Clinical Medicine & Research
Volume 20, Number 4: 185-194

clinmedres.org

Original Research

Changes in *Streptococcus pneumoniae* Susceptibility in Wisconsin: Implications for Clinical Treatment Decisions for Respiratory Infections

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Objective: In 2019, the American Thoracic Society and Infectious Diseases Society of America updated clinical practice guidelines for community-acquired pneumonia (CAP). In contrast to guidelines published in 2007, macrolide monotherapy for outpatients was made a conditional recommendation based on resistance levels. Local knowledge of current antimicrobial susceptibility is needed to guide management of CAP and other bacterial respiratory pathogens. The purpose of this study was to investigate antimicrobial susceptibility profiles and trending for Wisconsin *Streptococcus pneumoniae* isolates.

Design: Multi-center laboratory surveillance, with testing at a central location utilizing standardized susceptibility testing protocols.

Methods: Data published by the Wisconsin Department of Health Services (DHS) were augmented with data from the Surveillance of Wisconsin Organisms for Trends in Antimicrobial Resistance and Epidemiology (SWOTARE) program. Data were stratified by invasive or non-invasive sources, as well as DHS region and compared to data compiled from 2006-2010.

Results: Susceptibility rates for ≥ 916 invasive *S. pneumoniae* assessed from 2016-2020 were greater than 91% for ceftriaxone, tetracycline, and fluoroquinolone agents and were generally higher than those from 354 non-invasive isolates. Low susceptibility rates were observed for invasive isolates of penicillin (78.7%) and erythromycin (64.8%) and were even lower for non-invasive isolates (73.8% and 59.9%, respectively). This erythromycin susceptibility rate was a significant reduction from that observed in 2006-2010 (80.4; $P < 0.0002$). 24.8% of isolates generated an erythromycin MIC ≥ 8 µg/mL. Statewide geographic variability was noted.

Conclusions: Rates of *S. pneumoniae* susceptibility to parenteral penicillins and cephems, and oral tetracycline and fluoroquinolone agents, remain high throughout Wisconsin. However, low oral penicillin susceptibility rates, taken together with declining macrolide susceptibility rates, should cause clinicians to consider alternative treatment options for respiratory tract infections, especially with macrolides.

Keywords: *Streptococcus pneumoniae*; macrolides; doxycycline; SWOTARE

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Received: May 18, 2022
Revised: July 27, 2022
Accepted: August 9, 2022

doi:10.3121/cmr.2022.1767

Treatment of outpatient community-acquired pneumonia (CAP) has undergone transition over the past 15 years. In 2007, the Infectious Diseases Society of America/ American Thoracic Society (IDSA/ATS) published guidelines for therapy.[1] One of the more challenging aspects of the management of CAP has been age groups or medical conditions where a macrolide antibiotic was recommended as a sole agent for treatment. In 2007, in previously healthy individuals with no risk factors for drug-resistant *Streptococcus pneumoniae* (DRSP) infections, macrolide monotherapy was strongly recommended over doxycycline.[1] In regions with a high rate of infection (>25%) with "high-level" (minimum inhibitory concentration (MIC) of ≥ 16 μg/mL) macrolide-resistant *S. pneumoniae*, use of alternative agents (i.e., quinolone, or macrolide plus β-lactam) needed to be considered. Other experts chose a more conservative erythromycin breakpoint for resistance (≥ 8 μg/mL) as being of more clinical relevance.[2-4] With the IDSA/ATS revision in 2019, the consensus was modified to include "high-dose" oral amoxicillin, and placed doxycycline and macrolides on an equivalent status. However, experts continued to caution that in geographic areas with high prevalence of macrolide-resistant *S. pneumoniae* (>25%), other alternatives should be used if monotherapy is considered.[5] Most clinicians are not aware of differentiating low- and high-level resistance among macrolide agents, and the difficulty with this advice for clinicians is knowing what the actual rates of resistance are and how to apply this resistance information to their prescribing habits.

In addition to CAP, *S. pneumoniae* is also the most prevalent bacterium causing both otitis media in children and acute bacterial sinusitis in children and adults. There is no reason to believe that community isolate susceptibility should be appreciably different for these latter conditions compared to CAP. Hence, prescribing for these conditions is also affected by the local rates of resistance. Rates of antimicrobial resistance based on Clinical and Laboratory Standards Institute (CLSI) criteria for achievable oral penicillin/amoxicillin levels have exceeded the 25% threshold for many years, prompting the recommendation in pediatrics to use "high-dose" amoxicillin when necessary for acute otitis media.[6] For acute bacterial sinusitis, amoxicillin-clavulanate or amoxicillin is recommended when antibiotics are necessary in adults and children.[7] Furthermore, "high-dose" amoxicillin-clavulanate and amoxicillin is recommended for children and adults from geographic regions with high endemic rates (>10%) of invasive penicillin-non-susceptible *S. pneumoniae*.[7]

Penicillin and cephem agents have alternative interpretive criteria, as defined by CLSI, for parenteral/non-cerebrospinal fluid (non-CSF) serum and tissue levels of antibiotic that have generally continued to maintain clinical efficacy,[8,9] in spite of a trend towards decreased susceptibility. The final category of CLSI interpretive criteria for penicillin and cephem agents is for cerebrospinal fluid (CSF) infections. Following trends for CSF penicillin- and cephem-resistant *S. pneumoniae* has implications for treatment of suspected bacterial meningitis. Practically speaking though, the severity of the condition and need to have at least one effective antibiotic from the start has led to the use of vancomycin with the empiric regimen for coverage of possible DRSP infection.

We examined trends in antimicrobial resistance to *S. pneumoniae* in the state of Wisconsin for two time windows, 2006-2010 and 2016-2020. We also examined differences in susceptibility between invasive and non-invasive isolates for the latter period, and present recommendations for treatment of outpatient bacterial infections, especially CAP, based on these trends.

Materials and Methods

SWOTARE Data Collection

The Surveillance of Wisconsin Organisms for Trends in Antimicrobial Resistance and Epidemiology (SWOTARE) program is a Wisconsin antimicrobial resistance surveillance initiative operated out of Marquette University in prospective collaboration with nearly two dozen clinical microbiology laboratories. In general, SWOTARE attempts to focus study site recruitment efforts toward non-tertiary care facilities in order to provide a representative profile of Wisconsin antimicrobial resistance patterns.[10-13] On an annual basis, study sites were requested to forward clinically-significant isolates of *S. pneumoniae* to a centralized testing laboratory. Cerebrospinal fluid and bloodstream isolates were categorized as invasive isolates, while isolates from other anatomic sources were categorized as non-invasive isolates. To limit potential of bias during the collection process, isolates were collected in consecutive fashion and any duplicate or nonviable isolates were excluded from the study. Because of the lack of direct involvement in the collection of specimens and because of the utilization of de-identified isolates from routine clinical care, the SWOTARE program is not considered to be actively engaged in human research subjects research by the Marquette University Institutional Review Board.

Reference broth microdilution antimicrobial susceptibility testing was executed[14] using standards published by CLSI and interpreted using pertinent documents in the CLSI M100 series.[15] In brief, submitted isolates of *S. pneumoniae* were used to inoculate customized panels of cation-adjusted Mueller-Hinton broth supplemented with lysed horse blood. Panels were designed with antimicrobial agent dilution ranges that extended beyond individual CLSI breakpoints. Percentage susceptible, intermediate, and resistant values were provided to participating laboratories on a statewide and geographic basis for all agents tested.

Specialized MIC Considerations

Ceftriaxone MIC data have been interpreted using parenteral meningeal and non-meningeal CLSI interpretive criteria since 2003.[16] Penicillin susceptibility data have been interpreted using oral and parenteral non-meningeal CLSI interpretive criteria since 2008.[17] MIC breakpoints for susceptibility within these two criteria are ≤ 0.06 μg/mL and ≤ 2 μg/mL, respectively. A third

CLSI interpretive criterion, demarcated as parenteral meningeal, has also been published since 2008.[17] More stringent MIC breakpoints for susceptibility within the parenteral meningeal and oral criteria are equivalent (≤ 0.06 µg/mL).

Interpretive criteria for erythromycin broth microdilution susceptibility testing reflect breakpoints of ≤ 0.25 µg/mL (susceptible), 0.5 µg/mL (intermediate), and ≥ 1 µg/mL (resistant).[15] Within this reference, CLSI further states that susceptibility and resistance to azithromycin and clarithromycin can be predicted by testing erythromycin. To assess the potential for high-level erythromycin resistance in Wisconsin *S. pneumoniae* isolates, an MIC endpoint (≥ 8 µg/mL) commensurate with that described by Lynch and Martinez,[2] Jacobs,[3] and Pérez-Trallero[4] was selected. MIC data reflective of this endpoint were available for all non-invasive isolates and a subset of invasive isolates.

Data Collection by Wisconsin Department of Health Services (DHS)

S. pneumoniae isolates derived from an invasive source were submitted to the Wisconsin State Laboratory of Hygiene (WSLH) through DHS. Broth microdilution antimicrobial susceptibility testing was performed on a subset of these isolates using methods previously described.[14,15] Prior to 2017, WSLH performed antimicrobial susceptibility testing on all submitted isolates, later switching to performing surveillance on 20% of submitted isolates.

Data Compilation and Analysis

Two five-year intervals of *S. pneumoniae* isolate testing were investigated on the basis of data availability: 2006-2010 and 2016-2020. Antimicrobial agents assessed within this study included penicillin, ceftriaxone, erythromycin, tetracycline class representatives, and fluoroquinolone class agents. Compiled DHS data from invasive *S. pneumoniae* isolates tested in 2016-2020 were supplemented with compiled SWOTARE data from the same interval for all antimicrobial agent groupings. Compiled SWOTARE data relative to non-invasive *S. pneumoniae* isolates collected from 2016-2020 were available for an initial comparison to the combined DHS-SWOTARE 2016-2020 dataset for invasive *S. pneumoniae* isolates using all antimicrobial agent groupings.

The 2016-2020 data compilations were further stratified in regional fashion. Five DHS regions (Figure 1) that are typically

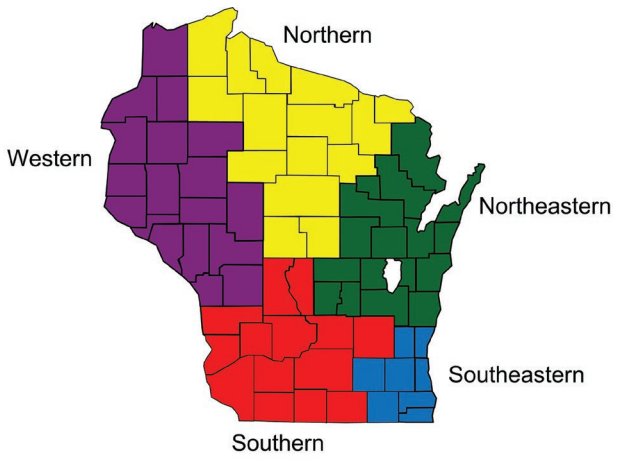

Figure 1. Distribution of five Wisconsin Department of Health Services geographic regions.

Table 1: Antimicrobial susceptibility profiles for *Streptococcus pneumoniae* non-invasive and invasive isolates, Wisconsin 2016-2020.

	Non-invasive		Invasive	
Antimicrobial Agent	**n**	**Percentage Susceptible**	**n**	**Percentage Susceptible**
Penicillin oral/CSF[a]	354	73.7	1070	78.7[b]
Penicillin non-CSF[a]	354	97.5	1020	99.4[c]
Ceftriaxone CSF[d]	354	93.8	1070	93.0
Ceftriaxone non-CSF[d]	354	97.7	1070	99.1[b]
Erythromycin	354	59.9	1070	64.8
Tetracycline	354	86.2[e]	916	91.4[c]
Levofloxacin	354	98.9	1070	99.0
Moxifloxacin	354	99.2	237	99.6

[a] Clinical and Laboratory Standards Institute has published oral, parenteral meningeal (CSF; each with a susceptibility breakpoint of 0.06 µg/mL), and parenteral non-meningeal (non-CSF) interpretive criteria for *Streptococcus pneumoniae* penicillin broth microdilution MIC data since 2008.

[b] P = 0.053 versus non-invasive isolates

[c] P ≤ 0.006 versus non-invasive isolates

[d] Clinical and Laboratory Standards Institute has published parenteral meningeal (CSF) and parenteral non-meningeal (non-CSF) interpretive criteria for *S. pneumoniae* ceftriaxone broth microdilution MIC data since 2003.

[e] Invasive *S. pneumoniae* doxycycline broth microdilution susceptibility data generated from 2018-2020 revealed 92.6% susceptibility to the agent. Data were not available for non-invasive isolates.

utilized for communication and data analysis within the agency served as the basis for geographic comparison within this study. The significance test of proportions determined if differences in percentage susceptibility were significant. The alpha level was set at 0.05 before the investigations commenced, and all *P* values are two-tailed.

DHS data from invasive *S. pneumoniae* isolates were also available from 2006-2010 for a temporal comparison of penicillin, ceftriaxone, erythromycin, and tetracycline susceptibility between that compilation and the combined DHS-SWOTARE dataset from 2016-2020. Within the earlier dataset, penicillin susceptibility data were analyzed only from 2008-2010, commensurate with CLSI interpretive criteria revisions of 2008.[17] Ceftriaxone susceptibility data interpreted by CLSI meningeal and non-meningeal criteria were only available from 2007-2010.[16] Penicillin and ceftriaxone interpretive criteria and breakpoints remained unchanged throughout the 2016-2020 testing interval. Moxifloxacin susceptibility data were generated only by SWOTARE; doxycycline data were only available through the DHS database.

As an additional means of characterizing geographic variation, the statewide mean susceptibility percentage for a given organism/antimicrobial combination established a baseline value. An interval of 5% on either side of that mean represented normal distribution. Region-specific values ≥ 5% less than the state mean indicated areas with increased resistance. Region-specific values ≥ 5% greater than the state mean indicated less resistance potential. These intervals were utilized in previous SWOTARE reports.[10-13,18]

Results

Contemporary Comparison of Antimicrobial Susceptibility Profiles from Invasive and Non-Invasive Streptococcus pneumoniae Isolates

SWOTARE assessed 354 non-invasive isolates collected from 2016 to 2020. Non-invasive isolates were largely derived from lower respiratory (66.9%) and upper respiratory (27.7%) sources. Data generated from these isolates were compared to a compiled invasive isolate dataset combining data from DHS with SWOTARE.

In general, non-invasive *S. pneumoniae* exhibited less susceptibility to penicillin, erythromycin (when using CLSI-published breakpoints), and tetracycline when compared to invasive isolates (Table 1). Differences in penicillin susceptibility between non-invasive and invasive isolates (73.7% versus 78.7%, respectively) relative to CLSI oral interpretive criteria trended toward significance ($P = 0.053$; Table 1), as did comparisons involving CLSI-published breakpoints for erythromycin (59.9% non-invasive, 64.8% invasive; $P = 0.10$). Differences in tetracycline susceptibility (86.2% non-invasive; 91.4% invasive) were significant ($P = 0.006$). Susceptibility differences between the datasets for two fluoroquinolone agents were nominal.

Differences in penicillin susceptibility between non-invasive and invasive isolates relative to CLSI parenteral non-meningeal interpretive criteria (97.5% and 99.4%, respectively; Table 1) were significant ($P = 0.002$). Trends toward diminished susceptibility included ceftriaxone using

Table 2: Geographic distribution of antimicrobial susceptibility profiles for *Streptococcus pneumoniae* non-invasive and invasive isolates, Wisconsin 2016-2020 (continued on page 189)

	Northern				Northeastern				Southern			
	Non-invasive		Invasive		Non-invasive		Invasive		Non-invasive		Invasive	
Antimicrobial Agent	n	% S[a]	n	% S[a]	n	% S[a]	n	% S[a]	n	% S[a]	n	% S[a]
Penicillin oral/CSF[b]	63	76.2	181	79.0	119	74.8	208	81.7	57	77.2	158	82.3
Penicillin non-CSF[b]	63	100.0	175	98.9	119	95.8	199	99.5	57	96.5	152	100.0
Ceftriaxone CSF[c]	63	95.2	181	95.6	119	94.1	208	92.3	57	91.2	158	94.3
Ceftriaxone non-CSF[c]	63	100.0	181	98.4	119	97.5	208	98.6	57	94.7	158	100.0
Erythromycin	63	61.9	181	68.5	119	58.8	208	61.1	57	57.9	158	59.5
Tetracycline[d]	63	85.7	152	91.4	119	86.6	184	90.8	57	78.9	126	92.1
Levofloxacin	63	98.4	181	99.4	119	99.2	208	99.0	57	100.0	158	100.0
Moxifloxacin	63	98.4	58	100.0	119	100.0	69	100.0	57	100.0	28	100.0

[a] Percentage susceptible

[b] Clinical and Laboratory Standards Institute has published oral, parenteral meningeal (CSF; each with a susceptibility breakpoint of 0.06 µg/mL), and parenteral non-meningeal (non-CSF) interpretive criteria for *Streptococcus pneumoniae* penicillin broth microdilution MIC data since 2008.

[c] Clinical and Laboratory Standards Institute has published parenteral meningeal (CSF) and parenteral non-meningeal (non-CSF) interpretive criteria for *S. pneumoniae* ceftriaxone broth microdilution MIC data since 2003.

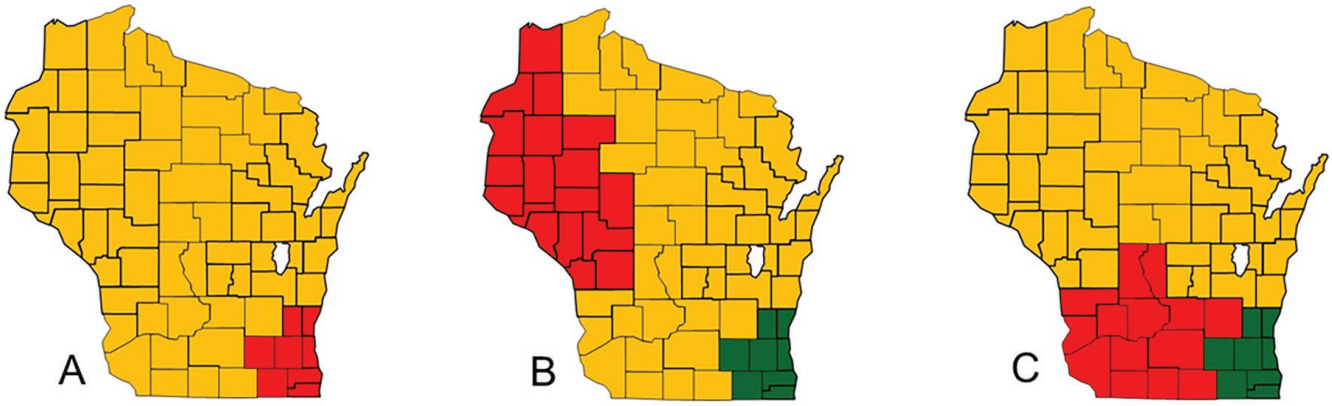

Figure 2. Geographic variation of *Streptococcus pneumoniae* non-invasive isolates with respect to penicillin (CLSI oral/parenteral meningeal interpretive criteria; A), erythromycin (CLSI-published breakpoints; B), and tetracycline (C) susceptibility relative to statewide mean, Wisconsin 2016-2020. Regions colored gold represent percentage susceptible rates ±5% of the Wisconsin mean for the antimicrobial agent. Regions colored red represent percentage susceptible rates ≥ 5% less than the state mean for the antimicrobial agent. Regions colored green represent percentage susceptible rates ≥ 5% greater than the state rate for the antimicrobial agent.

CLSI parenteral non-meningeal criteria (97.7% non-invasive, 99.1% invasive; $P = 0.053$).

Data regarding oral penicillin susceptibility extrapolate to the CLSI penicillin parenteral meningeal interpretive criterion due to the shared breakpoint for susceptibility. Ceftriaxone susceptibility rates did not show any appreciable differences between invasive and non-invasive isolates when CLSI parenteral meningeal criteria were applied ($P = 0.61$).

Table 2: (continued from page 188)

Southeastern				Western			
Non-invasive		Invasive		Non-invasive		Invasive	
n	% S[a]	n	% S[a]	n	% S[a]	n	% S[a]
65	67.7	382	75.7	50	72.0	141	78.0
65	100.0	358	99.2	50	96.0	136	100.0
65	93.8	382	90.8	50	94.0	141	95.0
65	98.5	382	99.0	50	98.0	141	100.0
65	66.2	382	66.2	50	54.0	141	67.4
65	92.3	334	92.5	50	86.0	120	88.3
65	100.0	382	98.4	50	96.0	141	98.6
65	100.0	51	98.0	50	96.0	31	100.0

[d] Doxycycline percentage susceptibility rates also determined for invasive isolates only. 95.2% in Northern region (n = 83 isolates tested); 95.5% in Northeastern region (n = 66); 94.8% in Southern region (n = 58); 87.7% in Southeastern region (n = 155); 96.6% in Western region (n = 59).

Geographic variation in non-invasive Wisconsin S. pneumoniae isolates

DHS region profiles of fluoroquinolone and ceftriaxone susceptibility within non-invasive *S. pneumoniae* exhibited little variability (Table 2). However, an approximate 10% difference was observed in penicillin susceptibility between Southern (77.2%) and Southeastern (67.7%) region isolates. A 12.2% difference in erythromycin susceptibility (when using CLSI-published breakpoints) was noted upon testing Southeastern and Western region isolates; a decreased rate of erythromycin susceptibility (57.9%) was also noted among Southern region isolates. Similarly, decreased tetracycline susceptibility (78.9%) was observed in Southern region isolates when compared to Southeastern region isolates.

When region-specific invasive *S. pneumoniae* susceptibility rates were compared to the Wisconsin state mean, differences in penicillin, tetracycline, and erythromycin (using CLSI-published breakpoints) susceptibility were noted (Figure 2). The Southeastern region trended toward increased erythromycin and tetracycline susceptibility when compared to other regions of Wisconsin. Western and Southern regions were more likely to exhibit decreased tetracycline or erythromycin susceptibility among non-invasive isolates.

Geographic Variation in Contemporary Invasive Wisconsin S. pneumoniae Isolates

The 2016-2020 invasive *S. pneumoniae* dataset was then stratified by DHS region. Inter-region comparisons (Table 2)

Table 3: Assessment and geographic distribution of high-level erythromycin resistance (defined by minimum inhibitory concentration ≥ 8 µg/mL) within subsets of non-invasive and invasive *Streptococcus pneumoniae* isolates, Wisconsin 2016-2020

	Northern		Northeastern		Southern		Southeastern		Western	
***Streptococcus pneumoniae* Isolate**	**n**	**Percentage ≥ 8 µg/mL**	**n**	**Percentage ≥ 8 µg/mL**	**n**	**Percentage ≥ 8 µg/mL**	**n**	**Percentage ≥ 8 µg/mL**	**n**	**Percentage ≥ 8 µg/mL**
Non-invasive	63	23.8	119	27.7	57	33.3	65	21.5	50	26.0
Invasive	141	20.6	135	23.7	86	24.4	206	26.2	89	23.6

mirrored those from non-invasive comparisons relative to penicillin (Southeastern and Southern regions) and CLSI-advocated erythromycin breakpoints (Southern region demonstrating lowest susceptibility rates, particularly when compared to Northern and Western region isolates). Moreover, when regional erythromycin susceptibility distribution was compared to the Wisconsin mean, a decrease was additionally noted in the Southern region (Figure 3).

Nominal geographic variation was observed with fluoroquinolone agents. Geographic differences in tetracycline susceptibility were of less magnitude with invasive *S. pneumoniae* when compared to non-invasive isolates. Analogous to the observation of decreased penicillin susceptibility in non-invasive *S. pneumoniae*, invasive isolates from the Southeastern region also exhibited lowest rates of ceftriaxone susceptibility when CLSI meningeal interpretive criteria were applied to MIC data (Table 2).

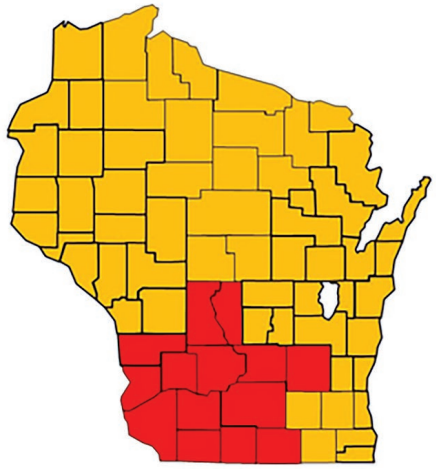

Figure 3. Geographic variation of *Streptococcus pneumoniae* invasive isolates with respect to erythromycin (CLSI-published breakpoints) susceptibility relative to statewide mean, Wisconsin 2016-2020. Regions colored gold represent percentage susceptible rates ±5% of the Wisconsin mean. Regions colored red represent percentage susceptible rates ≥ 5% less than the state mean.

Assessment of High-Level Erythromycin Resistance

When an MIC endpoint of ≥ 8 µg/mL was used to assess high-level erythromycin resistance, 26.6% and 23.9% of Wisconsin non-invasive and invasive *S. pneumoniae* isolates, respectively, demonstrated high-level resistance. The proportion of high-level resistant non-invasive isolates was higher in the Southern region and less in the Southeastern region (Table 3). Southeastern region findings were consistent with overall increased erythromycin susceptibility for non-invasive isolates within that region (Table 2; Figure 2B). Distribution of high-level erythromycin resistance within invasive isolates was consistent throughout Wisconsin, ranging from 20.6% in the Northern region to 26.2% in the Southeastern region ($P = 0.23$; Table 3).

Temporal Trends in Susceptibility among Invasive Streptococcus Pneumoniae

Antimicrobial susceptibility against four agents was compared using compiled data from 2006-2010 and 2016-2020 to examine longitudinal trends across Wisconsin (Table 4). Penicillin and ceftriaxone susceptibility has slightly increased over the last decade, particularly when applying CLSI parenteral non-meningeal interpretive criteria to each agent ($P < 0.0002$). Conversely, erythromycin susceptibility (when using CLSI-published breakpoints) has decreased significantly over this interval, as currently 64.8% of invasive isolates of *S. pneumoniae* demonstrate susceptibility ($P < 0.0002$). Tetracycline susceptibility is largely unchanged and remains greater than 91% ($P = 0.88$).

Discussion

S. pneumoniae is the most important bacterial cause of CAP, as well as acute otitis media and acute sinusitis. Recommendations regarding clinical treatment of these conditions is dependent on the prevalence of susceptible/resistant bacteria, which often varies nationally and regionally. Guidelines infer that when a certain threshold of antibiotic resistance is crossed, choices of antimicrobial therapy should be modified accordingly.

Table 4: Comparison of antimicrobial susceptibility profiles for *Streptococcus pneumoniae* invasive isolates, Wisconsin 2006-2010 and 2016-2020

	Wisconsin, 2006-2010		Wisconsin, 2016-2020	
Antimicrobial Agent	**n**	**Percentage Susceptible**	**n**	**Percentage Susceptible**
Penicillin oral/CSF	1231	76.4[a]	1070	78.7
Penicillin non-CSF	1198	93.2[a]	1020	99.4[b]
Ceftriaxone CSF	1604	91.5[c]	1070	93.0
Ceftriaxone non-CSF	1612	96.2[c]	1070	99.1[b]
Erythromycin	1978	80.4	1070	64.8[b]
Tetracycline	1978	91.2	916	91.4[d]

[a] Data from 2008-2010; Clinical and Laboratory Standards Institute has published oral, parenteral meningeal (CSF; each with a susceptibility breakpoint of 0.06 μg/mL), and parenteral non-meningeal (non-CSF) interpretive criteria for *Streptococcus pneumoniae* penicillin broth microdilution MIC data since 2008.
[b] $P < 0.0002$ versus isolates from 2006-2010
[c] Data available from 2007-2010; Clinical and Laboratory Standards Institute publishes parenteral meningeal (CSF) and parenteral non-meningeal (non-CSF) interpretive criteria for *S. pneumoniae* ceftriaxone broth microdilution MIC data.
[d] Invasive *S. pneumoniae* doxycycline broth microdilution susceptibility data generated from 2018-2020 revealed 92.6% susceptibility to the agent

While fluoroquinolone susceptibility rates between invasive and non-invasive *S. pneumoniae* were stable, differences were noted for penicillin, erythromycin, and tetracycline. Among invasive isolates only, there was a surprising increase in *S. pneumoniae* susceptibility to penicillin and ceftriaxone between the two time points, a stable temporal trend for tetracycline, but a significant declining trend for erythromycin susceptibility. Susceptibility and resistance to azithromycin and clarithromycin can be predicted by testing erythromycin.[15]

The 2007 and 2019 IDSA/ATS CAP guidelines recommend that in regions with a high rate of infection (>25%) with "high-level" (MIC ≥ 16 μg/mL) macrolide-resistant *S. pneumoniae*, use of alternative agents needs to be considered.[1,5] This erythromycin MIC endpoint for clinical significance and modification of treatment recommendations has been the subject of ongoing medical debate.[2,3,19-21] Resistance to erythromycin has been divided into "high-level" and "low-level" resistance, based on different mechanisms of resistance, beyond consideration of most clinicians.[2-4] While the CAP guidelines choose an MIC ≥ 16 μg/mL as a cutoff for clinically meaningful high-level resistance, others have suggested that an MIC ≥ 8 μg/mL is more appropriate.[2-4] We chose to analyze trends with this more conservative cutoff (i.e., more isolates interpreted as high-level resistant to erythromycin). Several authors have already cautioned regarding the concerns of using macrolides for the treatment of CAP.[19-21]

The overall resistance rates to erythromycin in Wisconsin exceed the 25% threshold cited in the CAP guidelines using the ≥ 8 μg/mL cutoff. There is no reason to believe that similar rates of resistance do not exist for ear and sinus infections. For non-invasive isolates, using the more conservative ≥ 8 μg/mL MIC endpoint, further examination of data shows that 26.6% of all isolates exhibit high-level resistance. There were regional differences, with the Southeastern region having the lowest rate with 21.5%, and the Southern region the highest with 33.3%. When the invasive isolates were examined using the ≥ 8 μg/mL MIC endpoint, overall 23.9% of isolates were resistant. Regional differences were also seen, with the Northern region having the lowest rate of 20.6%, and the Southeastern with the highest rate of 26.2%.

Oral doxycycline has largely replaced tetracycline for clinical use. Rates of tetracycline susceptibility among invasive isolates have remained high over time at approximately 91%. Among invasive isolates tested against doxycycline between 2018-2020, 92.6% were susceptible. While in the most recent time window there were differences between invasive and non-invasive isolates in the percentage susceptibility to tetracycline (91.4% vs 86.2%, respectively), such differences are likely clinically insignificant. Doxycycline exhibits increased *in vitro* potency compared to tetracycline, relative to *S. pneumoniae*.[22] Hence, doxycycline appears to be a viable alternative oral antibiotic for the treatment of bacterial respiratory infections.

While we observed several important findings with respect to temporal and geographic variation within *S. pneumoniae* susceptibility data, our findings should be viewed in the context of their limitations. First, analysis of *S. pneumoniae* penicillin data from the 2006-2010 interval was restricted to the years 2008, 2009, and 2010 to allow for a more direct comparison to the 2016-2020 interval with respect to the introduction of oral and non-meningeal interpretive criteria by CLSI in 2008. Evaluations of ceftriaxone MIC data through

CLSI parenteral meningeal and non-meningeal interpretive criteria were only available from 2007 through 2010. It is possible that penicillin and/or ceftriaxone susceptibility data from 2006 and/or 2007 could have altered overall interpretations of temporal comparisons. Secondly, non-invasive data were not available from DHS for both the 2006-2010 and 2016-2020 intervals, as this entity surveilled only invasive isolates of *S. pneumoniae*.

Additional limitations pertained to penicillin susceptibility testing. While both DHS and SWOTARE penicillin MIC data were derived from reference broth microdilution testing, dilution ranges of the agent differed between the two laboratories. Penicillin concentrations within the DHS assay extended to 1 μg/mL, yet the CLSI parenteral non-meningeal breakpoint for susceptibility is set at ≤ 2 μg/mL.[15] Fifty isolates had to be excluded from the presentations in Tables 1 and 4 due to a final MIC reading of >1 μg/mL. It is unclear whether these isolates were either susceptible with an MIC of 2 μg/mL or exhibited decreased susceptibility with higher MIC values. While the exact data within Tables 1 and 4 may be altered incrementally, the overall conclusions would likely not have changed. Reduced susceptibility of *S. pneumoniae* to penicillin has led to modifications in the oral dosing of β-lactams, especially amoxicillin for a variety of respiratory conditions.[5-7] Future investigations may be warranted to confirm the validity of CLSI-adjudicated β-lactam extrapolative data to oral cephems[23] on a local basis. Finally, different tetracycline class agents were surveilled between the two laboratory entities. While tetracycline-susceptible results do extrapolate into doxycycline susceptibility for *S. pneumoniae*, the converse scenario is not always observed.[15]

In conclusion, temporal and geographic differences in resistance to *S. pneumoniae* exist in Wisconsin, and likely elsewhere. Surveillance programs should be essential to assist in guiding clinician decisions on treatment of bacterial respiratory infections. Based on the observed trend in increasing macrolide resistance, and stable and relatively high percentage of tetracycline/doxycycline susceptibility in Wisconsin, it is suggested that when β-lactam antimicrobial alternatives are needed that clinicians strongly consider using doxycycline preferentially over macrolides in appropriate age groups for the treatment of CAP, and perhaps for acute sinusitis and otitis media as well.

Acknowledgments

The authors thank public health microbiologists at the Wisconsin State Laboratory of Hygiene for past and continual expert technical assistance. The authors further recognize the efforts of clinical microbiologists across Wisconsin for contributions to the SWOTARE program:

Kathy Lang (Ashland)

Thomas R. Fritsche, MD, PhD, Brooke Olson (Marshfield)

Ashley Gargulak, Mattie Pitts (Spooner)

Becky Brooks (Stevens Point)

Joshua Kropp, Jennifer Meyer (Weston)

Benjamin Kaetterhenry (Appleton)

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References

1. Mandell LA, Wunderink RG, Anzueto A, et al; Infectious Diseases Society of America; American Thoracic Society. Infectious Diseases Society of America/American Thoracic Society consensus guidelines on the management of community-acquired pneumonia in adults. Clin Infect Dis. 2007;44(Supplement_2):S27-S72. doi:10.1086/511159
2. Lynch JP III, Martinez FJ. Clinical relevance of macrolide-resistant *Streptococcus pneumoniae* for community-acquired pneumonia. Clin Infect Dis. 2002;34(Supplement_1):S27-S46. doi:10.1086/324527

3. Jacobs MR. *In vivo veritas*: *in vitro* macrolide resistance in systemic *Streptococcus pneumoniae* infections does result in clinical failure. Clin Infect Dis. 2002;35(5):565-569. doi:10.1086/341980
4. Pérez-Trallero E. Pneumococcal macrolide resistance—not a myth. J Antimicrob Chemother. 2000;45(3):401-402. doi:10.1093/jac/45.3.401
5. Metlay JP, Waterer GW, Long AC, et al. Diagnosis and treatment of adults with community-acquired pneumonia. An official clinical practice guideline of the American Thoracic Society and Infectious Diseases Society of America. Am J Respir Crit Care Med. 2019;200(7):e45-e67. doi:10.1164/rccm.201908-1581ST
6. Lieberthal AS, Carroll AE, Chonmaitree T, et al. The diagnosis and management of acute otitis media. Pediatrics. 2013;131(3):e964-e999. doi:10.1542/peds.2012-3488 [Correction published in Lieberthal AS, Carroll AE, Chonmaitree T, et al. Clinical Practice Guideline: The Diagnosis and Management of Acute Otitis Media. *Pediatrics*. 2013;131(3):e964–e999. *Pediatrics* February 2014; 133 (2): 346–347. doi:10.1542/peds.2013-3791].
7. Chow AW, Benninger MS, Brook I, et al; Infectious Diseases Society of America. IDSA clinical practice guideline for acute bacterial rhinosinusitis in children and adults. Clin Infect Dis. 2012;54(8):e72-e112. doi:10.1093/cid/cis370
8. Owens RC Jr, Tessier P, Nightingale CH, Ambrose PG, Quintiliani R, Nicolau DP. Pharmacodynamics of ceftriaxone and cefixime against community-acquired respiratory tract pathogens. Int J Antimicrob Agents. 2001;17(6):483-489. doi:10.1016/S0924-8579(01)00322-3
9. File TM Jr. Clinical implications and treatment of multiresistant *Streptococcus pneumoniae* pneumonia. Clin Microbiol Infect. 2006;12(Suppl 3):31-41. doi:10.1111/j.1469-0691.2006.01395.x
10. Munson E, Hueppchen E, Zeman H. Surveillance of Wisconsin Organisms for Trends in Antimicrobial Resistance and Epidemiology: introduction to the program and summary of 2016 geographic variation. WMJ. 2018;117(3):116-121.
11. Munson E, Zeman H, Hueppchen E. Surveillance of Wisconsin Organisms for Trends in Antimicrobial Resistance and Epidemiology (SWOTARE): epidemiologic correlates for 2016 surveillance isolates. Gundersen Medical Journal. 2017;10(1):41-48.
12. Schulte RH, Munson E. *Staphylococcus aureus* resistance patterns in Wisconsin: 2018 Surveillance of Wisconsin Organisms for Trends in Antimicrobial Resistance and Epidemiology (SWOTARE) program report. Clin Med Res. 2019;17(3-4):72-81. doi:10.3121/cmr.2019.1503
13. Reynoso A, Munson E. Surveillance of Wisconsin Organisms for Trends in Antimicrobial Resistance and Epidemiology (SWOTARE): 2018-2019 report on *Enterobacter cloacae* and *Klebsiella pneumoniae* clinical isolates. Clin Med Res. 2021;19(3):123-131. doi:10.3121/cmr.2021.1588
14. Clinical and Laboratory Standards Institute (CLSI). Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically. 11th ed. CLSI supplement M07. Clinical and Laboratory Standards Institute, Wayne, PA. 2018.
15. Clinical and Laboratory Standards Institute (CLSI). Performance standards for antimicrobial susceptibility testing, M100. 31st informational supplement. Wayne, PA: CLSI; 2021.
16. National Committee for Clinical Laboratory Standards (NCCLS). Performance standards for antimicrobial susceptibility testing, M100. 13th informational supplement (M7). Villanova, PA: NCCLS; 2003.
17. Clinical and Laboratory Standards Institute (CLSI). Performance standards for antimicrobial susceptibility testing, M100. 18th informational supplement. Wayne, PA: CLSI; 2008.
18. Lazzerini G, Lavey SC, Fox BC, Munson E. Surveillance of fluoroquinolone resistance in Wisconsin: geographic variation and impact of revised CLSI breakpoints. Clin Med Res. 2022;20(2):81-88. doi:10.3121/cmr.2021.1718
19. Gupta V, Yu KC, Schranz J, Gelone SP. A multicenter evaluation of the US prevalence and regional variation in macrolide-resistant *S. pneumoniae* in ambulatory and hospitalized adult patients in the United States. Open Forum Infect Dis. 2021;8(7):ofab063.
20. Low DE. What is the relevance of antimicrobial resistance on the outcome of community-acquired pneumonia caused by *Streptococcus pneumoniae*? (should macrolide monotherapy be used for mild pneumonia?). Infect Dis Clin North Am. 2013;27(1):87-97. doi:10.1016/j.idc.2012.11.013
21. Musher DM, Abers MS, Bartlett JG. Evolving understanding of the causes of pneumonia in adults, with special attention to the role of pneumococcus. Clin Infect Dis. 2017;65(10):1736-1744. doi:10.1093/cid/cix549
22. Jones RN, Stilwell MG, Wilson ML, Mendes RE. Contemporary tetracycline susceptibility testing: doxycycline MIC methods and interpretive criteria (CLSI and EUCAST) performance when testing Gram-positive pathogens. Diagn Microbiol Infect Dis. 2013;76(1):69-72. doi:10.1016/j.diagmicrobio.2013.01.023

23. Murphy ME, Powell E, Courter J, Mortensen JE. Predicting oral beta-lactam susceptibilities against *Streptococcus pneumoniae.* BMC Infect Dis. 2021;21(1):679. doi:10.1186/s12879-021-06341-y

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