Genomic surveillance of invasive meningococcal disease during a national menw outbreak in Australia, 2017 – 2018

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Background: In Australia, invasive meningococcal disease (IMD) incidence rapidly increased between 2014 and 2017 due to rising serogroup W (MenW) and MenY infections. We aimed to better understand the genetic diversity of IMD during 2017-18 using whole-genome sequencing (WGS) data.

Methods: Whole-genome sequencing data from 440 Australian IMD isolates collected during 2017-18 and 1737 international MenW:CC11 isolates collected in Europe, Africa, Asia, North America, and South America between 1974 and 2020 were used in phylogenetic analyses; genetic relatedness was determined from single nucleotide polymorphisms.

Results: Australian isolates comprised 181 MenW (41%), 144 MenB (33%), 88 MenY (20%), 16 MenC (4%), 1 MenW/Y (0.2%), and 10 non-genogroupable (2%) isolates. Eighteen clonal complexes (CC) were identified; three (CC11, CC23, CC41/44) accounted for 78% of isolates (343/440). These CCs were associated with specific serogroups; CC11 (199) predominated among MenW (181) & MenC (15), CC23 (80) among MenY (78), and CC41/44 (64) among MenB (64). MenB isolates were highly diverse, MenY intermediately diverse, and MenW & MenC isolates demonstrated the least genetic diversity. Thirty serogroup and CC-specific genomic clusters were identified. International CC11 comparison revealed diversification of MenW in Australia.

Conclusions: WGS comprehensively characterised Australian IMD isolates, indexed their genetic variability, provided increased within-CC resolution, and further elucidated the evolution of CC11 in Australia.

Keywords: Neisseria meningitidis, meningococcal disease, pathogen genomics, genomic epidemiology, whole genome sequencing, public health.

INTRODUCTION

Invasive meningococcal disease (IMD) is a rare but life-threatening infection caused by the bacterium Neisseria meningitidis, an obligate human pathogen that commonly colonises the nasopharynx [1]. IMD is an important cause of mortality and morbidity, due to its significant case fatality rate (5-15%) and high incidence of sequelae among survivors (10-20%) [2, 3].
N. meningitidis is classified into 12 serogroups based on capsular polysaccharide structure, with six (A, B, C, W, X, Y) causing most IMD worldwide [4, 5]. Vaccines against serogroups A/B/C/W/Y are currently available [6]. Meningococci are classified into lineages (sequence types, ST) based on multilocus sequence typing (MLST) [7]. Closely related STs form clonal complexes (CCs) [7]. Some CCs (e.g., CC41/44, CC11) cause most IMD and are termed ‘hyperinvasive lineages’ [8, 9].

The epidemiology of IMD is unpredictable. Disease incidence and serogroup distribution varies temporally and geographically [5, 10]. The emergence of serogroup W (MenW) exemplifies this dynamism. Previously a rare cause of IMD, the first recorded MenW outbreak occurred in 2000 during the annual Hajj pilgrimage in Saudi Arabia, followed by a near-identical outbreak in 2001 [11, 12]. MenW subsequently emerged in the ‘meningitis belt’ of sub-Saharan Africa, causing epidemics from the early 2000s [12, 13]. Thereafter, outbreaks and increases in endemic disease due to MenW were reported worldwide, including in South America, Europe, and Australasia [14-17]. Many countries responded with MenACWY vaccination programs [10, 18].

Molecular typing of Hajj outbreak isolates identified a hypervirulent CC11 (PorA type P1.5,2) strain [12, 19]. Globally, most emergent MenW was attributed to CC11 [14]. Analysis of whole-genome sequencing (WGS) data proved instrumental in elucidating the within-clone diversity of MenW:CC11 [20]. Two sub-lineages were identified: the Hajj and South American/UK sub-lineages; the latter now delineated into ‘original UK’ and ‘UK-2013’ strains [20, 21]. The original UK and UK-2013 strains were notable for atypical clinical presentations [22].

IMD is a notifiable disease in Australia; a federal nation comprising eight states and territories [23]. IMD incidence decreased between 2002 and 2013 (from 3.5 to 0.6 cases per 100,000 population) [24]. This was associated with declining MenB and MenC infections and followed the 2003 inclusion of MenC vaccines on the National Immunisation Program (NIP) [23, 24]. A rapid increase in MenW and (to a lesser extent) MenY reversed this trend and incidence increased yearly to 1.5 cases per 100,000 in 2017 [24].

Within Australia, MenW first emerged in 2014 in the state of Victoria, associated with older adults and atypical manifestations (e.g., pneumonia, epiglottitis) [25, 26]. Molecular typing identified P1.5,2:ST11 meningococci; phylogenetic analysis revealed clustering with South American/UK strains [25]. MenW supplanted MenB as the predominant cause of IMD Australia-wide in 2016 [24]. Beginning mid-2017, a multi-jurisdictional (Northern Territory, Queensland, South Australia, Western Australia) MenW outbreak occurred; most cases occurred among Indigenous children in remote Central Australia [27, 28].

In Australia, vaccines are provided mostly through the NIP [23]. Additional vaccines may be funded through state/territory programs [29]. Until 2018, the NIP included MenC vaccines only (administered at 12 months). The NIP was amended in 2018 (MenACWY vaccines replaced MenC vaccines), in 2019 (MenACWY eligibility expanded to include 14-16-year-olds), and
2020 (MenB vaccines funded for Aboriginal and Torres Strait Islander children, certain medical conditions) [29]. Jurisdictions funded additional programs in 2017/2018; South Australia funded MenB vaccines for children/adolescents (2018, ongoing), Victoria funded MenC vaccines for men who have sex with men (MSM) (2018-2019), and multiple jurisdictions funded MenACWY vaccines prior to their NIP inclusion [27-31].

In response to the increase in MenW, and to better understand the diversity of Australian meningococci, the Commonwealth Department of Health, in collaboration with states and territories, trialled a national IMD genomic surveillance initiative in 2017, with voluntary continuation in 2018. This initiative was coordinated by the Microbiological Diagnostic Unit Public Health Laboratory (MDU PHL), the Victorian bacterial reference laboratory. Here we outline the findings of this initiative and highlight valuable aspects of genomic surveillance.

METHODS

Epidemiological analysis

IMD is nationally notifiable; jurisdictions (New South Wales; NSW, Queensland; QLD, South Australia; SA, Tasmania; TAS, Victoria; VIC, Western Australia; WA, Australia Capital Territory; ACT, Northern Territory; NT) must report confirmed/probable cases to the federal health department for inclusion in the National Notifiable Diseases Surveillance System (NNDSS). Laboratory specimens positive for *N. meningitidis* are sent to reference laboratories in each jurisdiction (collectively the National Neisseria Network; NNN). The NNN undertakes national surveillance of *N. meningitidis* and *N. gonorrhoeae*. This study derived IMD notification data from NNDSS and NNN data.

Isolate selection

Australian isolates

During the genomic trial period *N. meningitidis* isolates or sequence data from culture-positive Australian IMD cases were submitted to MDU PHL. Laboratories in QLD, NSW, and WA provided Illumina short-read sequence data. Isolates underwent WGS, quality control (QC), and genomic analyses described below; sequence data underwent QC and genomic analyses.

Samples collected between 1/1/2017 and 23/5/2019 were eligible for inclusion, covering the genomic trial and extended study period (Supplementary Table 1). Isolates from 2019 were opportunistically included to provide additional context. Non-Victorian isolates from eye swabs were included (these jurisdictions classify such samples as having invasive potential).
International sequences

Short-read sequence data (FASTQ) from 1737 international ST-11 isolates were selected for comparison with Australian CC11 isolates (Supplementary Table 2). These isolates were collected between 1974 and 2020 in Africa (n=256, 11 countries represented), Asia (n=10, 2 countries represented), Europe (n=1458, 15 countries represented), North America (n=1, 1 country represented), and South America (n=11, 2 countries represented). Sixty-two isolates were of unknown collection year, 1 isolate was of unknown geographic origin. FASTQ files were preferred over genome assemblies for QC purposes. These were identified through the PubMLST Neisseria database [32] on 3/6/2022 using the search criteria: serogroup/genogroup/capsule=W, clonal complex=ST-11, ENA accession number is not null (accession numbers were used to download sequence data from the Sequence Read Archive). Sequences were downloaded from the National Centre for Biotechnology Information (NCBI) Sequence Read Archive (SRA) using SRA Toolkit v3.0.0 (https://github.com/ncbi/sra-tools). These data were subject to QC and genomic analyses described below.

Whole-genome Sequencing

Genomic DNA from pure isolates was extracted on QIAsymphony using the DSP Virus/Pathogen Mini Kit (Qiagen). Unique dual indexed libraries were prepared using the Nextera XT DNA sample preparation kit (Illumina). Libraries were sequenced on Illumina NextSeq 500/550 with 150-cycle paired end chemistry as described by the manufacturer.

Sequence quality control

Sequences were required to meet the following metrics for inclusion in genomic analyses: average Q-score ≥30, estimated genome size within range of N. meningitidis (expected size ~2.2 MB), average estimated coverage of ≥40, kmer identification of N. meningitidis.

Genomic analysis

Bioinformatics analysis was undertaken using bohra (https://github.com/MDU-PHL/bohra). De novo genome assemblies were used to establish in silico MLST (https://github.com/tseemann/mlst), and in silico serogroup and finetype (https://github.com/MDU-PHL/meningotype). Identification of core genome single nucleotide polymorphisms (SNPs) was undertaken using snippy v4.4.5 (https://github.com/tseemann/snippy). Reference genomes are listed in Supplementary Table 3.

Maximum likelihood phylogenies were generated using IQ-TREE [33] (http://www.iqtree.org/), with a GTR+G4 and 1000 bootstrap replicates. Phylogenies were annotated using R or iTOL (https://itol.embl.de/abo ut.cgi) [34]. Genomic clusters were defined by single-linkage clustering of pairwise SNP distances with two or more sequences, at the 30 SNP level. This cut-off was informed by the SNP distribution of the dataset and supported by literature. Recombination was
assessed using gubbins v3.2.1 (https://github.com/nickjcroucher/gubbins) [35] and was found not to impact cluster interpretation.

RESULTS

To understand the diversity of Australian meningococci from 2017/2018, we analysed 440 isolates from unique IMD cases (280 from 2017, 143 from 2018, 17 from 2019). Dataset representativeness was assessed by comparing the number of isolates with the number of laboratory-confirmed cases reported, noting that isolates require a positive culture (76% of cases (286/374) were culture-positive in 2017; 78% (216/278) were culture-positive in 2018 [24]). Overall representativeness was higher for 2017 than 2018 (74% vs. 51%). Representativeness by jurisdiction varied between years (e.g., NSW was third-most represented in 2017 (76%) but least represented in 2018 (18%)). Serogroup representativeness decreased between 2017 and 2018 for all serogroups but dropped more for MenC (100% to 50%), MenW (86% to 53%) and MenY (83% to 60%) than MenB (60% to 47%). Isolates from 2019 were opportunistic inclusions; representativeness was not calculated. For more detail see Supplementary Table 4.

The Australian N. Meningitidis population is diverse but dominated by a small number of clones.

WGS typing agreed with most PCR-based results and could serogroup 11/21 isolates non-groupable using PCR techniques. Using WGS results, the dataset comprises 181 MenW, 144 MenB, 88 MenY, 16 MenC, 1 MenW/Y, and 10 non-genogroupable (NG) isolates.

WGS typing assigned isolates to 127 genotypes (serogroup: PorA: FetA: MLST: CC), including seventy-seven finetypes (PorA/FetA), seventy-eight STs, and eighteen CCs (Supplementary Table 5). Most isolates in each serogroup were accounted for by a small number of finetypes belonging to one or two CCs. MenB was most diverse, being the only serogroup with three CCs assigned to ≥10 isolates (CC41/44, CC32, CC213) and accounting for 77 genotypes (61%) but only 144 isolates (33%). MenB isolates displayed the most phylogenetic diversity, as indicated by the longer branch lengths relative to MenC, MenY, and MenW (Figure 1).

Whole-genome sequencing can describe within-strain diversity.

The identification of closely related cases may improve public health responses by enhancing understanding of local transmission dynamics and at-risk populations. Using SNP-based analyses, genomic clusters of closely related isolates were identified. All clusters were identified within the three most prevalent CCs; they are described below.
ST-11 Complex

Clonal complex 11 was the dominant CC during 2017-2018 (199/440, 45%), observed mostly in MenW (178/199, 89%) and MenC isolates (15/199, 8%), but also in MenB (1/199, 0.5%), MenY (1/199, 0.5%) and NG (4/199, 2%) isolates (Supplementary Table 5).

Fourteen genotypes were identified in MenW:CC11; most prevalent (111/178, 62%) was W:P1.5,2:F1-1:ST11. Most remaining MenW:CC11 (64/67, 96%) and all NG:CC11 isolates (4/4, 100%) differed from this genotype by just one of PorA, FetA, ST. Most MenW:CC11 clusters (12/15) comprised isolates of genotype W:P1.5,2:F1-1:ST11 (Table 1).

Two MenW:CC11 clusters (W-5, W-9) were noticeably larger (W-5: 17 isolates, W-9: 21 isolates) than other MenW:CC11 clusters (range: 2-8 isolates) (Figure 2, Table 1). All W-5 and almost all W-9 isolates possessed the predominant MenW:CC11 finetype (P1.5,2:F1-1) but did not possess the predominant ST (ST-11). Instead, W-5 comprised ST-12351 (16/17) and ST-12897 (1/17) isolates. Comparatively, W-9 comprised ST-1287 (21/21) isolates. ST-12351 and ST-1287 are single locus variants of ST-11, ST-12897 is a double locus variant. Also of note, cases belonging to W-5 and W-9 were generally younger than those belonging to other MenW:CC11 clusters (W-5: median age 4 years, W-9: median age 6 years, other MenW:CC11 clusters: median age 53 years). W-5 comprised cases from WA (11), SA (3), QLD (2), and NT (1); W-9 comprised cases were from NT (14), QLD (4), and WA (3). By contrast, other MenW:CC11 clusters comprised cases mostly from VIC, NSW, QLD, and TAS.

Two genotypes were identified in MenC:CC11 isolates; most (12/15) were C:P1.5-1,10-8:F3-6:ST-11(CC11) (Supplementary Table 5). These isolates displayed limited phylogenetic diversity and one eight-isolate cluster (C-1) comprising males aged 25-39 from VIC and NSW was identified (Figure 1, Figure 2, Table 1).

ST-23 Complex

The second-most common CC was CC23 (80/440, 18%). CC23 was dominant among MenY (78/88, 89%). In turn, MenY isolates represented 98% of CC23 isolates (78/80); in addition to two NG isolates (2%) (Supplementary Table 5).

Twelve genotypes were identified in CC23 isolates, most (61/80, 77%) were Y:P1.5-1,10-1:F4-1:ST-1655. Six of eight CC23 clusters comprised Y:P1.5-1,10-1:F4-1:ST-1655 isolates (size range: 2-5 isolates). Cases belonging to these clusters were confined to QLD, NSW and VIC; nearly half were aged 65+ (Figure 2, Table 1).

ST-41/44 Complex

Clonal complex 41/44 was the third-most common CC (64/440, 15%). CC41/44 was solely associated with MenB (64/64, 100%) and was the dominant CC among serogroup B (64/144, 44%) (Supplementary Table 5).

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Nineteen genotypes were identified in MenB:CC41/44 isolates. Of these, most genotypes (15/19, 79%) were associated with only one or two isolates. The most prevalent genotype among MenB:CC41/44 isolates was B:P1.7-2.4:F1-5:ST-154 (31/64, 48%), followed by B:P1.18-1,34:F1-5:ST-6058 (8/64, 13%).

Six clusters were identified among MenB:CC41/44 isolates; four of these were within B:P1.7-2.4:F1-5:ST-154. The largest cluster (B-4, 6 isolates) was confined to SA. Isolates from SA appeared in most clusters (4/6, 80%) and comprised approximately half of all cluster-associated MenB:CC41/44 cases (9/20, 45%) (Figure 2, Table 1).

**Whole genome sequencing can reveal the likely origin of strains.**

The superior resolution of WGS enables assessment of diversity among isolates collected across multiple countries or years, potentially revealing clonal origin and evolution. Phylogenetic analysis of Australian MenW:CC11 sequences and a representative set of international CC11 sequences was undertaken (Figure 3).

Clades consistent with the known MenW:CC11 sub-lineages (based on sequence origin) can be observed (Hajj sub-lineage - sequences from Africa, Asia, Europe; South American/UK sub-lineage - sequences from Europe and South America). Most Australian isolates fall into one clade within the South American/UK sub-lineage. This Australian clade also contains a few European sequences however, the collection years are similar to those of the Australian sequences, making it challenging to determine whether these are importation or exportation events. This Australian clade likely represents one or more introductions of highly clonal strains, that underwent subsequent local diversification. Throughout the tree there are several other smaller groups of Australian sequences, indicative of separate introductions that never resulted in significant transmission in Australia.

**DISCUSSION**

This WGS-based study provides a detailed description of the *N. meningitidis* population circulating in Australia during 2017/2018. In line with notification data reported during this period, study isolates were predominantly MenW (n=181) and MenB (n=144), followed by MenY (n=88), and MenC (n=16). Five of 18 CCs accounted for the majority (389/440, 89%) of isolates (CC11; n=199, CC23; n=80, CC41/44; n=64, CC32; n=30). These CCs, bar CC11, were restricted to specific serogroups (CC23; MenY, CC41/44; MenB, CC32; MenB, CC213; MenB, CC11; MenB/C/W/Y).

In the early 2000s, hypervirulent MenW:CC11 strains emerged and MenW rapidly transitioned to a common cause of IMD worldwide [12]. Phylogenetic analyses illustrate that Hajj-related strains drove the initial global increase but subsided by the mid-2000s, with the subsequent rise due to genetically unrelated strains (the South American/UK sub-lineage and its variant strains).
More recently, there have been reports of emerging non-CC11 MenW lineages, e.g., ST-9316 in France, ST-3342(CC865) in the Czech Republic, which may have potential to supplant CC11 strains and alter IMD epidemiology [36, 37].

Australia experienced rapidly rising MenW; notifications inclined steeply between 2014 and 2017 and a multi-jurisdictional outbreak occurred in 2017 [24, 27]. During 2017/2018, CC11 was the dominant MenW lineage (178/181, 98%); remaining isolates were CC22 (2/181, 1%) and undefined/novel ST (1/181, 1%). In some countries, e.g., Canada, CC11 replaced CC22 as the dominant MenW lineage [38]. Almost all Australian MenW:CC11 isolates possessed the PorA P1.5,2 subtype (170/178, 96%), which is widespread and characteristic of all current MenW:CC11 global sub-lineages [20]. Based on WGS data, it appears that the increase in MenW in Australia was caused mainly by non-Hajj sub-lineages, in keeping with many countries where MenW emerged post mid-2000s. The data also indicated multiple MenW introductions to Australia, most of which did not result in significant transmission, plus limited phylogenetic diversity within Australia. Additionally, we observed two large genomic clusters (W-5, W-9) with associated metadata similar to the epidemiology of reported multi-jurisdictional MenW outbreaks in remote central Australia in 2017 [27, 28]. Together, this data is consistent with recent emergence, effective establishment, and rapid transmission in a naïve geographical area.

Among MenC isolates, CC11 was dominant (15/16, 94%); the one remaining MenC isolate was CC103. This predominance of CC11 among MenC is commonly observed internationally [20, 37, 39, 40]. We identified relatively few MenC isolates, reflective of the low incidence of MenC in Australia since routine MenC immunisation began in 2003 [23]. MenC isolates displayed little genetic variation and most MenC:CC11 isolates (8/15, 23%) formed a genomic cluster (C-1) comprising cases from men aged 25-37 from VIC and NSW. All C-1 isolates were C:P1.5-1,10-8:F3-6:ST-11. Since 2001, IMD outbreaks among MSM have been reported worldwide, mostly in European and North American cities (e.g. Los Angeles, New York City, Chicago, Toronto, Berlin, Paris) [41]. These were predominantly caused by MenC:CC11 meningococci of the same genotype as C-1 isolates [42]. The metadata associated with C-1 isolates is consistent with the epidemiological profile (age, sex, geography) of a 2017 MenC outbreak reported among MSM in Melbourne, Victoria [31]. C-1 may correspond to this outbreak, though matched epidemiological data would be needed to confirm this.

MenY infections were uncommon in Australia before 2014, but increased steadily between 2014 and 2017, mainly in QLD, NSW, and VIC [24]. In this dataset, MenY isolates were predominately CC23 (78/88, 89%) and almost all CC23 isolates were MenY (78/80, 98%). Clusters identified among MenY:CC23 isolates were mostly associated with older adults in QLD, NSW, and VIC. Together, this suggests a pattern of clonal expansion of CC23, likely concentrated in eastern Australia. This trend – emergence and expansion of MenY:CC23 – has been observed in many countries since the early 2000s [37, 40, 43].

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Our MenB population displayed high heterogeneity relative to MenC/W/Y, alone grouping into several CCs and displaying the most phylogenetic diversity. This aligns with many other developed countries who also report a high degree of genetic diversity in their MenB populations [37, 40, 44]. The most prevalent CCs in our dataset were CC41/44 (64/144, 44%), CC32 (30/144, 21%), and CC213 (16/144, 11%); the same CCs responsible for most endemic MenB globally between 2000 and 2015 [45]. Both CC41/44 and CC32 warrant monitoring due to their proven epidemic potential; C41/44 was responsible for prolonged outbreaks in the Netherlands and New Zealand, while CC32 has caused major outbreaks in Norway, Spain, Cuba, the USA, and France [44]. CC213 also warrants monitoring due to its increasing prevalence in some countries and low predicted 4CMenB vaccine coverage [46]. In Australia, MenB predominated from 2002 to 2015, though overall incidence decreased annually between 2001 and 2016, except in South Australia where most cases were caused by the 'New Zealand epidemic strain' (PorA P1.7-2,4; CC41/44) [24, 30]. The predominance of South Australian isolates within B:P1.7-2,4:F1-5:ST-154(CC41/44) clusters, the largest of which was confined to SA (B-4, n=6), suggests protracted transmission of this strain in SA during 2017/18.

This study underlines the value of routine and nationalised genomic surveillance of IMD. Interpretation of epidemiological IMD data alone is challenging due to low disease incidence and the predominant role of asymptomatic carriage in transmission, which often results in large or variable time periods and/or geographic distances between observed cases in a transmission chain [47]. Additionally, the dominance of a small number of lineages that may be seeded through multiple introductions may give rise to separate transmission chains with identical genotypes. As demonstrated in this study, WGS provides the additional resolution required to disentangle these issues though cluster identification, description of the overall bacterial population, and identification of new, expanding, or retracting clones.

A growing number of countries use genomic surveillance of N. meningitidis to inform and evaluate meningococcal vaccination programs. Traditionally, the evaluation of such programs has broadly involved predicting counterfactuals based on pre-immunisation incidence, while considering variables such as vaccine efficacy and coverage [48]. Determining vaccine efficacy against MenB however has proven difficult due to its low incidence and high antigenic variability (the MenB vaccines target surface antigens rather than the polysaccharide capsule and do not cover all MenB strains) [49, 50]. DNA-based typing schemes such as the genetic Meningococcal Antigen Typing Scheme (gMATS) and Bexsero Antigen Sequence Typing (BAST) have been developed to enable prediction of MenB vaccine coverage [50]. Insights generated through other methods of genomic surveillance, such as those outlined in this study, could further augment the evaluation of meningococcal vaccination programs. For example, if genomic surveillance identified highly related cases in the target serogroup following a change to vaccine strategy, this could indicate new or ongoing transmission regardless of any observed reduction in the number of infections. As a result, it may be possible to identify sub-populations or areas in need of targeted vaccination or other interventions.

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To fully realise the benefits of genomic surveillance, re-establishing the collation of a highly representative dataset (including current and historical samples) will be required. In this context, representativeness refers to the proportion of cases isolated and sequenced, noting that this requires a positive culture. Lower representativeness, decentralised analyses, and inconsistent coverage leads to the omission of intermediary isolates, which reduces the ability to identify transmission chains or to identify their full scope, potentially delaying the recognition of, or response to, epidemic activity. For example, in this dataset, 2018 cases had a lower proportion sequenced than 2017 cases, which may explain why proportionally fewer isolates clustered. However, this may also have been due to reduced epidemic activity and increased sporadic disease.

Our findings demonstrate several benefits offered by routine, nationalised IMD genomic surveillance, but also stress the necessity of comprehensive participation nationwide and greater integration of clinical and epidemiological data. This study is strengthened by cross-jurisdictional representation during a year characterised by MenC and MenW outbreaks. Lower representation during 2018, captured via voluntary participation, highlights the need for sustainable funding and formalisation of systems to ensure representative datasets and robust analyses going forward. Future integration of enhanced epidemiological information, such as travel history, clinical outcome data, vaccination status, and contact information, would augment genomic insights and strengthen IMD surveillance.

References


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AUTHOR NOTES

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Author contributions

ES, KH, PA, BH designed the study; ES, KH performed analysis; KH, PA, BH, CL, CG, JMV, ML, AVJ, DJS, LL provided critical input and discussions; ES, KH, PA, BH wrote first draft; and all authors contributed to the final manuscript.

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Potential Conflicts of Interest

No author has any conflict of interest to declare.

Submission declaration

This manuscript has not been published elsewhere and is not under consideration by another journal. A previous version of this research was presented as an oral presentation at the Communicable Disease Control Conference 2019, Canberra, Australia, 2019, under the title Genomic surveillance of Invasive Meningococcal Disease (IMD) in Australia, 2017 – 2018.

Ethics statement

Ethical approval was received from the University of Melbourne Human Research Ethics Committee (study number 1954615).

Patient consent statement

A patient consent waiver was included in the above local ethics committee approval.

Data availability

Sequencing data will be deposited in the NCBI Sequence Read Archive (SRA) and made publicly available by the time of publication.
FIGURE LEGENDS

Figure 1. Maximum likelihood phylogenetic tree for all serogroup B, C, W, Y, W/Y and non-genogroupable (NG) invasive meningococcal disease isolates submitted to MDU PHL, collected between 1 January 2017 and 23 May 2019. Isolate metadata is shown by the coloured rings surrounding the tree. Isolate serogroup is displayed by the inner ring, and jurisdiction by the outer ring (see legend for colour scheme).
Figure 2. Dot plot displaying genomic clusters identified among invasive meningococcal disease isolates submitted to MDU PHL, collected between 1 January 2017 and 23 May 2019. Cluster designation is displayed on the y-axis; clusters are grouped according to their predominant serogroup. The coloured dots correspond to isolates and the size of the dot is relative to the number of isolates collected on a given day (date of collection is displayed on the x-axis). Dots are coloured according to jurisdiction (see legend for colour scheme).
Figure 3. Maximum likelihood phylogenetic tree for Australian and international serogroup W Clonal Complex 11 isolates. Australian isolates were submitted to MDU PHL and collected between 1 January 2017 and 23 May 2019, and represent instances of invasive disease. International isolates were identified through the PubMLST *Neisseria* database and represent instances of carriage and invasive disease. Isolates’ continent of collection is displayed by the coloured ring surrounding the tree (see legend for colour scheme).

Table 1. Description of genomic clusters detected within invasive meningococcal disease isolates submitted to the MDU PHL, collected between 1 January 2017 and 23 May 2019.
<table>
<thead>
<tr>
<th>Genomic cluster</th>
<th>Number of isolates</th>
<th>Age; mean (range)</th>
<th>Duration (days)</th>
<th>Average time between samples (days)</th>
<th>WGS Genotype (number of isolates)</th>
<th>Jurisdictions (number of isolates)</th>
</tr>
</thead>
<tbody>
<tr>
<td>B-1</td>
<td>2</td>
<td>3 (2-3)</td>
<td>0</td>
<td>0.0</td>
<td>B: P1.18-1,34: F1-5: ST-6058 (cc41/44) (2)</td>
<td>QLD</td>
</tr>
<tr>
<td>B-2</td>
<td>3</td>
<td>21 (15-28)</td>
<td>362</td>
<td>120.7</td>
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