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EPIDEMIOLOGY

Persistent Transmission of Shigellosis in England Is Associated with a Recently Emerged Multidrug-Resistant Strain of *Shigella sonnei*

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ABSTRACT Whole-genome sequencing has enhanced surveillance and facilitated detailed monitoring of the transmission of *Shigella* species in England. We undertook an epidemiological and phylogenetic analysis of isolates from all cases of shigellosis referred to Public Health England between 2015 and 2018 to explore recent strain characteristics and the transmission dynamics of *Shigella* species. Of the 4,950 confirmed cases of shigellosis identified during this period, the highest proportion of isolates was *Shigella sonnei* (54.4%), followed by *S. flexneri* (39.2%), *S. boydii* (4.1%), and *S. dysenteriae* (2.2%). Most cases were adults (82.9%) and male (59.5%), and 34.9% cases reported recent travel outside the United Kingdom. Throughout the study period, diagnoses of *S. flexneri* and *S. sonnei* infections were most common in men with no history of recent travel abroad. The species prevalence was not static, with cases of *S. flexneri* infection in men decreasing between 2015 and 2016 and the number of cases of *S. sonnei* infection increasing from 2017. Phylogenetic analysis showed this recent increase in *S. sonnei* infections was attributed to a novel clade that emerged from a Central Asia sublineage exhibiting resistance to ciprofloxacin and azithromycin. Despite changes in species prevalence, diagnoses of *Shigella* infections in England are persistently most common in adult males without a reported travel history, consistent with sexual transmission among men who have sex with men. The trend toward increasing rates of ciprofloxacin resistance in *S. sonnei*, in addition to plasmid-mediated azithromycin resistance, is of significant public health concern with respect to the transmission of multidrug-resistant gastrointestinal pathogens and the risk of treatment failures.

KEYWORDS shigellosis, epidemiology, whole-genome sequencing, sexual transmission, multidrug resistance

The four species of *Shigella* (*Shigella sonnei*, *S. flexneri*, *S. boydii*, and *S. dysenteriae*) cause dysentery and are transmitted via the fecal-oral route. Shigellosis is the second leading cause of diarrheal deaths worldwide, and the highest burden is found in lower- to middle-income countries (1). In high-income countries, shigellosis was historically associated with either travel-associated infections or self-limited transmission in community risk groups. However, since being identified as a sexually transmitted infection (STI) in 1974 in the San Francisco, CA, homosexual male community (2, 3), sustained sexual transmission has become an important component of *Shigella* species epidemiology. Outbreaks of *S. sonnei* and *S. flexneri* among homosexual men, bisexual men, and other men who have sex with men (GBMSM) have been reported in urban settings across North America, Europe, Asia, and Oceania (4–12).

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In England, the epidemiology of infections with *Shigella* species has changed markedly in the past decade, from being predominantly travel-associated infections to being domestically acquired infections in men, which account for a large and increasing proportion of diagnoses. National surveillance data, case questionnaires, and outbreak investigations highlight considerable and sustained sexual transmission within specific sexual networks (4, 13, 14). Since 2009, overlapping GBMSM-associated epidemics of *S. flexneri* serotype 3a, *S. flexneri* serotype 2a, and, more recently, *S. sonnei* have emerged, while the numbers have remained stable in females and travel-associated cases (13).

Whole-genome sequencing (WGS) studies have been used to determine the factors driving the emergence and transmission of *Shigella* species and have previously demonstrated that certain community outbreaks of shigellosis in the United Kingdom belonged to prolonged, global epidemics (5, 15–18). These analyses showed that lineages of *S. flexneri* serotypes 3a and 2a exhibited intercontinental spread via sexual transmission in GBMSM networks and subsequently acquired multiple antimicrobial resistance (AMR) determinants (5, 17). Within the *S. sonnei* population, Baker et al. (2018) (16) defined four clades associated with transmission in GBMSM communities, and all were within lineage III, the dominant extant lineage both in the United Kingdom and around the world (19, 20). A 2-year population-level study of all cultured *Shigella* isolates in the state of Victoria, Australia (2016 to 2018), identified two predominant lineages circulating among GBMSM (21). Specifically, these consisted of the *S. flexneri* 2a lineage described above and a multidrug-resistant *S. sonnei* clade associated with reduced susceptibility to azithromycin, trimethoprim-sulfamethoxazole, and ciprofloxacin.

The implementation of WGS in 2015 at Public Health England (PHE) has enhanced surveillance and facilitated the monitoring of transmission of *Shigella* species in England (20, 22). This article uses national surveillance data from 2015 to 2018 to describe the epidemiology of *Shigella* species in England in the post-WGS era. We undertook epidemiological and phylogenetic analyses of *Shigella* isolates referred to the Gastrointestinal Bacterial Reference Unit (GBRU) of PHE to explore recent characteristics and the patterns of transmission of shigellosis in England to better inform the public health response. We examined trends in travel-associated and non-travel-associated acquisition by gender to assess evidence for the ongoing sexual transmission of *Shigella* species among men and to report on genomic subtype and antimicrobial resistance changes across the population.

MATERIALS AND METHODS

Data collection. The UK Standards for Microbiology Investigation of Faecal Specimens for Enteric Pathogens recommends testing of all fecal specimens for *Shigella* species in individuals reporting symptoms of gastrointestinal disease (https://www.gov.uk/government/publications/smi-b-30-investigation-of-faecal-specimens-for-enteric-pathogens). Analysis of the number of *Shigella* species reported by the local hospital laboratories to the Second Generation Surveillance System (https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/739854/PHE_Laboratory_Reporting_Guidelines.pdf) shows that approximately two-thirds of isolates are submitted from the local hospital laboratories to Public Health England's Gastrointestinal Bacterial Reference Unit (GBRU) for confirmation of species identification and typing. Subsequent microbiological typing data, including serotyping and single nucleotide polymorphism (SNP) typing, and patient demographic data, including, sex, age, and recent travel, collected from laboratory request forms upon isolate submission are stored in an integrated molecular national surveillance database.

Data analysis. Microbiological typing data from all isolates of *Shigella* species submitted to GBRU between January 2015 and December 2018 were extracted and analyzed. Travel-associated cases were defined as those reporting recent foreign travel to any country (including both high-risk countries where shigellosis is endemic and low-risk countries) 7 days prior to the onset of symptoms, based on information from laboratory reports. Travel history is captured for between 60 and 70% of cases of *Shigella* species; if no travel history is reported, it cannot be inferred that the case did not travel. Laboratory surveillance data lack information on patient sexual orientation. However, in line with previous work, we calculated gender ratios as an indicator of cases that may be attributed to sexual transmission among GBMSM (13, 23).

First, we analyzed the demographic data available, specifically, age, sex, and travel history, linked to the reference laboratory-confirmed cases of shigellosis in England for all *Shigella* species and then for each individual species of *Shigella*. We then analyzed the WGS data for genomic markers of resistance to azithromycin (*ermB* and *mphA*) and ciprofloxacin (mutations in the quinolone resistance-determining

region [QRDR] of *gyrA* and *parC*) for all *Shigella* species and for each individual species of *Shigella*. Finally, we investigated the trends in the number of cases and resistance to azithromycin and ciprofloxacin among *S. flexneri* serotype 2a and *S. flexneri* serotype 3a isolates and the clades of *S. sonnei* previously associated with sexual transmission in men.

Whole-genome sequencing. Since August 2015, microbiological typing, including confirmation of the species and the serotype, has been performed at PHE using WGS (20, 22). DNA was extracted for sequencing on an Illumina HiSeq 2500 instrument. Quality and adapter trimmed Illumina reads were aligned to a reference genome, consisting of the *S. sonnei* strain Ss46 (GenBank accession number NC_007384.1) or *S. flexneri* serotype 2a strain 2457T (GenBank accession number AE014073.1) genome, using the BWA MEM (v0.7.12) program (24). Single nucleotide polymorphisms (SNPs) were identified using the GATK (v2.6.5) tool (25) in the unified Genotyper mode. Core genome positions that had a high-quality SNP (>90% consensus; minimum depth, 10 times; mapping quality [MQ] $\geq$ 30) in at least one isolate were extracted, and the RAxML (v8.2.8) program (26) was used to derive the maximum likelihood phylogeny of the isolates, after first removing regions of the genome predicted to have undergone horizontal exchange, using the Gubbins (v2.0.0) program (27). Hierarchical single-linkage clustering was performed on the pairwise SNP difference between all isolates at various distance thresholds (Δ250, Δ100, Δ50, Δ25, Δ10, Δ5, Δ0) (28).

Identification to the species level was done by kmer identification, as described by Chattaway et al. (2017) (29). Genome-derived serotyping and AMR determinant profiling were performed using the GeneFinder tool (https://github.com/phe-bioinformatics/gene_finder), run using the default parameters. For serotyping, a reference database containing the gene sequences encoding the 12 O-antigen synthesis or modification genes, including *wzxc1-5*, *wzxe1-5*, *wzx6*, *gtrI*, *gtrII*, *gtrIV*, *gtrV*, *gtrX*, *gtr1c*, *oac*, *oac1b*, and *opt*, was constructed (22). Only *in silico* predictions of serotype that matched a gene determinant at >80% nucleotide identity over >80% of the target gene length were accepted.

For AMR determinant profiling, genes were defined as present if they represented 100% of the reference sequence with greater than 90% nucleotide identity (20, 30). The reference database for AMR determinants can also be found in the GeneFinder github repository (https://github.com/phe-bioinformatics/gene_finder/tree/master/refs). The rates of resistance to streptomycin, tetracycline, the sulfonamides, and trimethoprim in *Shigella* species are high, and the trends have been consistent for many years (5, 15–17, 30). We therefore focused our AMR analysis on the presence of genomic makers of resistance to macrolides (specifically, *ermB* and *mphA*) and fluroquinolones (specifically, mutations in the quinolone resistance-determining regions [QRDR] of *gyrA* and *parC*), as the trends in resistance to these clinically relevant classes of antimicrobials fluctuated during the study period. The presence of the IncFII plasmid (pKSR100) (5, 17) was determined by mapping to the reference sequence with GenBank accession number LN624486 using the BWA MEM (v0.7.12) program (24), with a positive match being defined as a >90% read coverage of pKSR100. For international context, 110 *S. sonnei* sequences from isolates from GBMSMs in Australia, previously reported by Ingle et al. (2019) (21), were processed as described above for phylogenetic comparison.

Timed phylogenies were constructed using the BEAST-MCMC (v2.4.7) program (31) with the SNP alignments prepared as described above. Alternative clock models and population priors were computed and assessed based on Bayes factor (BF) tests using the Tracer (v1.6) program. The highest supported model was a strict clock rate under a Bayesian skyline coalescent population. All models were run with a chain length of 1 billion. A maximum clade credibility tree was constructed using the TreeAnnotator (v1.75) program (31).

Data availability. FASTQ reads from all sequences in this study can be found at the PHE BioProject under accession number PRJNA315192. The Short Read Archive accessions numbers for all 4,950 sequenced isolates in this study are listed Table S1 in the supplemental material, along with corresponding information on the isolate (month and date of isolation, species, serotype, clade, and the presence of genetic determinants known to confer resistance to fluroquinolones or azithromycin) and the patient (sex, age status, and travel status). Cases were assigned to be non-travel associated if they reported no recent travel or if no travel history was reported.

RESULTS

Epidemiology of shigellosis in England. Between 2015 and 2018, there were 4,950 reference laboratory-confirmed cases of shigellosis, of which the majority were in males (2,946/4,950, 59.5%) and adults aged ≥16 years (4,105/4,950, 82.9%) (see Table S1 in the supplemental material). In total, 1,725/4,950 (34.9%) cases reported recent travel outside the United Kingdom. When stratified by age, children (age < 16 years) (262/836, 31.3%) reported significantly less recent travel than adults (1,462/4,105, 35.6%) (chi-square test, $P < 0.05$). Among adult shigellosis cases, the proportion of males reporting recent travel outside the United Kingdom during this period was significantly lower than the proportion of females (626/2,518 [24.9%] for males versus 825/1,544 [53.4%] for females; chi-square test, $P < 0.001$).

During the study period, the number of isolates of *Shigella* species referred from non-travel-associated adult male cases exceeded the number referred from cases reporting recent travel at every time point, although marked fluctuations were observed (Fig. 1). An increase of adult, male, non-travel-associated cases was first ob-

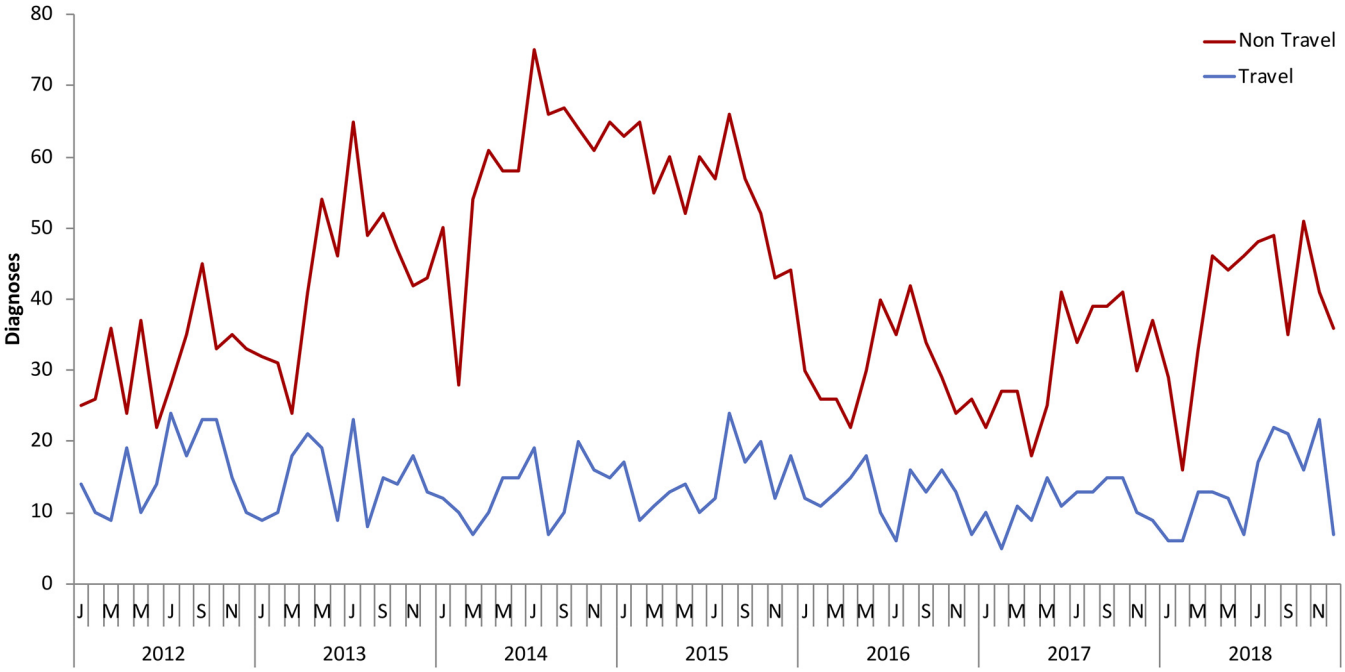

FIG 1 Trends in association of travel with *Shigella* species infection diagnoses among men aged ≥16 years in England. Data from Simms et al. (2015) (13), analyzed prior to this study, are included in this figure for context.

served in 2013, as previously reported by Simms et al. (2015), and was attributed to transmission among GBMSM (13) (Fig. 1). During the time frame of this study, adult, male, non-travel-associated cases of shigellosis remained at epidemic levels throughout the first quarter of 2015, before falling by 46% from 2015 to 2016 (674 to 364 cases), but then subsequently reemerged from 2017 onwards.

Species determination, serotyping, and analysis of patient demography. Of the 4,950 isolates of *Shigella* submitted to GBRU between 2015 and 2018, the highest proportion consisted of *S. sonnei* ($n = 2,695$), followed by *S. flexneri* ($n = 1,943$), *S. boydii* ($n = 201$), and *S. dysenteriae* ($n = 111$) (Table 1; Table S1). The most common *S. flexneri* serotypes were 2a ($n = 1,113/1,943$, 57.2%), 1b ($n = 254/1,943$, 13.1%), and 3a ($n = 206/1,943$, 10.6%). The most common serotypes of *S. boydii* were serotype 1 ($n = 24/201$, 11.9%), serotype 2 ($n = 42/201$, 20.9%), and serotype 4 ($n = 23/201$, 11.4%), and the most common serotype of *S. dysenteriae* was serotype 2 ($n = 33/110$, 30.0%). All isolates of *S. sonnei* expressed the same somatic antigen and therefore could not be serotyped.

Cases of *S. dysenteriae* (60.4%) or *S. boydii* (59.2%) infection were significantly more associated with recent foreign travel than cases of *S. sonnei* (36.8%) or *S. flexneri* (28.2%) infection ($P < 0.001$) (Table 1). For all species, similar proportions of cases of infection were found among adults, with the lowest proportion being cases of *S. dysenteriae* infection (77.5%) and the highest proportion being cases of *S. flexneri* infection (83.7%). Significantly higher proportions of cases of infection with *S. sonnei* (54.6%) and *S.*

TABLE 1 *Shigella* species in England from 2015 to 2018 by age, gender, and travel[a]

Species	No. (%) of cases						
	Total	Adults	Children	Males	Females	Travel associated	Non-travel associated
S. sonnei	2,695 (54.4%)	2,228 (82.7)	467 (17.3)	1,472 (54.6)	1,223 (45.4)	992 (36.8)	1,703 (63.2)
S. flexneri	1,943 (39.2)	1,627 (83.7)	316 (16.3)	1,342 (69.1)	601 (30.9)	547 (28.2)	1,396 (71.8)
S. boydii	201 (4.1)	164 (81.6)	37 (18.4)	88 (43.8)	113 (56.2)	119 (59.2)	82 (40.8)
S. dysenteriae	111 (2.2)	86 (77.5)	25 (22.5)	44 (39.6)	67 (60.4)	67 (60.4)	44 (39.6)

[a]Data are for 4,950 individuals.

TABLE 2 Presence of AMR determinants encoding resistance to macrolides and to fluroquinolones in adult, non-travel-associated cases of shigellosis[a]

AMR determinant	No. (%) of cases					
	Male			Female		
	All cases (*n* = 1,458)	*S. flexneri* (*n* = 720)	*S. sonnei* (*n* = 738)	Total (*n* = 573)	*S. flexneri* (*n* = 171)	*S. sonnei* (*n* = 402)
ermB and *mphA*	774 (53.1)	390 (54.2)	384 (52.0)	30 (5.2)	8 (4.7)	22 (5.5)
Triple QRDR mutations	412 (28.3)	63 (8.8)	349 (47.3)	133 (23.2)	41 (24.0)	92 (22.8)

[a]*ermB* and *mphA* are the determinants of resistance to macrolides, and triple mutations in the QRDR are the determinants of resistance to fluroquinolones.

flexneri (69.1%) than with *S. boydii* (43.8%) and *S. dysenteriae* (39.6%) were seen among males (chi-square test, $P < 0.001$) (Table 1).

Genomic surveillance of markers of antimicrobial resistance to azithromycin and ciprofloxacin. The genome-derived AMR profiles were available for 4,057/4,950 (81.4%) isolates of *Shigella* in the study, as the study period started in January 2015 and WGS was implemented in August 2015 (Table S1). Determinants encoding azithromycin resistance, *ermB* and *mphA*, were most commonly detected in isolates responsible for male, non-travel-associated cases of *S. flexneri* and *S. sonnei* infection, with 45% of isolates displaying this resistance genotype but with <5% of isolates from non-travel-associated female cases displaying this resistance genotype (for males reporting travel, *n* = 63/660 [9.5%]; for males not reporting travel, *n* = 776/1,728 [45.0%]; for females reporting travel, *n* = 5/794 [0.6%]; for females not reporting travel, *n* = 42/875 [4.8%]). Analysis of the available data from adult non-travel-associated cases of *S. flexneri* and *S. sonnei* infections showed that *ermB* and *mphA* were most commonly identified in men (Table 2). No azithromycin resistance was detected in *S. boydii* or *S. dysenteriae* isolates. Overall, the rate of resistance to azithromycin remained stable during the study period (Fig. 2, top left), although a downward trend in the rate of azithromycin resistance associated with *S. flexneri* isolates from male cases and an upward trend in the rate of azithromycin resistance identified among isolates of *S. sonnei* from male cases were observed (Fig. 2, bottom left).

The prevalence of triple mutations in the QRDR, known to confer resistance to ciprofloxacin, was higher in isolates from males than in isolates from females, but similar proportions were detected in isolates from travel- and non-travel-associated cases in both men and woman (for males reporting travel, *n* = 172/660 [26.1%]; for males not reporting travel, *n* = 456/1,728 [26.4%]; for females reporting travel, *n* = 171/794 [21.5%]; for females not reporting travel, *n* = 178/875 [20.3%]). Analysis of the available data from adult non-travel-associated cases of *S. flexneri and S. sonnei* infections showed that triple QRDR mutations were most commonly identified in isolates of *S. sonnei* from males (Table 2). Throughout the study period, the proportion of shigellae with ciprofloxacin resistance-conferring QRDR mutations was stable in females and males reporting travel. However, a proportional increase was identified in the male non-travel-associated cases (Fig. 2, top right). Stratification of these data by *Shigella* species showed that this trend was caused by an increase in the number of isolates of *S. sonnei* (Fig. 2, bottom right). By 2018, 80% of adult male non-travel-associated cases had triple QRDR mutations.

Phylogenetic trends in *S. sonnei* and *S. flexneri* serotype 2a and 3a GBMSM-associated clades. Among all adult cases without a recent travel history, *S. flexneri* serotype 2a and *S. flexneri* serotype 3a (previously implicated in GBMSM-associated shigellosis epidemics) accounted for 32.3% (1,041/3,225) of all laboratory-confirmed infections between 2015 and 2018. The age and sex distributions for cases of *S. flexneri* 2a and *S. flexneri* 3a infection were strongly associated with males (82.3%, 860/1,041) over the age of 16 years (91.5%, 787/860). The fall in the incidence of laboratory-confirmed infections with *S. flexneri* 3a in adult males with no travel history first reported in 2014 has continued throughout this study period, and the numbers of isolates submitted nationally are now at preepidemic levels (Fig. 3; Fig. S1). Diagnoses

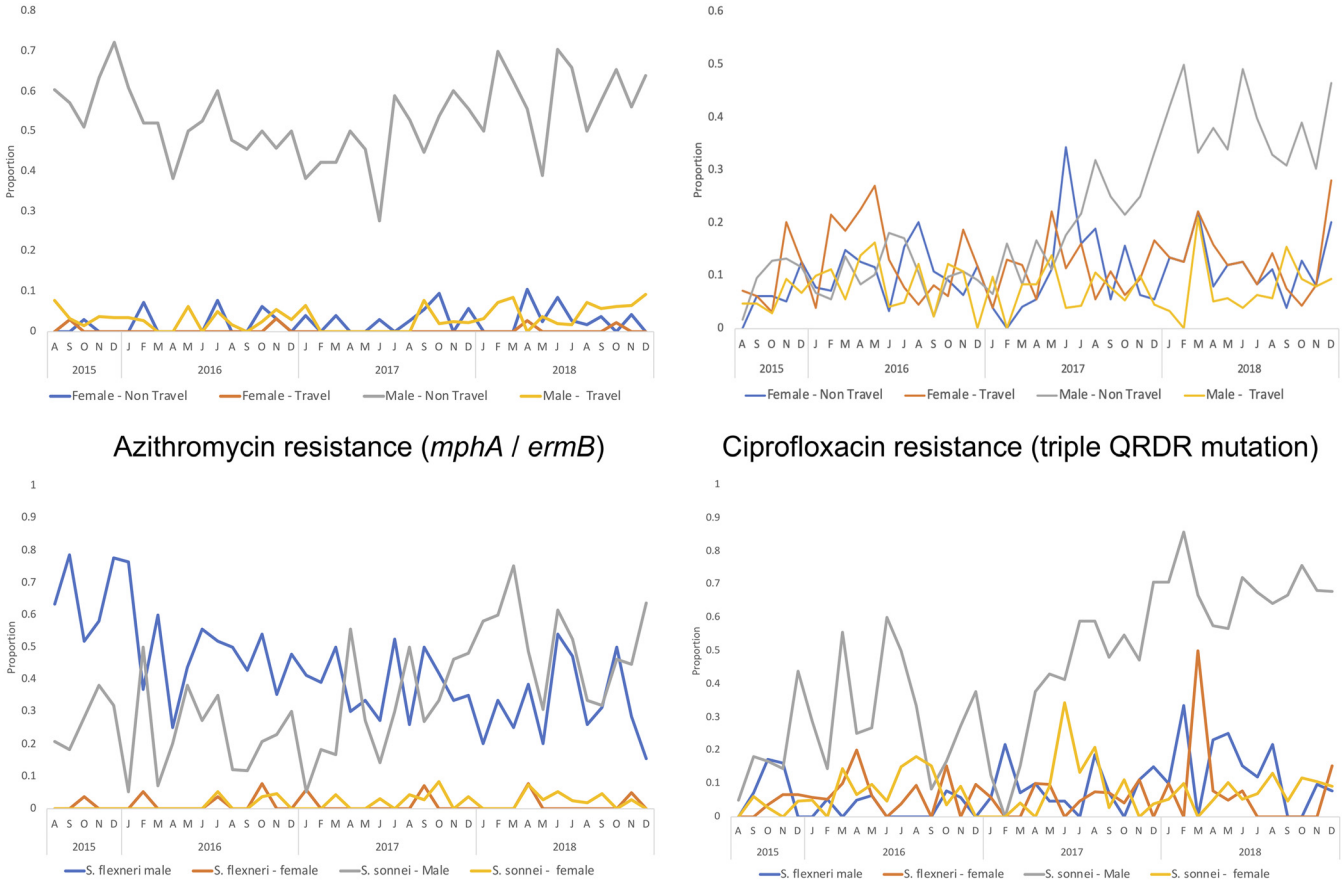

FIG 2 (Top) Trends in male and female cases reporting travel outside the United Kingdom within 7 days of the onset of symptoms and those designated non-travel associated (defined as cases either reporting no travel or not reporting any travel information) caused by isolates harboring genomic markers for resistance to azithromycin and ciprofloxacin. (Bottom) Trends in adult male and female cases designated non-travel associated caused by isolates of *S. flexneri* or *S. sonnei* harboring genomic markers for resistance to azithromycin and ciprofloxacin.

of *S. flexneri* 2a infections in men also declined, with a 71.4% fall during this study period; from the peak in 2015 (297 cases), cases fell annually to 144 cases in 2016, 140 cases in 2017, and just 85 cases in 2018 (Fig. 3). Diagnoses from women during this period remained low at less than 12 cases in each quarter (Table 1; Fig. 2).

Phylodynamic analysis of the genome sequences of *S. flexneri* serotype 2a and 3a GBMSM-associated clades across the two epidemics showed that the decrease in case numbers described above is reflected in decreased sample diversity over time. Bayesian skyline plots revealed that the effective population size of *S. flexneri* 3a has decreased to preepidemic levels, whereas for *S. flexneri* 2a, the effective population size is a log fold change less than that at the height of the epidemic in 2015, although it is still above preepidemic levels (Fig. S1).

S. sonnei, also previously implicated in transmission among GBMSM, accounted for over half (52.8%, 1,703/3,225) of reported cases of shigellosis among those without a recent travel history between 2015 and 2018. The age and sex distributions for cases of *S. sonnei* infection without a recent travel history showed that they were also associated with males (61.8%, 1,052/1,703) over the age of 16 years (85.9%, 904/1052). Non-travel-associated cases of *S. sonnei* among men were reduced from 2015 to 2016, in parallel with the reduction of *S. flexneri* serotype 2a infections, but increased again from mid-2017, with the highest number of diagnoses in the study period being reported in the second to fourth quarters of 2018 (Fig. 4). Female cases remained relatively stable, except for a notable decline in the first quarter of 2018 and a subsequent peak in the third quarter of 2018.

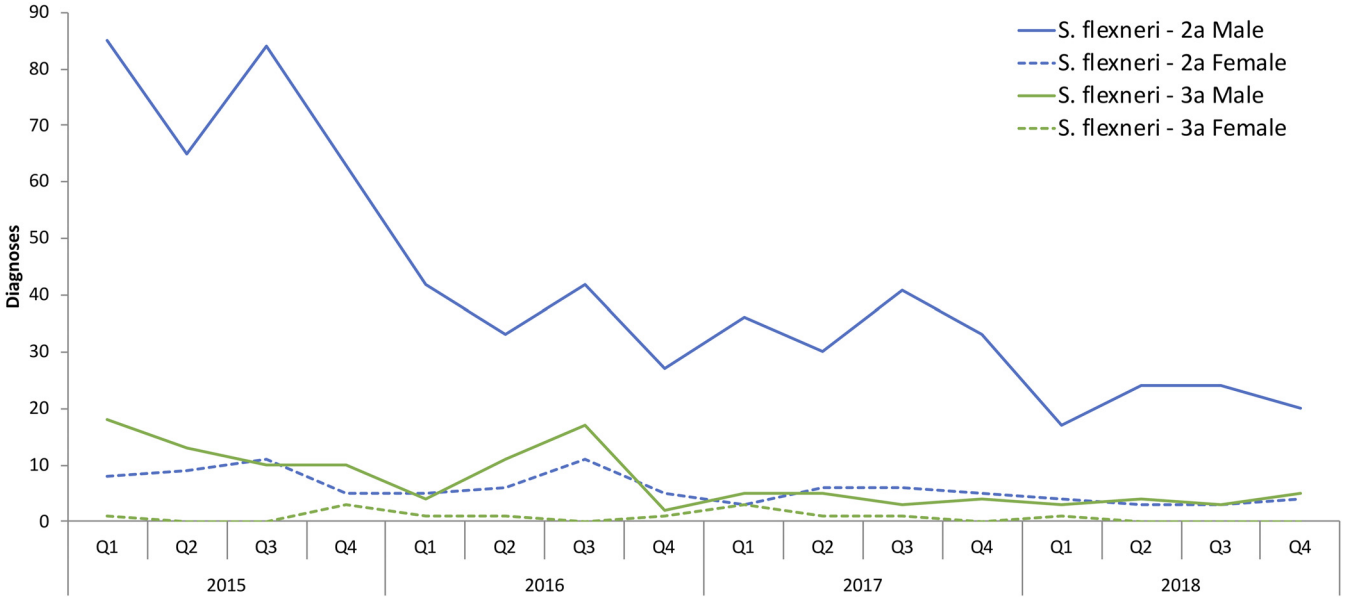

FIG 3 Trends in the number of cases of *S. flexneri* serotype 2a and 3a infections in adult males and females. Q1, Q2, Q3, and Q4, first, second, third, and fourth quarters, respectively.

A phylogenetic analysis was performed to determine whether the four clades of *S. sonnei* previously determined to be circulating in the United Kingdom GBMSM population between 2008 and 2014 (*S. sonnei* clades 1 to 4 in 2018) were contributing to the recent rise in suspect GBMSM-associated cases of *S. sonnei* infection (Fig. 5; Table S1). Of these four clades, only clades 2 and 4 caused cases during the study period, and this was at a stable rate, with both clades causing a median of two cases per month. Isolates in both clade 2 and clade 4 have acquired an IncFII plasmid (pKSR100) harboring macrolide resistance genes (*mphA* and *ermB*) on multiple occasions. In addition, isolates in clade 2 have a single QRDR mutation known to result in reduced susceptibility to ciprofloxacin, whereas isolates in clade 4 have no QRDR mutations.

Phylodynamics analysis suggests that both clade 2 and clade 4 have a declining effective population size, consistent with the epidemiological evidence of stable case rates attributable to these lineages, suggesting that these clades are not contributing to the recent increase in transmission. The recent increase in cases in this community were attributed to a novel clade, hereby designated clade 5. Like clade 1, clade 5 has emerged from the Central Asia III sublineage. Isolates from both clade 1 and clade 5

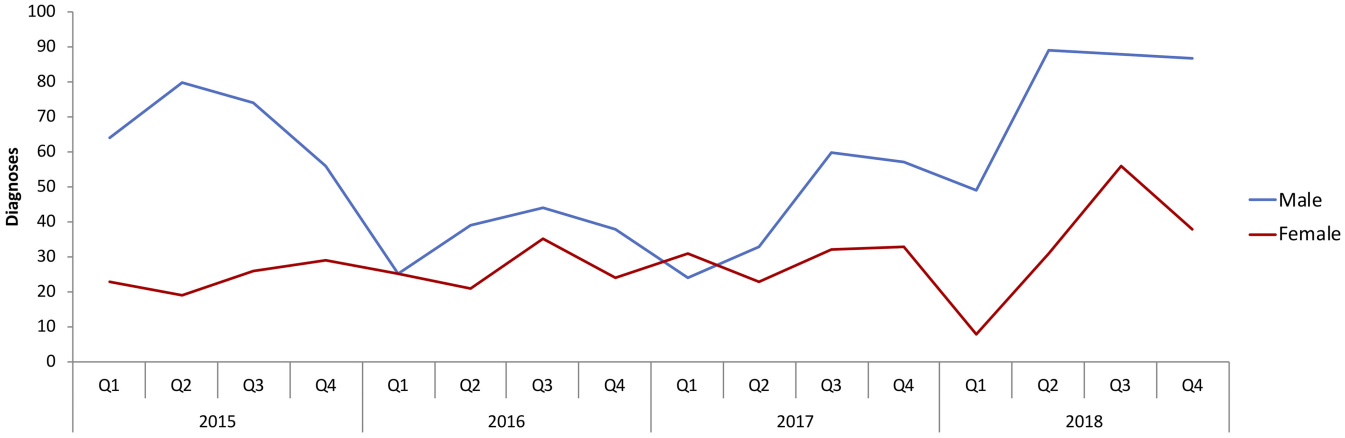

FIG 4 Diagnoses of non-travel-associated cases of *Shigella sonnei* infection among males aged ≥16 years.

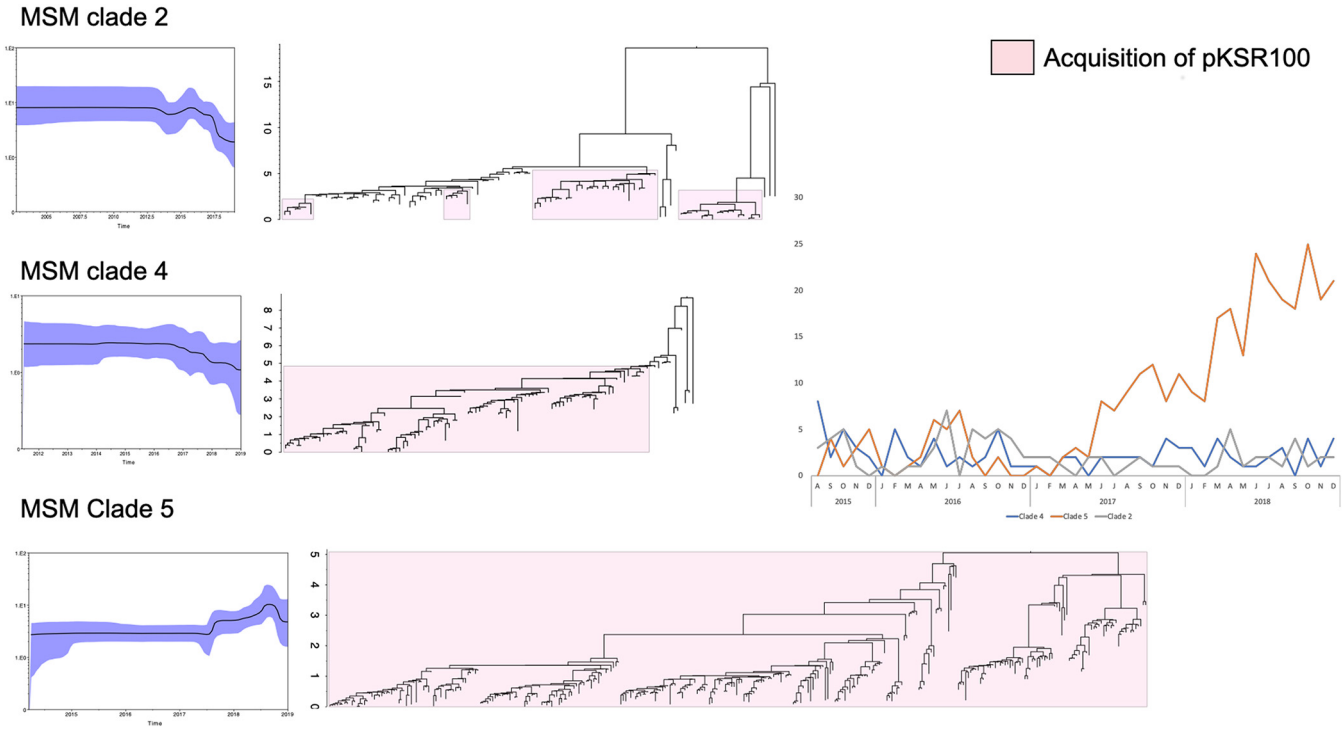

FIG 5 (Left) Phylodynamic analysis of *S. sonnei* clades 2, 4, and 5 associated with transmission among MSM. Maximum clade credibility trees are presented, with Bayesian skyline plots showing the temporal changes of the effective population size of each clade. *x* axis, time in years; *y* axis, effective population size. The 95% highest posterior density interval is highlighted in purple. (Top right) Frequency of cases clustering within *S. sonnei* clades 2, 4, and 5 associated with transmission among GBMSM. Gray line, clade 2; blue line, clade 4; orange line, clade 5.

exhibited resistance to ciprofloxacin, mediated by triple mutations (*gyrA* S83L D87G, *parC* S80I) in the QRDR, and isolates from both clades have acquired pKSR100, resulting in macrolide resistance. Phylodynamics analysis suggests that clade 5 originated in about 2015 (95% highest posterior density, 2014.2 to 2012.7), and the increased isolation of clade 5 *S. sonnei* from male cases is reflected in an increased effective population size of this clade since 2017 (Fig. 5).

To place these findings in an international context, 110 *S. sonnei* genome sequences obtained from Australian GBMSMs (21) were analyzed to determine which, if any, of the clades identified in this study were contemporaneously circulating globally. Clade 2 isolates accounted for 88% (97/110) of the Australian GBMSM *S. sonnei* isolates, 6% (7/110) were part of clade 5, and the remaining 6 *S. sonnei* isolates were in other parts of the *S. sonnei* population not previously implicated in transmission among GBMSM. Of the Australian clade 5 isolates; one was from 2016, four were from 2017 and two were from 2018, with none of the individuals from whom these isolates were recovered reporting travel outside of Australia. All 7 isolates contained triple mutations in QRDR and carried the pKRS100 plasmid, encoding resistance to azithromycin, as reported in the English clade 5 isolates.

DISCUSSION

In this study, we present evidence that the previous GBMSM-associated epidemics of *S. flexneri* serotypes 2a and 3a have abated, while in contrast, non-travel-associated transmission of *S. sonnei* among males has intensified since mid-2017 and is associated with the recent emergence of a novel, specific multidrug-resistant clone, clade 5. Despite changes in species prevalence, diagnoses of infections caused by *Shigella* species remain largely restricted to males without a reported travel history and concentrated in urban settings, consistent with continued sexual transmission among GBMSM. As with previously observed shigellosis epidemics associated with transmission among GBMSM, there is evidence of the global dissemination of clade 5 strains

between continents. Furthermore, clade 5 strains have an enhanced ciprofloxacin resistance profile (triple QRDR mutations) compared to that of other GBMSM-associated lineages circulating, and as such, their emergence represents a significant public health concern.

Research has shown that proactive campaigns through targeted social media and leaflets in sexual health clinics have failed to raise awareness of shigellosis among GBMSM (32). Despite this, the number of cases of *S. sonnei* and *S. flexneri* serotype 2a infection diagnosed followed similar trends of decline between August 2015 and August 2017. This decrease in the number of cases reflects the decrease in the effective population size and strain diversity and results in lower levels of sexual transmission occurring within the GBMSM community. While the incidences of infections with *S. flexneri* 2a and 3a continued to fall, we report an emerging epidemic of *S. sonnei* among men in England without a travel history from August 2017 onwards. This increase in the number of cases reflects the increase in the effective population size and strain diversity and results in higher levels of sexual transmission occurring within the GBMSM community. Currently, *S. sonnei* is the most prevalent endemic species acquired among individuals in this population by orders of magnitude. It is possible that changes to the dominant subtype reflect levels of herd immunity (33) and that new subtypes have the potential to enter and spread within the GBMSM population under the right conditions.

Studies of ultraorthodox Jewish communities in Israel and abroad (34) have shown that in regions of endemicity the incidence of *S. sonnei* shigellosis follows a cyclical pattern, with epidemics occurring every 2 years. The timing of these cyclical epidemics of *S. sonnei* shigellosis are attributed to the waning rate of natural immunity to the organism (33, 34). It is likely that a similar phenomenon occurs among GBMSM; natural exposure to *Shigella* species through intensive shigellosis epidemics in the GBMSM community may also increase immunity levels in individuals belonging to sexual networks. Herd immunity may be sufficient to temporarily reduce the circulation of *Shigella* species in these networks and prevent epidemics from being sustained in subsequent years. However, eventual waning levels of antibodies may lead to a decrease in herd immunity below a critical level and thus enable renewed epidemic transmission of shigellosis in GBMSM. This phenomenon may also facilitate strain replacement events, in which circulating strains are replaced by strains with different immunogenic properties or fitness profiles. Given the indication here that we should anticipate repeated epidemics of strains imported from areas of endemicity, future studies to develop a predictive framework from genomic information would be beneficial.

Given the ever-increasing reports of AMR in *Shigella* species and evidence that strains harboring mobile resistance-conferring plasmids can spread intercontinentally through sexual transmission (5), therapy guided according to susceptibility testing should be a priority. Previous studies have discussed the role of AMR in driving the transmission of shigellosis in the GBMSM community, and azithromycin resistance is regarded as a marker for isolates associated with sexual transmission among men who have sex with men (MSM) (5, 17). During the peak of transmission among GBMSM in 2014, only one of the four *S. sonnei* clades (clades 1 to 4) associated with transmission among GBMSM was resistant to ciprofloxacin (16). Of concern in this study was that the majority of suspect GBMSM-associated cases had become resistant to both azithromycin and ciprofloxacin. The clade 5 strain causing the majority of cases in 2017 and 2018 emerged from the same travel-associated clade as clade 1. Given this repeated epidemiological occurrence, it is likely that resistance to ciprofloxacin has driven, or at least contributed to, the emergence and dissemination of clade 5.

Current WHO guidelines recommend the use of fluoroquinolones as the first-line treatment for shigellosis. The trend toward increases in the rates of resistance to fluroquinolones observed in *S. sonnei* is a public health concern with respect both to the increased likelihood of treatment failures and to the transmission of fluoroquinolone-resistant strains of *S. sonnei* to the wider community. Evidence of the transmission of *S. flexneri* previously associated with transmission among GBMSM

spreading to the wider community has been described (35). Data on epidemiologically dominant AMR profiles in *Shigella* species would be a beneficial resource to be shared with clinicians to raise awareness and direct appropriate clinical management (35).

While WGS provides highly detailed and informative information on genetically linked clusters, a lack of patient exposure data, including sexual behavioral data, congregational settings, food, and travel history, linked to isolates within these clusters hinders the accurate determination of the impact of sexual transmission in shigellosis epidemiology and the most appropriate public health intervention. Collecting enhanced surveillance data for cases of shigellosis is essential in order to help distinguish between transmission among GBMSM and other forms of transmission requiring different public health actions.

Given the diversity of clinical settings in which shigellosis-affected GBMSM may present, it is important to consider awareness among clinicians. Health care professionals should be aware of the importance of sensitively obtaining a sexual history and provide advice about the risk of sexual transmission, the need to avoid sexual activity for at least 1 week after symptoms cease, and how to prevent onward transmission through sexual and nonsexual contacts. GBMSM with sexually acquired shigellosis are at high risk of STIs and HIV infection and should be referred to a sexual health clinic for comprehensive testing for HIV infection and STIs and partner notification (4). From a patient perspective, dense sexual networks, diverse sexual practices (including chemsex), and STI and/or HIV coinfection may contribute to the ongoing transmission of *Shigella* species among GBMSM (32, 35). Given the increasing rates of diagnoses of other sexually transmitted infections seen among GBMSM, such as gonorrhea, syphilis, and lymphogranuloma venereum, in the United Kingdom in recent years (36, 37; https://www.gov.uk/government/statistics/sexually-transmitted-infections-stis-annual-data-tables) and the threat of AMR, strengthened surveillance of *Shigella* species transmission is warranted.

SUPPLEMENTAL MATERIAL

Supplemental material is available online only.

SUPPLEMENTAL FILE 1, PDF file, 0.8 MB.

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