

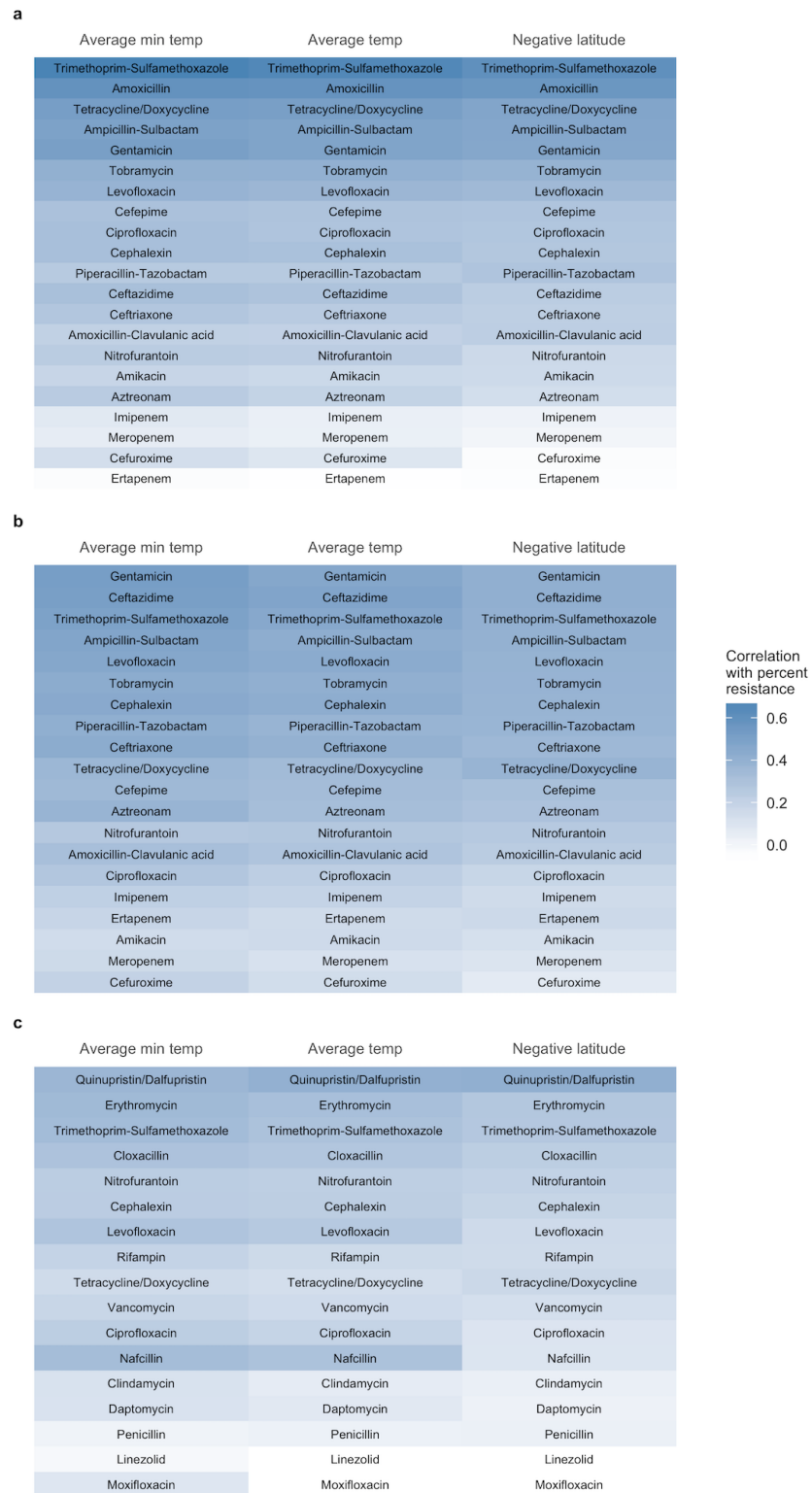
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Antibiotic resistance increases with local temperature

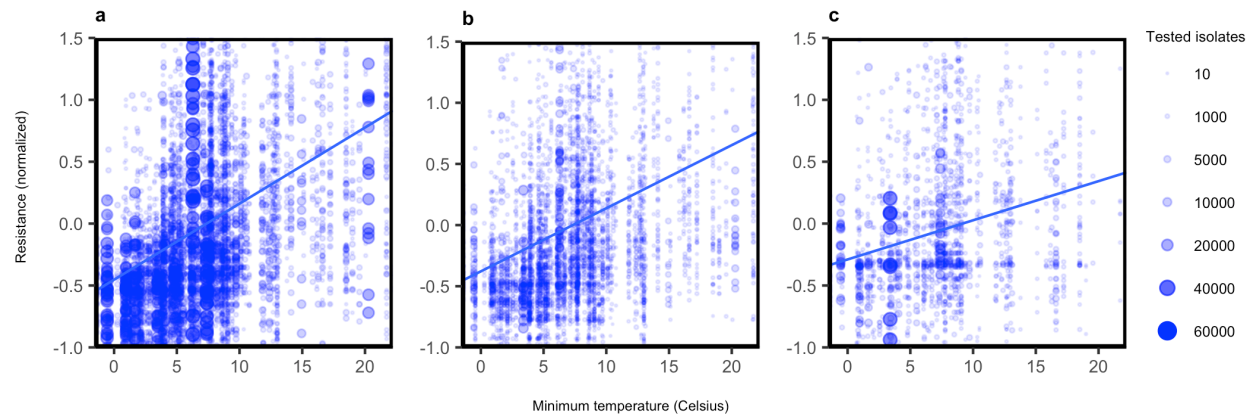
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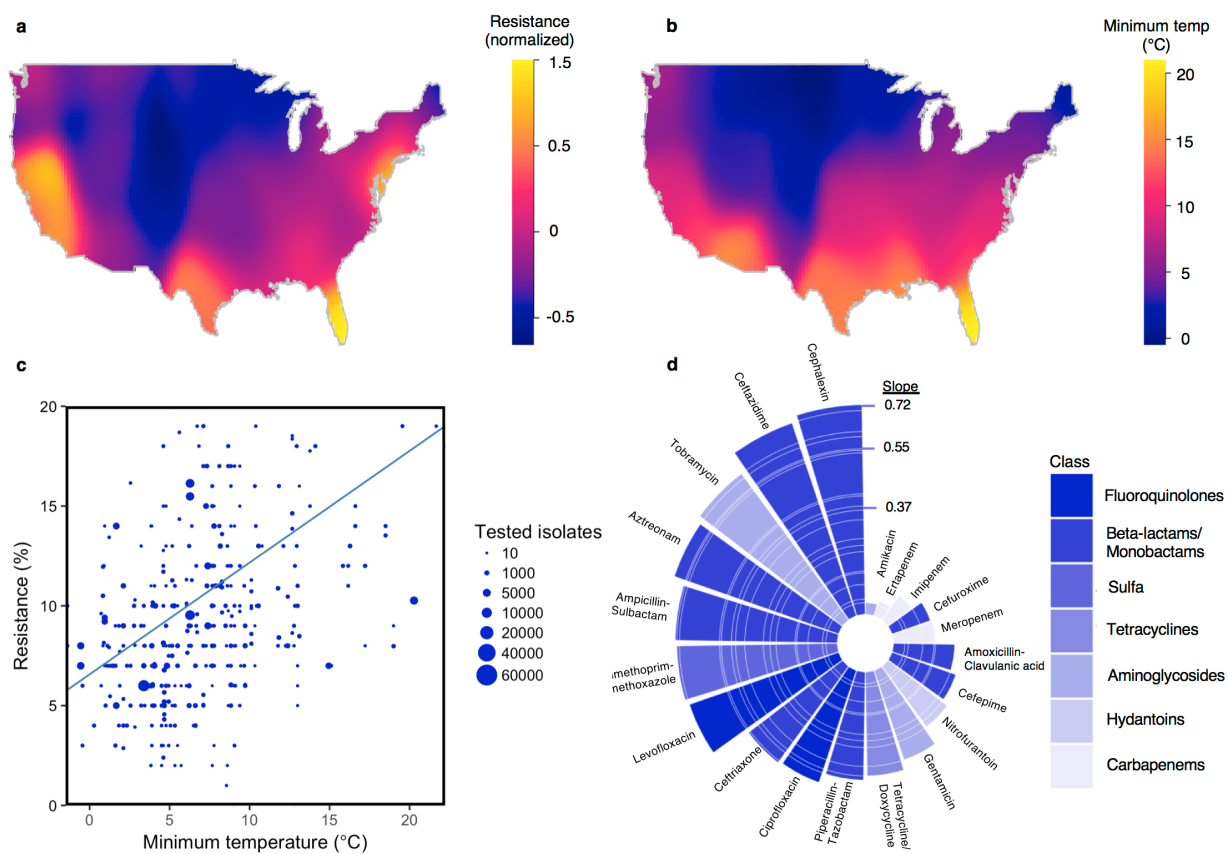
Antibiotic Resistance Increases with Local Temperature: Supplementary Information



Supplementary Figure 1 | Correlation between climate and latitude variables with antibiotic resistance. Correlation of predictor (30-year mean minimum temperature, 30-year mean temperature, latitude) with antibiotic resistance (%) by pathogen: (A) *E. coli*; (B) *K. pneumoniae*; and (C) *S. aureus*.

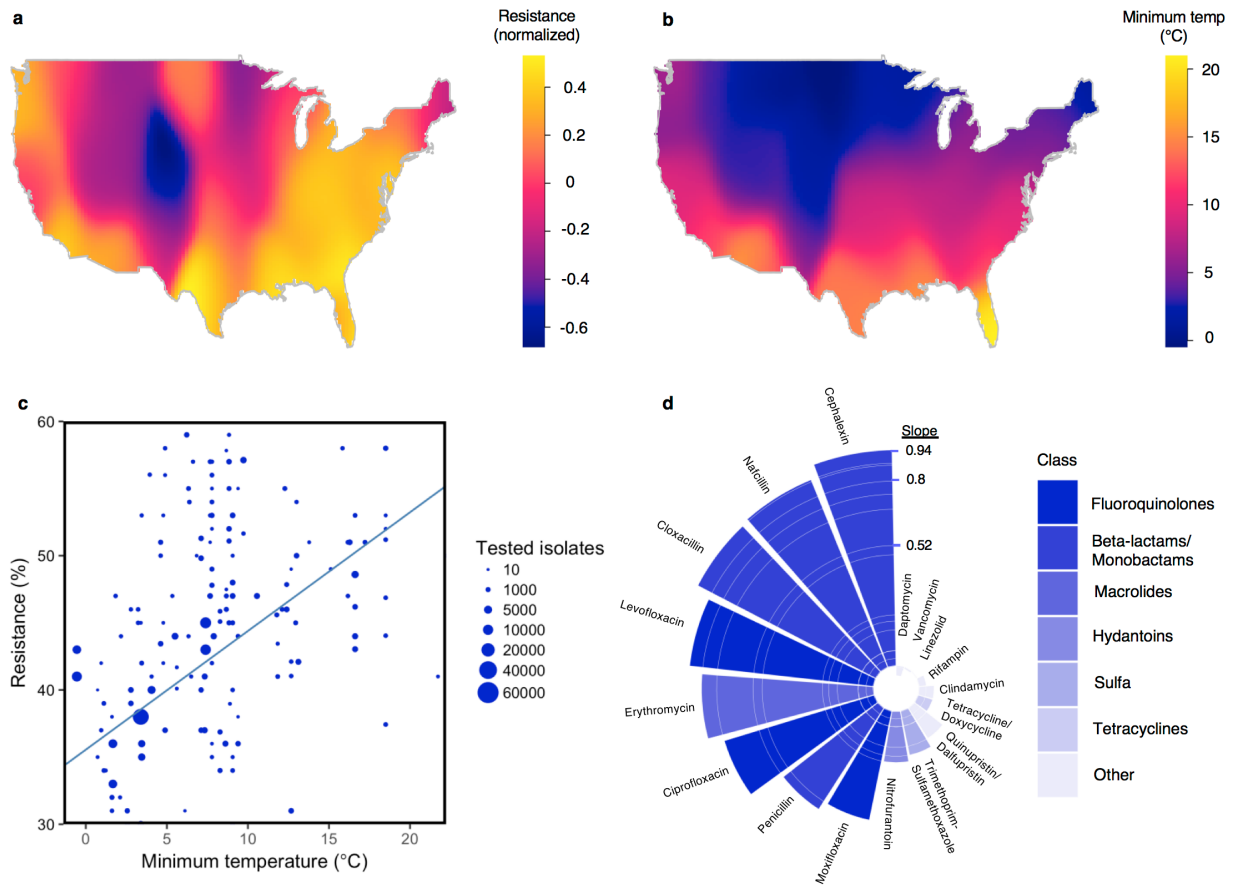


Supplementary Figure 2 | Association between minimum temperature and antibiotic resistance. Scatter plots of mean normalized antibiotic resistance versus minimum temperature ($^{\circ}\text{C}$) for all tested antibiotics, by pathogen: (A) *E. coli*; (B) *K. pneumoniae*; and (C) *S. aureus*. Unadjusted weighted linear trend lines are shown in blue.



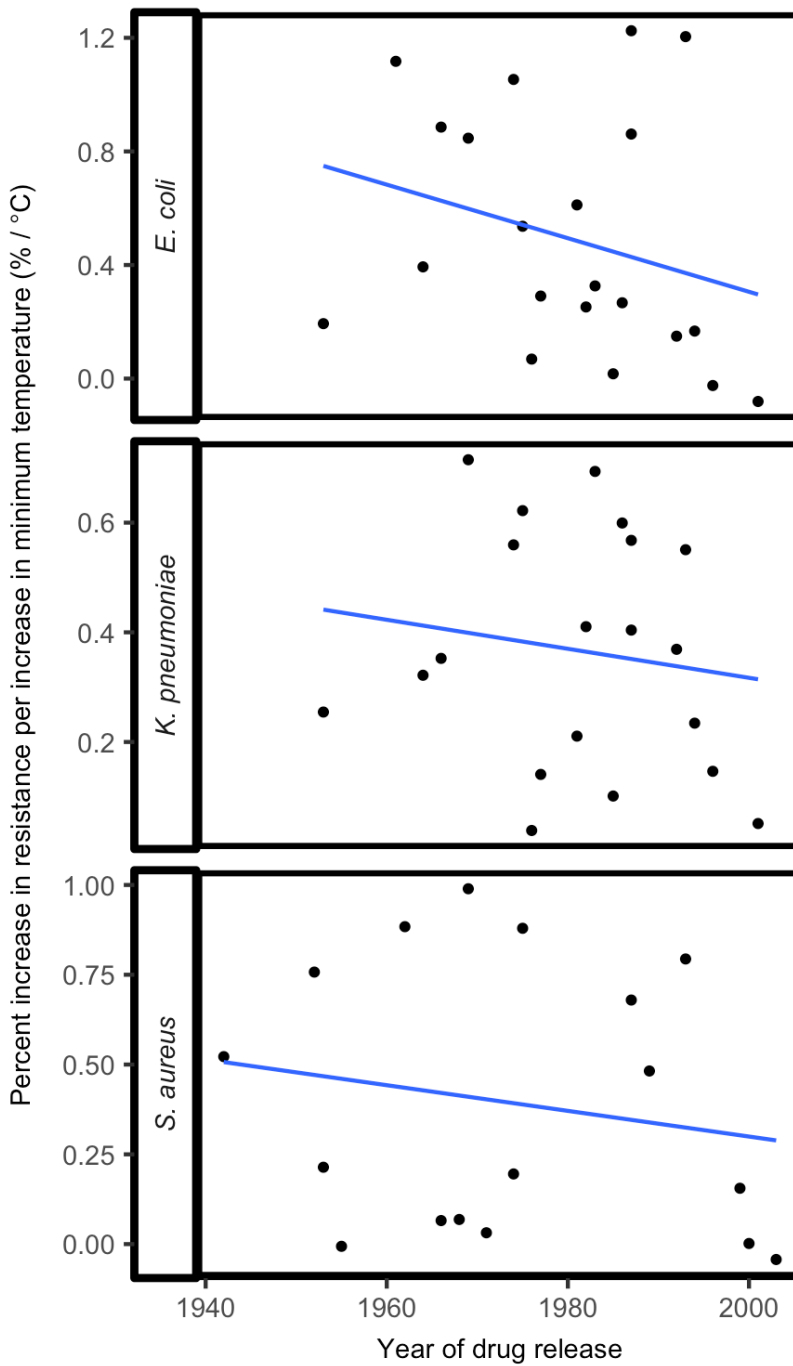
Supplementary Figure 3 | Antibiotic resistance and minimum temperature (*K.*

pneumoniae). (A) A heatmap of mean normalized antibiotic resistance for *K. pneumoniae* for all antibiotics across the United States. (B) A heatmap of 30-year average minimum temperature (°C) across the United States. (C) A scatter plot of antibiotic resistance versus minimum temperature for *K. pneumoniae* and trimethoprim-sulfamethoxazole. Unadjusted weighted linear trend line is shown in blue. (D) Slope of unadjusted relationship (% Resistance/°C) between minimum temperature and antibiotic resistance by antibiotic for *K. pneumoniae*. Antibiotic class coded by color shading.

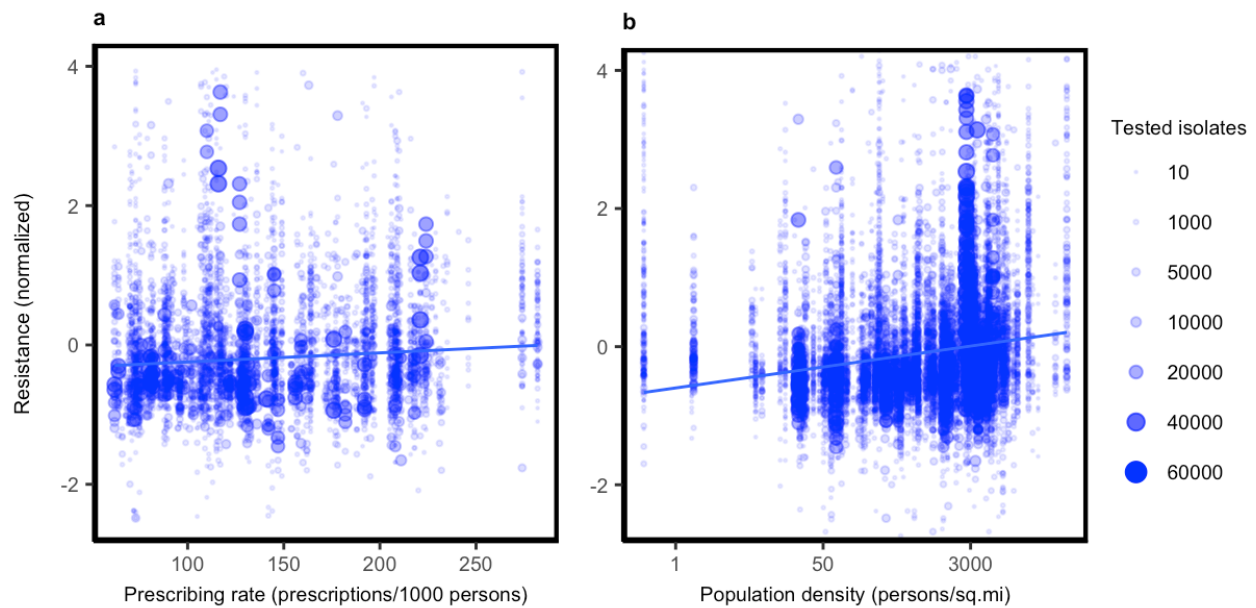


Supplementary Figure 4 | Antibiotic resistance and minimum temperature (*S. aureus*). (A)

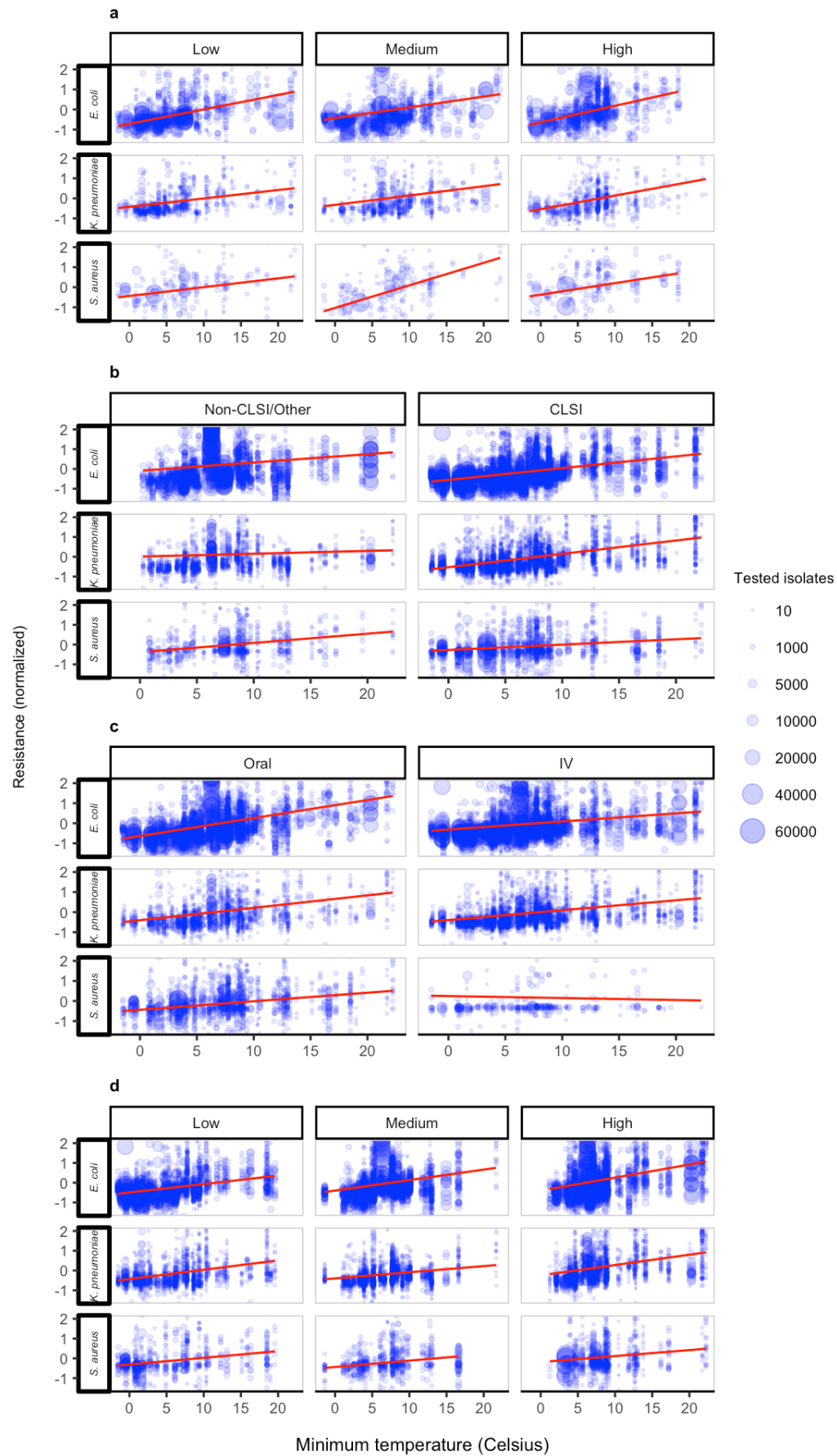
A heatmap of mean normalized antibiotic resistance for *S. aureus* for all antibiotics across the United States. (B) A heatmap of 30-year average minimum temperature (°C) across the United States. (C) A scatter plot of antibiotic resistance versus minimum temperature for *S. aureus* and cloxacillin. Unadjusted weighted linear trend line is shown in blue. (D) Slope of unadjusted relationship (% Resistance/°C) between minimum temperature and antibiotic resistance by antibiotic for *S. aureus*. Antibiotic class coded by color shading.



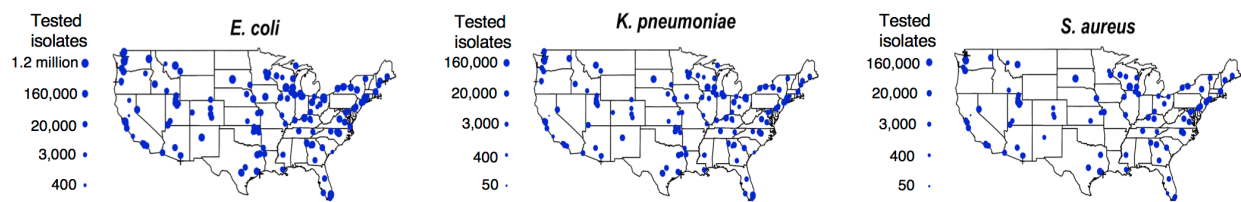
Supplementary Figure 5 | Antibiotic specific association between minimum temperature and antibiotic resistance versus approximate antibiotic introduction time in the United States¹.



Supplementary Figure 6 | Additional important predictors of antibiotic resistance, population density and prescribing rate. Scatter plots of the association between mean normalized antibiotic resistance with (A) annual prescription rate (prescriptions per 1000 persons); and (B) population density (persons/mi²), in *E. coli*, *K. pneumoniae*, and *S. aureus*. Data point size corresponds to number of isolates. Unadjusted weighted linear trend lines are shown in blue.



Supplementary Figure 7 | Evaluating relationship of minimum temperature and antibiotic resistance across other covariates. Scatter plots of mean normalized antibiotic resistance (%) versus minimum temperature (°C) by: (A) tertile of prescription rate (annual prescriptions/1000 persons); (B) reported laboratory standard (CLSI vs. other/not reported); (C) oral/IV formulation; and (D) tertile of population density (persons/mi²). Unadjusted weighted linear trend lines are shown in red.



Supplementary Figure 8 | Distribution of antibiotic resistance data across the United

States. A map of the United States showing locations of antibiotic resistance indices used as data points in this analysis, as a function of number of isolate tests (data point size), by pathogen (A) *E. coli*, (B) *K. pneumoniae*, and (C) *S. aureus*.

Supplementary Table 1 | Antibiotic and organism specific adjusted estimates of association between minimum temperature and antibiotic resistance.

Bacteria	Drug	Minimum temp	P-Value
<i>E. coli</i>	Amoxicillin	0.98	<0.0001
	Amoxicillin-Clavulanic acid	0.31	0.08
	Ampicillin-Sulbactam	0.64	<0.0001
	Cefepime	0.06	0.22
	Ceftazidime	0.26	0.0005
	Ceftriaxone	0.15	0.13
	Cefuroxime	0.19	0.40
	Cephalexin	0.61	<0.0001
	Ciprofloxacin	0.56	0.12
	Levofloxacin	0.67	0.0018
	Piperacillin-Tazobactam	0.14	<0.0001
<i>K. pneumoniae</i>	Amoxicillin-Clavulanic acid	-0.12	0.11
	Ampicillin-Sulbactam	0.36	<0.0001
	Cefepime	0.04	0.17
	Ceftazidime	0.45	<0.0001
	Ceftriaxone	0.19	<0.0001
	Cefuroxime	0.02	0.83
	Cephalexin	0.52	<0.0001
	Ciprofloxacin	0.11	0.05
	Levofloxacin	0.28	<0.0001
	Piperacillin-Tazobactam	0.23	<0.0001
<i>S. aureus</i>	Cephalexin	0.51	0.10
	Ciprofloxacin	0.29	0.0001
	Cloxacillin	0.58	<0.0001
	Erythromycin	0.54	<0.0001
	Levofloxacin	0.39	0.18
	Moxifloxacin	0.89	0.16
	Nafcillin	0.84	0.10
	Penicillin	-0.07	0.87

Sub-analysis of selected effect estimates and p-values for the association between minimum temperature and antibiotic resistance from full adjusted multivariable models restricted to specific antibiotic susceptibilities (for those antibiotics with available prescribing data). Individual models allow for different effects (modification) by antibiotic type and organism, but are subject to reduced sample size/power.

Supplementary Table 2 | Model predictor variable correlations.

Correlation Matrix						
<i>E. coli</i>						
	T min	Prescription rate	Outpatient	Lab standard	Population density	Median income
T min	1.000					
Prescription rate	0.008	1.000				
Outpatient	-0.123	0.042	1.000			
Lab standard	-0.048	0.009	-0.025	1.000		
Population density	0.237	-0.096	0.040	-0.060	1.000	
Median income	0.081	-0.017	0.028	-0.086	0.443	1.000
<i>Klebsiella</i>						
	T min	Prescription rate	Outpatient	Lab standard	Population density	Median income
T min	1.000					
Prescription rate	0.005	1.000				
Outpatient	-0.119	0.042	1.000			
Lab standard	-0.033	0.015	-0.014	1.000		
Population density	0.224	-0.116	0.047	-0.057	1.000	
Median income	0.073	-0.033	0.046	-0.095	0.451	1.000
<i>S. aureus</i>						
	T min	Prescription rate	Outpatient	Lab standard	Population density	Median income
T min	1.000					
Prescription rate	0.029	1.000				
Outpatient	-0.226	0.018	1.000			
Lab standard	-0.105	0.036	-0.070	1.000		
Population density	0.188	-0.072	0.061	-0.092	1.000	
Median income	0.046	-0.094	-0.061	-0.156	0.500	1.000
Variance inflation factor (VIF)						
Bacteria	T min	Prescription rate	Outpatient	Lab standard	Population density	Median income
<i>E. coli</i>	1.217	1.012	1.020	1.114	1.273	1.239
<i>Klebsiella</i>	1.195	1.011	1.029	1.108	1.392	1.422
<i>S. aureus</i>	1.274	1.024	1.124	1.253	1.473	1.450

Supplementary Table 3 | IV and Oral antibiotic classification.

<i>E. coli</i>		<i>K. pneumoniae</i>		<i>S. aureus</i>	
Amoxicillin	Oral	Amoxicillin-Clavulanic acid	Oral	Cephalexin	Oral
Amoxicillin-Clavulanic acid	Oral	Cefuroxime	Oral	Clindamycin	Oral
Cefuroxime	Oral	Cephalexin	Oral	Cloxacillin	Oral
Cephalexin	Oral	Ciprofloxacin	Oral	Erythromycin	Oral
Ciprofloxacin	Oral	Levofloxacin	Oral	Levofloxacin	Oral
Levofloxacin	Oral	Tetracycline/Doxycycline	Oral	Linezolid	Oral
Nitrofurantoin	Oral	Trimethoprim-Sulfamethoxazole	Oral	Moxifloxacin	Oral
Tetracycline/Doxycycline	Oral	Amikacin	IV	Penicillin	Oral
Trimethoprim-Sulfamethoxazole	Oral	Ampicillin-Sulbactam	IV	Rifampin	Oral
Amikacin	IV	Aztreonam	IV	Tetracycline/Doxycycline	Oral
Ampicillin-Sulbactam	IV	Cefepime	IV	Trimethoprim-Sulfamethoxazole	Oral
Aztreonam	IV	Ceftazidime	IV	Daptomycin	IV
Cefepime	IV	Ceftriaxone	IV	Nafcillin	IV
Ceftazidime	IV	Ertapenem	IV	Quinupristin/Dalfupristin	IV
Ceftriaxone	IV	Gentamicin	IV	Vancomycin	IV
Ertapenem	IV	Imipenem	IV		
Gentamicin	IV	Meropenem	IV		
Imipenem	IV	Piperacillin-Tazobactam	IV		
Meropenem	IV	Tobramycin	IV		
Piperacillin-Tazobactam	IV				
Tobramycin	IV				

Supplementary Table 4 | Additional sensitivity analyses.

Bacteria	Variable	Sensitivity A	Sensitivity B	Sensitivity C	Sensitivity D	Sensitivity E	Sensitivity F
<i>E. coli</i>	Minimum temp	0.44 (<i><0.0001</i>)	0.47 (<i><0.0001</i>)	0.34 (<i><0.0001</i>)	0.41 (<i><0.0001</i>)	0.38 (<i><0.0001</i>)	
	Prescription rate	0.12 (<i><0.0001</i>)	0.11 (<i><0.0001</i>)	0.11 (<i><0.0001</i>)	0.12 (<i><0.0001</i>)	0.12 (<i><0.0001</i>)	
	Outpatient	-2.99 (0.09)	-3.30 (<i><0.001</i>)	-4.23 (0.04)	-2.98 (0.07)	-3.08 (0.07)	
	Lab standard	-3.82 (0.09)	-2.43 (0.18)	-5.49 (0.07)	-3.20 (0.14)	-3.13 (0.16)	
	Population density	0.00028 (0.13)	0.00030 (0.06)	0.00020 (0.20)	--	0.00015 (0.28)	
	Median income		--		0.00029 (<i><0.001</i>)	0.00025 (0.001)	
	Blood		-3.93 (0.04)				
	Respiratory		2.95 (0.29)				
	Urine		-1.15 (0.28)				
	Other sterile		11.91 (<i><0.001</i>)				
	Other nonsterile		-14.29 (<i><0.0001</i>)				
<i>Klebsiella</i>	Minimum temp	0.23 (<i><0.001</i>)	0.22 (<i><0.0001</i>)	0.29 (<i><0.0001</i>)	0.28 (<i><0.0001</i>)	0.21 (<i><0.0001</i>)	
	Prescription rate	0.04 (<i><0.0001</i>)	0.03 (<i><0.0001</i>)	0.02 (<i><0.0001</i>)	0.03 (<i><0.0001</i>)	0.03 (<i><0.0001</i>)	
	Outpatient	-2.68 (0.20)	-2.75 (0.13)	-4.00 (0.15)	-2.55 (0.19)	-2.91 (0.15)	
	Lab standard	-0.40 (0.76)	-0.38 (0.66)	-0.43 (0.74)	-0.71 (0.65)	-0.34 (0.76)	
	Population density	0.00062 (<i><0.0001</i>)	0.00060 (<i><0.0001</i>)	0.00064 (<i><0.0001</i>)	--	0.00058 (<i><0.0001</i>)	
	Median income		--		0.00028 (0.16)	0.00004 (0.79)	
	Blood		-0.53 (0.65)				
	Respiratory		1.27 (0.37)				
	Urine		-1.86 (0.27)				
	Other sterile		-10.64 (<i><0.0001</i>)				
	Other nonsterile		10.35 (<i><0.0001</i>)				
<i>S. aureus</i>	Minimum temp	0.68 (0.002)	0.19 (<i><0.0001</i>)	0.39 (0.14)	0.28 (0.18)	0.28 (0.18)	0.53 (0.002)
	Prescription rate	0.12 (<i><0.0001</i>)	0.13 (<i><0.0001</i>)	0.10 (<i><0.001</i>)	0.13 (<i><0.0001</i>)	0.13 (<i><0.0001</i>)	0.17 (<i><0.0001</i>)
	Outpatient	-7.93 (<i><0.0001</i>)	-4.47 (<i><0.001</i>)	-0.18 (0.89)	-3.92 (0.006)	-3.86 (0.008)	-6.22 (<i><0.0001</i>)
	Lab standard	-1.38 (0.55)	-3.89 (0.06)	-9.50 (0.05)	-3.62 (0.03)	-3.65 (0.05)	-2.69 (0.08)
	Population density	0.00006 (0.14)	0.00000 (0.97)	-0.00013 (0.44)	--	-0.00003 (0.84)	0.00014 (0.15)
	Median income		--		0.00010 (0.38)	0.00020 (0.33)	
	Blood		-4.24 (<i><0.001</i>)				
	Respiratory		2.78 0.22				
	Urine		8.59 (<i><0.001</i>)				
	Other sterile		-17.63 (<i><0.0001</i>)				
	Other nonsterile		19.80 (<i><0.0001</i>)				

(A) restriction of the full adjusted model to only health systems, hospitals, or laboratories (excluding surveillance bodies); (B) full adjusted model with restriction of the dataset to only locations with multiple years represented within the database; (C) full adjusted model with addition of binary predictors for isolate types; (D) full adjusted model with substitution of population density variable with median income; (E) full adjusted model with addition of median income; and (F) full adjusted model for *S. aureus* with cephalexin and nafcillin excluded. P-values are shown in parentheses (). All analyses were restricted to subset of antibiotic susceptibilities for dates and classes of antibiotics corresponding to available prescribing rate data.

Supplementary References

1. IUPAC, Fischer, J. & Robin Ganellin, C. *Analogue-based Drug Discovery*. (John Wiley & Sons, 2006).