma N, Grundmann H, Verloo D, Tiemersma E, Monen J,
391 Goossens H, et al. Antimicrobial drug use and resistance in Europe. Emerg Infect
392 Dis. 2008 Nov;14(11):1722–30.

393 10. Livermore DM. The 2018 Garrod Lecture: Preparing for the Black Swans of
394 resistance. J Antimicrob Chemother. 2018 Nov 1;73(11):2907–15.

395 11. Turnidge J, Christiansen K. Antibiotic use and resistance--proving the obvious.
396 Lancet Lond Engl. 2005 Feb;365(9459):548–9.

397 12. Arason VA, Gunnlaugsson A, Sigurdsson JA, Erlendsdottir H, Gudmundsson S,
398 Kristinsson KG. Clonal Spread of Resistant Pneumococci Despite Diminished
399 Antimicrobial Use. Microb Drug Resist. 2002 Sep;8(3):187–92.

400 13. Lipsitch M. The rise and fall of antimicrobial resistance. Trends Microbiol. 2001
401 Sep;9(9):438–44.

402 14. Olesen SW, Barnett ML, MacFadden DR, Brownstein JS, Hernández-Díaz S,
403 Lipsitch M, et al. The distribution of antibiotic use and its association with antibiotic
404 resistance. eLife [Internet]. 2018 Dec 18 [cited 2019 Sep 23];7. Available from:
405 https://elifesciences.org/articles/39435

406 15. Costelloe C, Metcalfe C, Lovering A, Mant D, Hay AD. Effect of antibiotic
407 prescribing in primary care on antimicrobial resistance in individual patients:
408 systematic review and meta-analysis. BMJ. 2010 May 18;340:c2096.

409 16. Harbarth S, Harris AD, Carmeli Y, Samore MH. Parallel analysis of individual and
410 aggregated data on antibiotic exposure and resistance in gram-begative bacilli. Clin
411 Infect Dis. 2001;33:1462–8.

412 17. Lipsitch M. Measuring and Interpreting Associations between Antibiotic Use and
413 Penicillin Resistance in Streptococcus pneumoniae. Clin Infect Dis. 2001 Apr
414 1;32(7):1044–54.

415 18. Hannah EL, Angulo FJ, Johnson JR, Haddadin B, Williamson J, Samore MH. Drug-
416 resistant Escherichia coli, Rural Idaho. Emerg Infect Dis. 2005 Oct;11(10):1614–7.

417 19. Kalter HD, Gilman RH, Moulton LH, Cullotta AR, Cabrera L, Velapatiño B. Risk
418 factors for antibiotic-resistant Escherichia coli carriage in young children in Peru:
419 community-based cross-sectional prevalence study. Am J Trop Med Hyg. 2010
420 May;82(5):879–88.

421 20. Samore MH, Magill MK, Alder SC, Severina E, Morrison-De Boer L, Lyon JL, et al.
422 High rates of multiple antibiotic resistance in Streptococcus pneumoniae from
423 healthy children living in isolated rural communities: association with cephalosporin
424 use and intrafamilial transmission. Pediatrics. 2001 Oct;108(4):856–65.

425 21. Gottesman B-S, Low M, Almog R, Chowers M. Quinolone Consumption by Mothers
426 Increases Their Children's Risk of Acquiring Quinolone-Resistant Bacteriuria. Clin
427 Infect Dis [Internet]. 2019 Aug 29 [cited 2019 Dec 2]; Available from:
428 https://academic.oup.com/cid/advance-article/doi/10.1093/cid/ciz858/5556473

429 22. Lipsitch M, Samore MH. Antimicrobial use and antimicrobial resistance: a
430 population perspective. Emerg Infect Dis. 2002;8:347–54.

431 23. Moellering RC. NDM-1 — A Cause for Worldwide Concern. N Engl J Med. 2010
432 Dec 16;363(25):2377–9.

433 24. Wang R, van Dorp L, Shaw LP, Bradley P, Wang Q, Wang X, et al. The global
434 distribution and spread of the mobilized colistin resistance gene mcr-1. Nat
435 Commun [Internet]. 2018 Dec [cited 2020 Jun 26];9(1). Available from:
436 http://www.nature.com/articles/s41467-018-03205-z

437 25. Bogoch II, Utzinger J, Lo NC, Andrews JR. Antibacterial mass drug administration
438 for child mortality reduction: Opportunities, concerns, and possible next steps.
439 Bhutta ZA, editor. PLoS Negl Trop Dis. 2019 May 23;13(5):e0007315.

440 26. Doan T, Arzika AM, Hinterwirth A, Maliki R, Zhong L, Cummings S, et al. Macrolide
441 Resistance in MORDOR I — A Cluster-Randomized Trial in Niger. N Engl J Med.
442 2019 Jun 6;380(23):2271–3.

443 27. Cooper BS, Medley GF, Stone SP, Kibbler CC, Cookson BD, Roberts JA, et al.
444 Methicillin-resistant Staphylococcus aureus in hospitals and the community: Stealth
445 dynamics and control catastrophes. Proc Natl Acad Sci. 2004 Jul 6;101(27):10223–
446 8.

447 28. MacFadden DR, Fishman DN, Hanage WP, Lipsitch M. The Relative Impact of
448 Community and Hospital Antibiotic Use on the Selection of Extended-Spectrum
449 Beta-lactamase-Producing Escherichia coli. Clin Infect Dis [Internet]. 2018 [cited
450 2018 Nov 29]; Available from: https://www.ncbi.nlm.nih.gov/pubmed/30462185

451 29. Knight GM, Costelloe C, Deeny SR, Moore LSP, Hopkins S, Johnson AP, et al.
452 Quantifying where human acquisition of antibiotic resistance occurs: a
453 mathematical modelling study. BMC Med. 2018 23;16(1):137.

454 30. Klevens RM, Caten E, Olesen SW, DeMaria A, Troppy S, Grad YH. Outpatient
455 Antibiotic Prescribing in Massachusetts, 2011–2015. Open Forum Infect Dis
456 [Internet]. 2019 May 1 [cited 2019 Sep 15];6(5). Available from:
457 https://academic.oup.com/ofid/article/doi/10.1093/ofid/ofz169/5430941

458 31. Davies NG, Flasche S, Jit M, Atkins KE. Within-host dynamics shape antibiotic
459 resistance in commensal bacteria. Nat Ecol Evol. 2019 Mar;3(3):440–9.

460 32. Lehtinen S, Blanquart F, Croucher NJ, Turner P, Lipsitch M, Fraser C. Evolution of
461 antibiotic resistance is linked to any genetic mechanism affecting bacterial duration
462 of carriage. Proc Natl Acad Sci. 2017 Jan 31;114(5):1075–80.

463 33. Blanquart F, Lehtinen S, Lipsitch M, Fraser C. The evolution of antibiotic resistance
464 in a structured host population. J R Soc Interface. 2018 Jun;15(143).

465 34. Truven Health MarketScan Database. Commercial Claims and Encounters. Ann
466 Arbor, MI; 2015.

467 35. MacFadden DR, Fisman D, Andre J, Ara Y, Majumder MS, Bogoch II, et al. A
468 Platform for Monitoring Regional Antimicrobial Resistance, Using Online Data
469 Sources: ResistanceOpen. J Infect Dis. 2016 Dec 1;214(suppl_4):S393–8.

470 36. US Centers for Disease Control and Prevention. Patient Safety Atlas - Outpatient
471 Antibiotic Use [Internet]. [cited 2018 Oct 25]. Available from:
472 https://gis.cdc.gov/grasp/PSA/indexAU.html

37. US Centers for Disease Control and Prevention. Patient Safety Atlas - Antibiotic Resistance [Internet]. [cited 2018 Oct 25]. Available from: https://gis.cdc.gov/grasp/PSA/AboutTheData.html

38. European Centre for Disease Prevention and Control. Antimicrobial consumption database [Internet]. European Centre for Disease Prevention and Control. [cited 2018 Oct 25]. Available from: http://ecdc.europa.eu/en/antimicrobial-consumption/surveillance-and-disease-data/database

39. European Centre for Disease Prevention and Control. Surveillance Atlas of Infectious Diseases [Internet]. [cited 2018 Oct 25]. Available from: https://atlas.ecdc.europa.eu/public/index.aspx

40. US Census. County Adjacency File [Internet]. Available from: https://www.census.gov/geo/reference/county-adjacency.html

41. Correlates of War Project. Direct Contiguity Data, 1816-2016 (v3.2) [Internet]. Available from: http://correlatesofwar.org/data-sets/direct-contiguity

42. Stinnett DM, Tir J, Diehl PF, Schafer P, Gochman C. The Correlates of War (Cow) Project Direct Contiguity Data, Version 3.0. Confl Manag Peace Sci. 2002 Sep;19(2):59–67.

43. US Census. 2011-2015 5-Year ACS Commuting Flows [Internet]. Available from: https://www.census.gov/data/tables/2015/demo/metro-micro/commuting-flows-2015.html

44. Eurostat. Air transport measurement [Internet]. Available from: https://ec.europa.eu/eurostat/cache/metadata/en/avia_pa_esms.htm

45. R Core Team. R: A language and environment for statistical computing [Internet]. Vienna; 2018. Available from: www.R-project.org

46. Oksanen J, Blanchet FG, Friendly M, Kindt R, Legendre P, McGlinn D, et al. vegan: Community Ecology Package. R package version 2.5-6.

47. Hennessy TW, Petersen KM, Bruden D, Parkinson AJ, Hurlburt D, Getty M, et al. Changes in Antibiotic-Prescribing Practices and Carriage of Penicillin-Resistant *Streptococcus pneumoniae:* A Controlled Intervention Trial in Rural Alaska. Clin Infect Dis. 2002 Jun 15;34(12):1543–50.

48. Sundqvist M, Geli P, Andersson DI, Sjölund-Karlsson M, Runehagen A, Cars H, et al. Little evidence for reversibility of trimethoprim resistance after a drastic reduction in trimethoprim use. J Antimicrob Chemother. 2010 Feb;65(2):350–60.

49. Enne VI, Livermore DM, Stephens P, Hall LM. Persistence of sulphonamide resistance in Escherichia coli in the UK despite national prescribing restriction. Lancet Lond Engl. 2001 Apr 28;357(9265):1325–8.

50. Pouwels KB, Freeman R, Muller-Pebody B, Rooney G, Henderson KL, Robotham JV, et al. Association between use of different antibiotics and trimethoprimresistance: going beyond the obvious crude association. J Antimicrob Chemoth. 2018;

51. Tedijanto C, Olesen SW, Grad YH, Lipsitch M. Estimating the proportion of bystander selection for antibiotic resistance among potentially pathogenic bacterial flora. Proc Natl Acad Sci. 2018 Dec 18;115(51):E11988–95.

52. Olesen SW, Torrone EA, Papp JR, Kirkcaldy RD, Lipsitch M, Grad YH. Azithromycin Susceptibility Among Neisseria gonorrhoeae Isolates and Seasonal Macrolide Use. J Infect Dis [Internet]. 2018 [cited 2018 Nov 6]; Available from: http://academic.oup.com/jid/advance-article/doi/10.1093/infdis/jiy551/5098400

53. Dagan R, Barkai G, Givon-Lavi N, Sharf AZ, Vardy D, Cohen T, et al. Seasonality of Antibiotic-Resistant Streptococcus pneumoniae That Causes Acute Otitis Media: A Clue for an Antibiotic-Restriction Policy? J Infect Dis. 2008 Apr 15;197(8):1094–102.

54. Sun L, Klein EY, Laxminarayan R. Seasonality and Temporal Correlation between Community Antibiotic Use and Resistance in the United States. Clin Infect Dis. 2012 Sep 1;55(5):687–94.

55. Blanquart F, Lehtinen S, Fraser C. An evolutionary model to predict the frequency of antibiotic resistance under seasonal antibiotic use, and an application to Streptococcus pneumoniae. Proc R Soc B. 2017 May 31;284(1855):20170679.

56. Dingle KE, Didelot X, Quan TP, Eyre DW, Stoesser N, Golubchik T, et al. Effects of control interventions on Clostridium difficile infection in England: an observational study. Lancet Infect Dis. 2017 Apr;17(4):411–21.

57. McCormick AW, Whitney CG, Farley MM, Lynfield R, Harrison LH, Bennett NM, et al. Geographic diversity and temporal trends of antimicrobial resistance in Streptococcus pneumoniae in the United States. Nat Med. 2003 Apr;9(4):424–30.

58. Collignon P, Beggs JJ, Walsh TR, Gandra S, Laxminarayan R. Anthropological and socioeconomic factors contributing to global antimicrobial resistance: a univariate and multivariable analysis. Lancet Planet Health. 2018 Sep;2(9):e398–405.

59. Mossong J, Hens N, Jit M, Beutels P, Auranen K, Mikolajczyk R, et al. Social contacts and mixing patterns relevant to the spread of infectious diseases. PLoS Med. 2008 Mar 25;5(3):e74.

60. Garett G, Hughes J, Anderson R, Stoner B, Aral S, Whittington W, et al. Sexual mixing patterns of patients attending sexually transmitted diseases clinics. Sex Transm Dis. 1996;23(3):248–57.

61. Newman MEJ. Mixing patterns in networks. Phys Rev E Stat Nonlin Soft Matter Phys. 2003 Feb;67(2 Pt 2):026126.

62. Hicks LA, Bartoces MG, Roberts RM, Suda KJ, Hunkler RJ, Taylor TH, et al. US Outpatient Antibiotic Prescribing Variation According to Geography, Patient Population, and Provider Specialty in 2011. Clin Infect Dis. 2015 May 1;60(9):1308–16.

63. Olesen SW, Grad YH. Racial/Ethnic Disparities in Antimicrobial Drug Use, United States, 2014–2015. Emerg Infect Dis [Internet]. 2018 [cited 2018 Nov 6];24(11). Available from: https://wwwnc.cdc.gov/eid/article/24/11/18-0762_article

64. Charu V, Zeger S, Gog J, Bjørnstad ON, Kissler S, Simonsen L, et al. Human mobility and the spatial transmission of influenza in the United States. PLoS Comput Biol. 2017;13(2):e1005382.

65. Schechner V, Temkin E, Harbarth S, Carmeli Y, Schwaber MJ. Epidemiological interpretation of studies examining the effect of antibiotic usage on resistance. Clin Microbiol Rev. 2013 Apr;26(2):289–307.

66. O'Brien KS, Emerson P, Hooper P, Reingold AL, Dennis EG, Keenan JD, et al. Antimicrobial resistance following mass azithromycin distribution for trachoma: a systematic review. Lancet Infect Dis [Internet]. 2018 Oct 3 [cited 2018 Nov 7]; Available from: http://www.sciencedirect.com/science/article/pii/S1473309918304444

Disclaimers

The views and opinions of the authors expressed herein do not necessarily state or reflect those of the ECDC. The accuracy of the authors' statistical analysis and the findings they report are not the responsibility of ECDC. ECDC is not responsible for conclusions or opinions drawn from the data provided. ECDC is not responsible for the correctness of the data and for data management, data merging and data collation after provision of the data. ECDC shall not be held liable for improper or incorrect use of the data.

Funding

This work was supported by the National Institutes of Health (grant number U54GM088558 to ML). The funders had no role in study design, data collection and interpretation, or the decision to submit the work for publication.

Acknowledgements

We thank Dr. Stephen M. Kissler for helpful comments on the manuscript.

Figure 1. **Interactions between populations attenuate the effect of interventions.** (*a*) Schematic of the 2-population model. (*b*) Results of simulations of the 2-population WHN model for a modest intervention (difference in antibiotic use between populations $\Delta\tau$ = 0.05 monthly treatments per capita; average of control and intervention treatment rates 0.125). As interaction strength (ε, horizontal axis) increases, the difference in antibiotic resistance between the two populations decreases. Dotted line shows resistance level in populations before the intervention. (*c*) The same pattern holds for a stronger intervention ($\Delta\tau$ = 0.1, same average treatment rate). (*d*) In general, the difference in resistance between populations ($\Delta\rho$, vertical axis) increases with the difference in antibiotic use ($\Delta\tau$, horizontal axis). (*e*) However, in the WHN model, the use-resistance relationship ($\Delta\rho/\Delta\tau$, vertical axis) depends mostly on the interaction strength ε and is mostly independent of the difference in antibiotic use $\Delta\tau$.

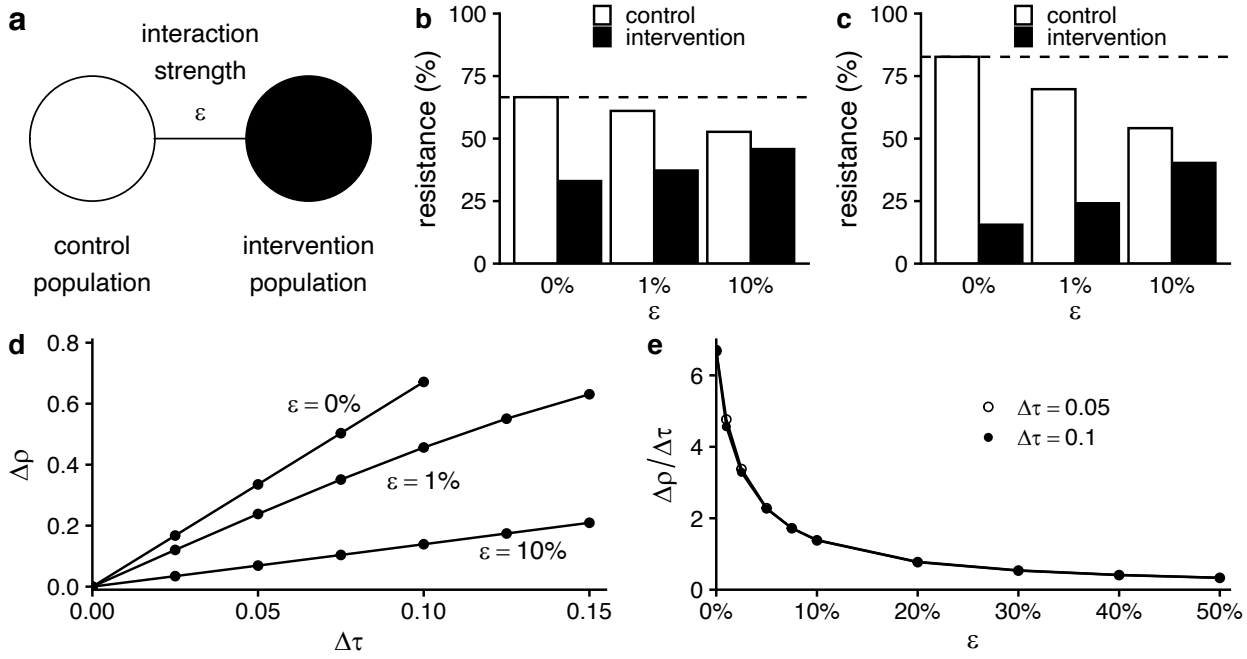
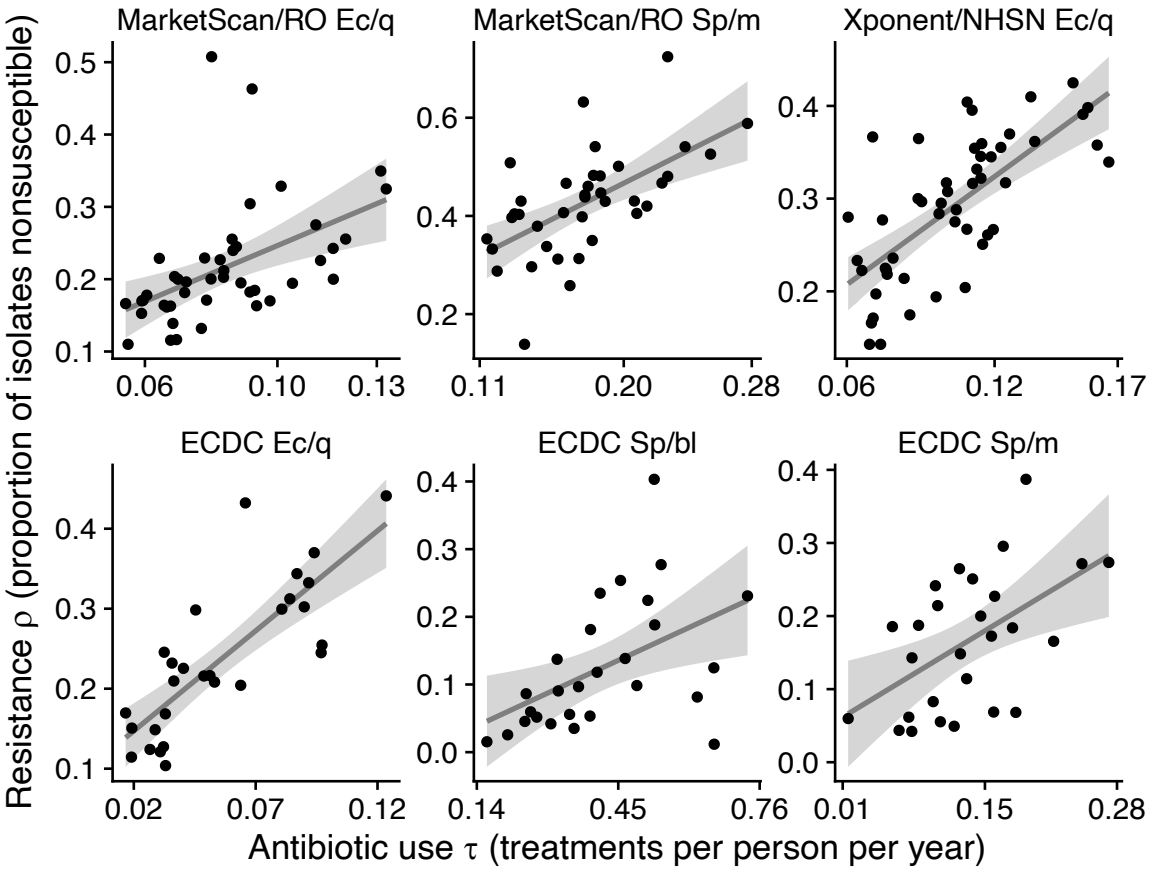
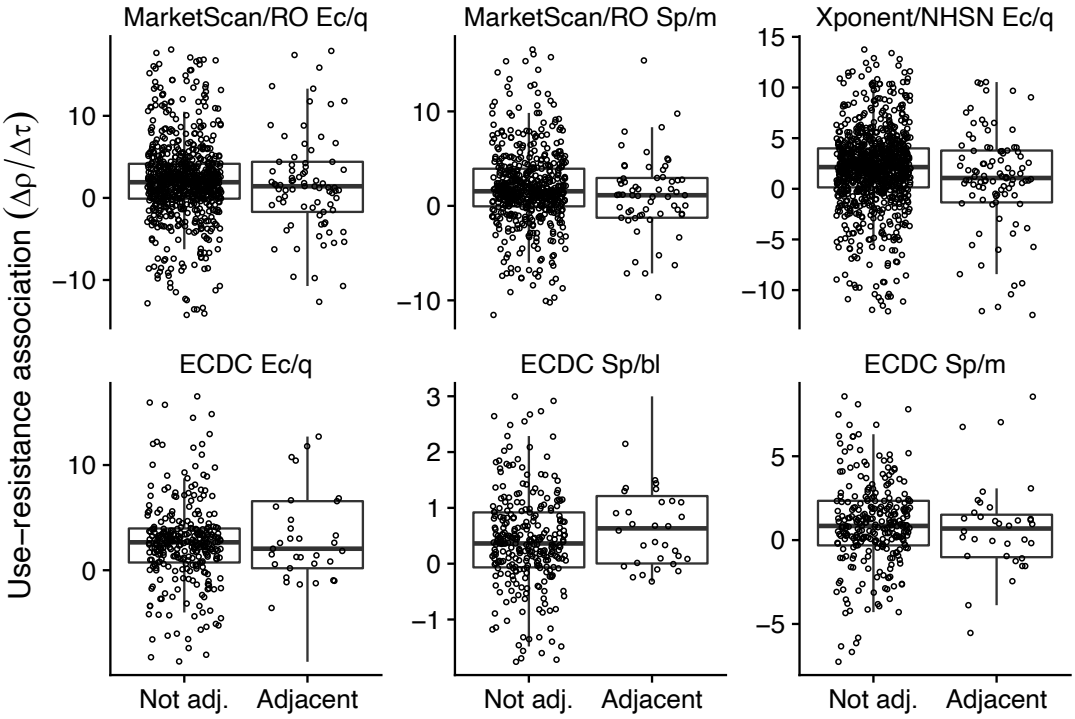


Figure 2. **Use-resistance relationships across US states and European countries.** Each point represents antibiotic use and resistance in a US state (top row) or European country (bottom row). Lines show simple linear regression best fit. Gray areas show 95% confidence interval. Ec/q: *E. coli* and quinolones. Sp/m: *S. pneunomiae* and macrolides. Sp/bl: *S. pneumoniae* and β-lactams. RO: ResistanceOpen. ECDC: European CDC.



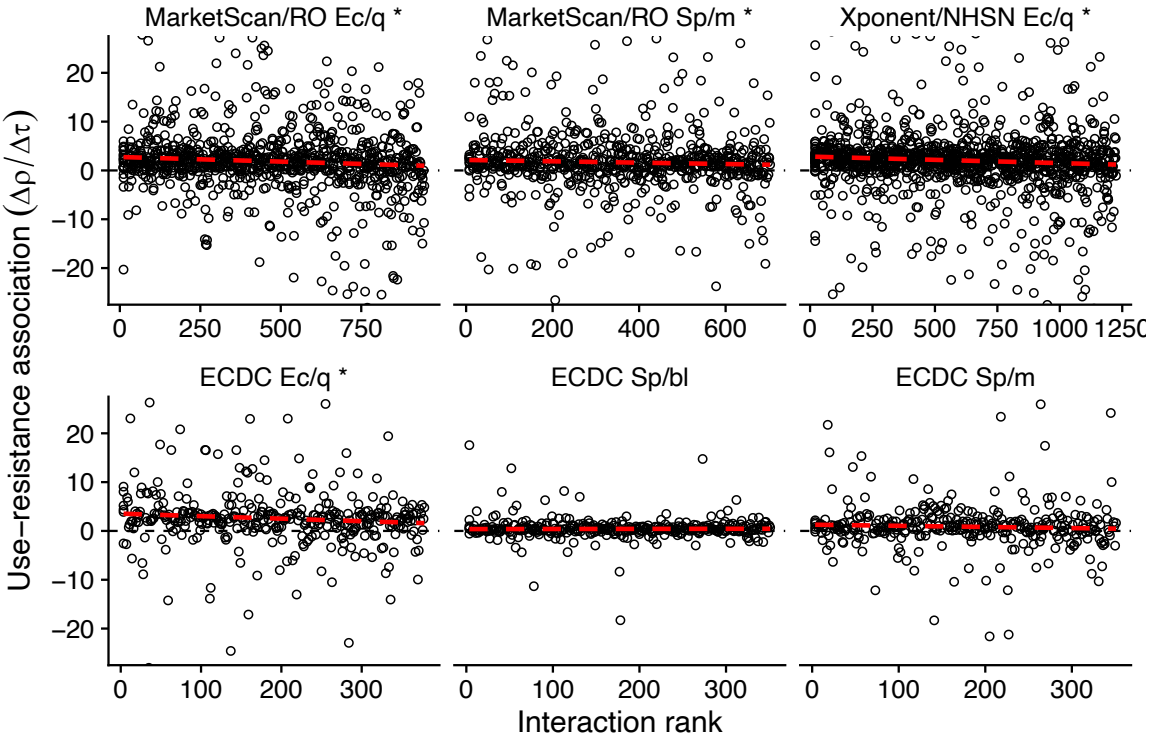
607 Figure 3. **Use-resistance relationships by adjacency.** Each point represents the use-
608 resistance association in populations (top row, US states; bottom row, European
609 countries), the same as those shown in Figure 2, arranged by whether the pair of
610 populations is physically adjacent. Physically adjacent populations tend to have weaker
611 use-resistance associations, but differences were not statistically significant. For visual
612 clarity, the vertical axes are truncated to show only the central 90% of data points. Ec/q:
613 *E. coli* and quinolones. Sp/m: *S. pneunomiae* and macrolides. Sp/bl: *S. pneumoniae*
614 and β-lactams. RO: ResistanceOpen. ECDC: European CDC.



615

616

617 Figure 4. **Use-resistance associations by ranked interaction.** Each point represents
618 the use-resistance association in a pair of US states (top row) or European countries
619 (bottom row), the same pairs as shown in Figure 3, rank ordered by increasing inter-
620 population interaction as inferred from transportation data. For visual clarity, the
621 horizontal axes are truncated to exclude outliers. The dashed red line is a visual
622 illustration of how increasing interaction is correlated with decreasing use-resistance
623 associations (robust regression; compare Supplemental Tables 3 and 4). The asterisk
624 (*) indicates a statistically significant association between increased interaction and
625 decreased use-resistance relationship (Supplemental Table 3). Ec/q: *E. coli* and
626 quinolones. Sp/m: *S. pneunomiae* and macrolides. Sp/bl: *S. pneumoniae* and β-
627 lactams. RO: ResistanceOpen. ECDC: European CDC.



628

629