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The global meningitis genome partnership

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SUMMARY

Genomic surveillance of bacterial meningitis pathogens is essential for effective disease control globally, enabling identification of emerging and expanding strains and consequent public health interventions. While there has been a rise in the use of whole genome sequencing, this has been driven predominately by a subset of countries with adequate capacity and resources. Global capacity to participate in surveillance needs to be expanded, particularly in low and middle-income countries with high disease burdens. In light of this, the WHO-led collaboration, Defeating Meningitis by 2030 Global Roadmap, has called for the establishment of a Global Meningitis Genome Partnership that links resources for: *N. meningitidis* (Nm), *S. pneumoniae* (Sp), *H. influenzae* (Hi) and *S. agalactiae* (Sa) to improve worldwide co-ordination of strain identification and tracking. Existing platforms containing relevant genomes include: PubMLST: Nm (31,622), Sp (15,132), Hi (1935), Sa (9026); The Wellcome Sanger Institute: Nm (13,711), Sp (> 24,000), Sa (6200), Hi (1738); and BMGAP: Nm (8785), Hi (2030). A steering group is being established to coordinate the initiative and encourage high-quality data curation. Next steps include: developing guidelines on open-access sharing of genomic data; defining a core set of metadata; and facilitating development of user-friendly interfaces that represent publicly available data.

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Introduction

Meningitis, and related invasive bacterial infections such as bacteraemic pneumonia and sepsis, are devastating diseases that represent a major global health challenge.[1,2] While successful immunisation programmes against three of the main bacterial causes, *Neisseria meningitidis* (Nm, the meningococcus), *Streptococcus pneumoniae* (Sp, the pneumococcus), and *Haemophilus influenzae* type b (Hib), have enabled great progress to be made, the burden remains high with progress substantially behind that of other infectious diseases. According to one estimate, between 1990 and 2017 child (under five) meningitis deaths fell by just 53%, compared to 70% for diarrhoea, 87% for measles, and 93% for tetanus.[3]

In the African meningitis belt, large epidemics of meningococcal disease occur periodically. Although there have been dramatic declines in Nm serogroup A (NmA) disease following successful MenAfriVac® mass vaccination campaigns, the threat of both endemic and epidemic disease caused by non-A Nm and Sp persists[4], with Africa as a whole still experiencing the greatest global burden of bacterial meningitis. Epidemiological data underlying global burden estimates, particularly in Asia, remain incomplete, however.[5] Nucleotide sequence-based typing, in particular whole genome sequencing (WGS), has important applications for tracking and responding to endemic and epidemic disease.

WGS to elucidate the origin and spread of new strains for public health management

Outbreaks of Nm serogroup C (NmC) disease were rare in the meningitis belt until 2013, when North-Western Nigeria experienced localised outbreaks caused by a novel NmC strain (the ST-10217 clonal complex; cc10217).[6] In 2015, this strain caused a severe epidemic in Niger[7] and, in 2017, the largest ever known outbreak of NmC disease in Nigeria, with over 14,000 suspected cases.[8,9] The analysis and comparison of WGS data successfully elucidated the origin and spread of this new strain that emerged after the acquisition of virulence genes by a non-encapsulated, non-virulent ancestor.[10] Metagenomic approaches have since revealed that NmC cc10217 has spread outside the meningitis belt, causing an outbreak associated with a funeral in Liberia in 2017.[11]

Genomic approaches were also able to resolve Nm serogroup W (NmW) ST-11 clonal complex (cc11) isolates into multiple strains within two divergent sublineages: (i) the 'Hajj-strain sublineage', (named after a constituent strain that caused a global outbreak among Hajj pilgrims in the early 2000s); and, (ii) the 'South American-strain sublineage' that recently spread from South America to Europe and beyond.[12] Identification of the emergence and expansion of the South American-strain sublineage in the United Kingdom (U.K.) led to introduction of MenACWY conjugate vaccination for adolescents. A novel, seemingly more virulent, variant, the '2013-strain' has since expanded into Europe, Australasia and North America[13] with consequential vaccine policy changes in Ireland, the Netherlands, Switzerland and Australia.

WGS also has important applications in the development of vaccines to control endemic disease. Indeed the identification of the recombinant antigens used in Bexsero®, a protein- based vaccine that targets Nm serogroup B (NmB) disease, marked the first use of genomics for vaccine antigen discovery through a 'reverse vaccinology' approach.[14] Prior to this, broadly protective MenB vaccine development was hindered by the fact that NmB polysaccharide is poorly immunogenic and has possible autoimmune effects, due to its similarities with human glycoproteins[15], while candidate outer-membrane vesicle vaccines offered limited protection. Two recombinant protein MenB vaccines, Trumenba® and Bexsero® are currently licensed for use in multiple countries worldwide, with the latter routinely used in the U.K., Ireland, Lithuania, Andorra, San Marino and South Australia[16], while regions of Italy routinely use both Trumenba® and Bexsero®. While these vaccines are expected to provide broad coverage of circulating NmB strains[17,18], on-going global genomic surveillance is required to provide evidence of vaccine impact, document strain coverage, monitor vaccine escape and to facilitate development of next generation vaccines.[19,20]

WGS has an important role to play in the surveillance of invasive pneumococcal disease (IPD), which includes meningitis, bacteraemic pneumonia, and sepsis and remains a leading global cause of mortality, especially among children.[21–23] Under immune selective pressure[24] the pneumococcus can escape pneumococcal conjugate vaccine (PCV) control through serotype replacement. The increase of non-vaccine serotypes, concomitant with a decrease in vaccine serotypes,[25] highlights the importance of monitoring serotypes circulating pre- and post-PCV introduction to enable invasive non-vaccine pneumococcal serotypes to be rapidly identified.[17] WGS has enabled far greater insight into this process.

Genomics has been used to investigate serotype replacement after the introduction of both the original (PCV7) and higher valency (PCV10 and PCV13) vaccines, and emerging non-vaccine serotypes have been found to vary extensively among countries. Using genome-wide variation, an international definition of pneumococcal population structure 'Global Pneumococcal Sequence Clusters (GPSCs)' has been developed and enables the production of pneumococcal datasets, independent of previous definitions of lineages.[26] Using this cluster definition, it was revealed that, post-PCV13 introduction, the top emerging cluster, GPSC3, expressed serotype 8 in South Africa, but 33F in Israel and the United States (U.S.).[27] This demonstrates how species-wide approaches can enable the evolution of the pneumococcus to be better tracked, beyond the limits of serotype.

At the time of writing, comprehensive genome studies investigating the meningococcus and the pneumococcus were available, but such studies for *H. influenzae* were more limited. It is important to ensure that this pathogen is adequately examined, as, despite dramatic reductions in Hib disease following global vaccine implementation, with 192 countries having introduced a universal Hib vaccination programme by 2019[28], *H. influenzae* meningitis caused by other types still occurs.[29] For example, increasing rates of invasive disease due to *H. influenzae* type a (Hia) have been described in indigenous populations in northern Canada, Alaska, the southwestern U.S. and Australia, with an additional study indicating that *H. influenzae* strains are able to capsule-switch or modulate the expression of the type b capsule.[30–32] Unencapsulated non-typeable *H. influenzae* strains (NTHi) are also increasingly associated with invasive disease, including bacteraemic pneumonia. These organisms represent a growing challenge in healthcare settings, particularly as many are antibiotic resistant.[33]

As of mid-2020 *Streptococcus agalactiae* (GBS), a leading cause of neonatal meningitis and sepsis, was not yet vaccine-preventable, although vaccines were in development.[34–36] An increasing number of WGS studies investigating GBS were becoming available, including some from low income countries, indicating a growing awareness of this infection alongside the benefits of WGS data in understanding the epidemiology of this pathogen.[37]

Consequently, continued surveillance and characterisation of bacteria associated with meningitis remain a priority, if the global burden of meningitis is to be reduced. The availability of sequence-based data will play an essential role in strengthening surveillance of disease types and enabling interventions to be appropriately targeted.

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Strengthening global genomic surveillance: a global meningitis genome partnership

The need for a Global Meningitis Genome Library (GMGL) was identified in May 2017 at a meeting of international experts hosted by Wilton Park,[38] and led by Meningitis Research Foundation (MRF) in collaboration with the World Health Organization (WHO). This meeting first outlined a global vision to defeat meningitis and called for an action plan in line with the United Nations Sustainable Development Goals (SDGs).[39,40] Subsequent consultations led to the establishment of a Global Meningitis Genome Partnership (GMGP) to link resources for the four leading causes of acute bacterial meningitis, with the aim of improving worldwide co-ordination of strain identification and tracking, and enabling the public health benefits of WGS to be delivered.

A Technical Task Force[41] co-ordinated by WHO developed a Global Roadmap to Defeat Meningitis by 2030, which will be submitted for consideration at the World Health Assembly in 2020. The roadmap focuses on the main causes of acute bacterial meningitis that are already vaccine-preventable or may be in the foreseeable future: *N. meningitidis, S. pneumoniae, H. influenzae,* and *S. agalactiae*. Sequence-based data are one key element to the global vision to defeat meningitis, and therefore the importance of the establishment of a GMGP functional for the four pathogens has been highlighted by the Task Force. The terminology 'partnership' (within which libraries, or data repositories and platforms play a crucial part), reflects the collaborative approach required for the work that needs to be done.

This article provides an account of the current status of meningitis genomics globally and, in particular, focuses on: (i) reviewing existing platforms that facilitate the submission and analysis of WGS data; (ii) describing the efforts of existing global projects and partners focused on enhancing surveillance and increasing our understanding of these bacteria, particularly in resource-poor settings; and, (iii) discussing the complexities associated with ensuring globally representative datasets can be obtained and made publicly available.

Examples of existing meningitis pathogen genome libraries and analysis platforms

PubMLST

The open-access PubMLST.org website and its underlying web-based software platform, the Bacterial Isolate Genome Sequence Database (BIGSdb), include all levels of sequence data enabling a hierarchical approach to studying microbial species from single gene fragments, to functional genes grouped into schemes, up to full genome comparisons.[42] The integration of isolate characterisation, linking genotypes and genetic variation with provenance and phenotype data, addresses a wide range of functional questions, from population biology to antimicrobial resistance (AMR) and vaccine formulation, all of which can inform public health policies.

At the time of writing the PubMLST databases hosted over 100 species and/or genera, including curated databases for the four meningitis pathogens. This included > 30,000 *N. meningitidis* genomes, > 15,000 *S. pneumoniae* genomes, > 9000 *S. agalactiae* genomes and 1900 *H. influenzae* genomes, predominantly submitted from high-income regions or international centres.

PubMLST is based on gene-by-gene population consensus annotation, where genes are identified and their variation catalogued systematically.[43] As a result, the genetic variation of every gene in every bacterial genome can, in principle, be linked with provenance and phenotypic information.[44] Users can perform a suite of analyses, including: evolutionary analyses, bacterial species classification, disease surveillance, phylogeography and outbreak analyses. The provision of a common nomenclature, *i.e.* a universal language to describe bacterial strains and lineages, is integral to its success, enabling portable use of this infrastructure and allowing global laboratories and public health agencies to compare isolates of interest, including foodborne pathogens. As a result, enteric pathogen outbreaks have been detected and halted through quick recognition of the same strain from different sources.[45–47]

In addition to providing curated genomes, visual analytic tools enable users to assess WGS genetic relatedness with integrated, third-party software, such as: (i) GrapeTree[48] and/or PhyloViz[49]; (ii) Interactive Tree of Life (iTOL)[50]; and (iii) MicroReact, which respectively enable the creation of minimum spanning trees, phylogenetic trees and the visualisation of genomic epidemiology and phylogeography (Fig. 1).[51]

The PubMLST.org *Neisseria* site is one of the largest single databases within PubMLST (Fig. 2).[52] Data are actively curated with genomes submitted by users internationally. Additionally, curators sometimes assemble genomes from short read data from the European Nucleotide Archive (ENA) and associate these with published metadata. Curated data are made freely available and accessible via the website or application programming interfaces (APIs). This enables third party analysis sites to interconnect with the database, facilitating open sharing of publicly available genomic data. Through APIs, comparisons can be made between private data, containing clinically sensitive information, and publicly available data, enabling individual databases to act synergistically.[53]

The PubMLST *Neisseria* database contains several datasets, including comprehensive meningococcal genome libraries for England and Wales (July 2010-present),[54] Scotland (2009-present) and Ireland (2010–2014); however, it is not yet globally representative. As of March 2020, 127 countries had submitted meningococcal isolates with sequence data but only 97 countries had submitted meningococcal genomes and the number of genomes submitted ($n = 31{,}622$) was approximately half of the number of isolates ($n = 60{,}776$); the proactive collection and curation of comprehensive and representative country-specific genome data from around the world could remedy this.

The PubMLST.org *S. pneumoniae* database contains over 15,000 genomes of which over 9000 were included in the first version of the Pneumococcal Genome Library. The library aims to provide users with assembled pneumococcal genome data and corresponding metadata, as well as an assessment of genome quality among published data. It was developed following a review of articles with at least one pneumococcal genome; 979 articles were screened on PubMed, and 173 peer-reviewed publications identified, of which 97 (56%) provided retrievable pneumococcal genomic data without discrepancies (*e.g.* missing or incorrect genome sequence accession numbers). Upon first release, the library contained data from 92 publications, from 41 countries collected between 1916 and 2018, representing > 90 serotypes. Efforts are underway to resolve discrepancies in the remaining 76 publications and enable upload of the associated genomes.

BIGSdb is also used by the Institut Pasteur, Paris to run their 18 databases of MLST and genome-based typing schemes. The platform provides reference nomenclatures for microbial isolates, primarily intended for molecular epidemiology of pathogens of public health importance, detection of virulence and AMR genes, and for population biology research.

The Wellcome Sanger Institute

The Wellcome Sanger Institute has active research for a number of meningitis-causing pathogens.[55] Datasets represent broad geographic and temporal ranges that are of value for evolutionary and epidemiological analyses, but also include densely sampled collections that elucidate patterns of pathogen transmission and spread.

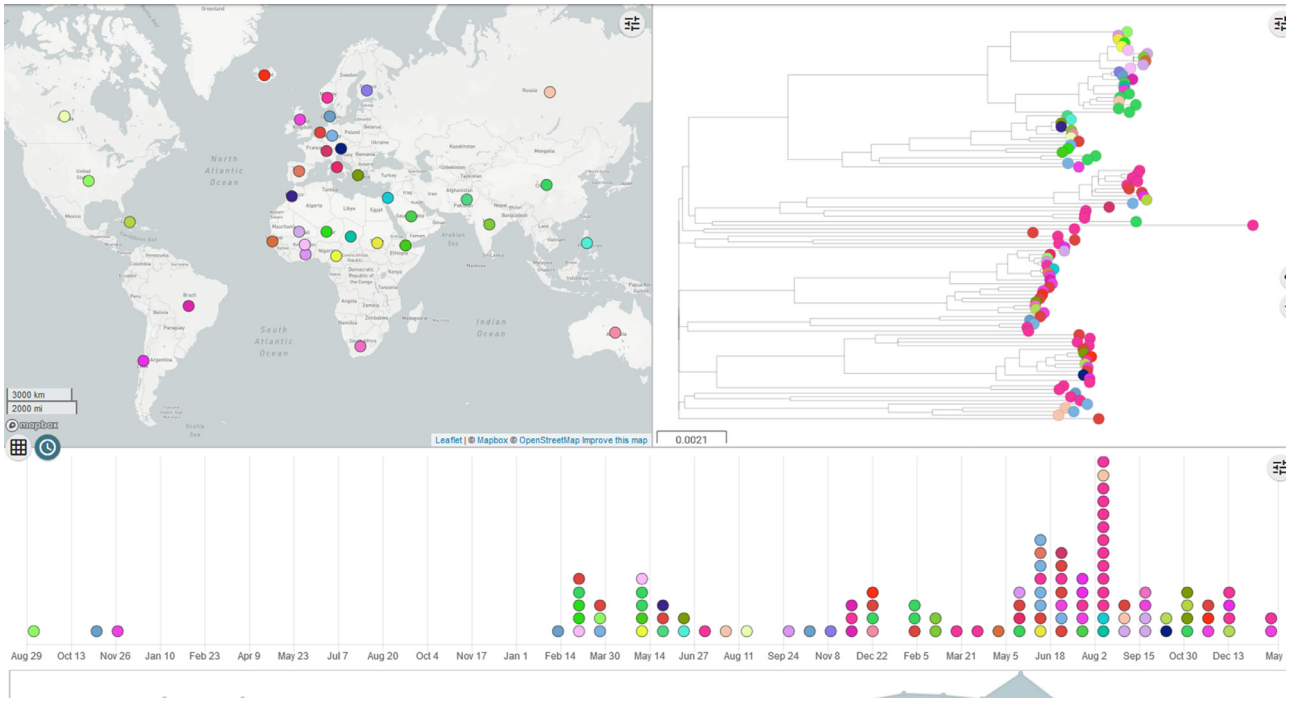

Fig. 1. Phylogeographic, temporal, and phylogenic visualisation tool Microreact available through the PubMLST.org website. Collection of 107 characterised meningococcal isolates chosen to be representative of disease globally in the latter half of the 20th century.

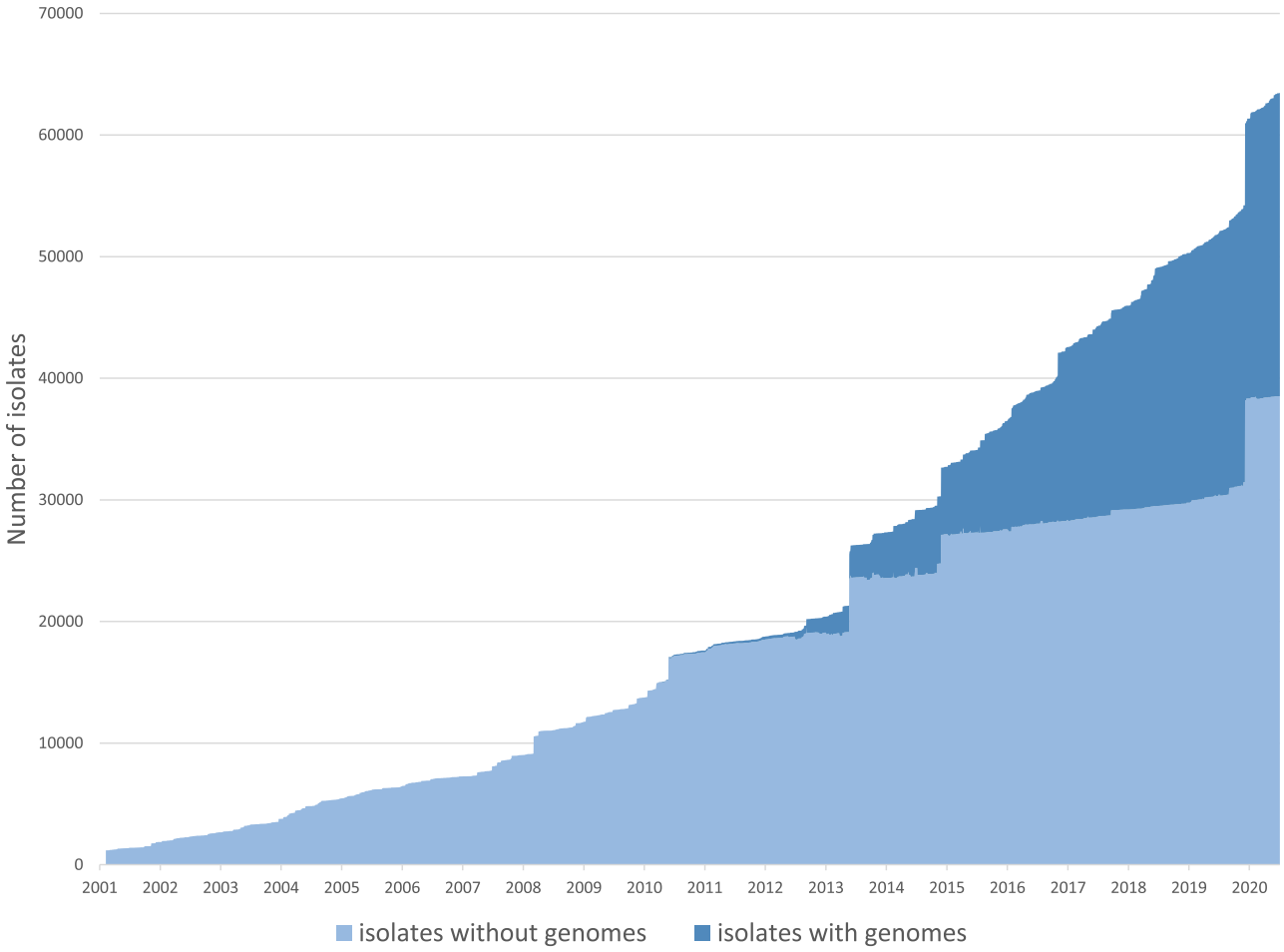

Fig. 2. PubMLST Neisseria database: isolate and genome submissions over time.

Databases of all publicly available genomes are compiled and infrastructure is being developed to enable public access to data and analysis tools.

A data collection for *Neisseria* genome analyses has been established that gathers all publicly available *Neisseria* WGS data from the ENA and all available associated metadata, accessed from the PubMLST.org *Neisseria* database and the published literature. This has generated a collection of genomes from 32,538 isolates, from 77 countries, collected between 1937 and 2014. Of these, 16,164 are Nm isolates, with metadata available for 13,711. However, a significant proportion of the Nm isolates are from the U.K. ($n = 4256$) and other resource-rich countries; therefore some effort is still necessary to establish truly global collections of *Neisseria* isolates.

The Sanger pneumococcal database includes the Global Pneumococcal Sequencing (GPS) project, which aims to assess the risk of pneumococcal vaccine evasion by sampling population genomic evolution before and after vaccine implementation, to inform future vaccine and surveillance strategies.[56,57] The GPS project has generated over 24,000 high-quality genomes (3105 from meningitis cases) from 58 countries, with an emphasis on low- and middle-income countries (LMICs) where disease burden is highest. Extensive metadata for 13,454 of the genomes and interactive visualisations are available via Pneumogen.net[58] and Pathogenwatch.[59]

A global genomic survey of GBS ('JUNO') is now underway, aiming to provide data and insights to inform vaccine strategies and elucidate the routes of infection leading to infant mortality. Like the GPS project, it will focus on training partners from LMICs to increase access to genome data analysis, and 26 founding partner institutions from 12 countries (mostly African countries) have already agreed to participate in this initiative.

The Bacterial Meningitis Genome Analysis Platform

In addition to global genome collections, platforms customised for country specific needs have been developed, including the U.S. Centers for Disease Control and Prevention (CDC) Bacterial Meningitis Genome Analysis Platform (BMGAP). BMGAP is an automated genomic analysis platform focused on *H. influenzae* and *N. meningitidis* that streamlines workflows and reduces analysis time.

BMGAP relies on PubMLST nomenclature and many functions overlap with PubMLST; however, clinical data access is tightly restricted, requiring state permission, and access to BMGAP requires user registration. Due to the low burden of disease in the US, even anonymised data may be traceable to an individual if precise details of time, place, and clinical data are published with genomes; therefore, clinical data sharing is closely regulated, often requiring additional approval. However, U.S. sequencing data from BMGAP can be made publicly available in PubMLST, in compliance with CDC data-sharing policies, with strictly limited access to sensitive metadata and epidemiological data, to enable the systems to cross-talk.

Enhancing country representation and access through global partner support

Current bacterial meningitis genome repositories are over-representative of the locations where users already have capacity and access to sample collections and understand the benefits of WGS, or where users are collaborating with those who can obtain, culture and sequence patient samples. As a result, sequences from countries important in the emergence and spread of strains, but who lack capacity, resources, and/or funding, or who are not authorised to share data with new partners or collaborators, may be absent from current collections.

To develop globally representative genome repositories, a proactive approach which utilises partner engagement is needed. WHO is committed to helping promote the benefits of WGS through regional and country links, and through existing networks such as the Invasive Bacterial Vaccine-Preventable Diseases (IB-VPD) global surveillance network.[60]

The Global Meningococcal Initiative (GMI)[61] also has an important role to play in promoting the benefits of WGS and participation in a GMGP among its collaborators. The GMI is a multidisciplinary expert group, with members across 37 countries. It aims to prevent invasive meningococcal disease worldwide through education, research, and international collaboration. The role of the GMI is particularly important in regions such as Eastern Europe, Asia and the Pacific where formal meningitis surveillance is less established.[62] A GMI meeting in China fostered a China-U.K. collaboration for the characterisation of a collection of Chinese NmW isolates[63] that were shown to be distinct from the Hajj- and South American-strain sublineages previously described.[64] The collaboration further documented the disappearance of NmA and the emergence of new NmB clones. The GMI is currently engaging with Eastern European countries to investigate the persistence of NmA disease in Kazakhstan.[65]

A further approach to engaging with countries not currently submitting to WGS repositories is through funding time-limited projects that establish partnerships between countries that have capacity and expertise in DNA extraction and sequencing, with those that do not. Data ownership and control over the level of metadata shared would reside with the corresponding country, owing to the codes of practice to be established regarding use of repositories and publication of data. Projects such as the Meningitis Research Foundation-Meningococcal Genome Library (MRF-MGL), which includes comprehensive meningococcal genome libraries for England, Wales and Scotland, have been very successful in establishing representative epidemiological collections of isolates, which have continued to be maintained and updated (Fig. 3a, b).[66]

Pneumococcal African Genomes (PAGe) consortium

A project-based approach can also successfully deliver training and capacity building. The Pneumococcal African Genomes project (PAGe) is a consortium led by the Malawi-Liverpool-Wellcome Trust, with primary partners in Niger, South Africa, The Gambia, and the U.K. PAGe focused on genomic analyses of the pneumococcus across Africa, collecting over 800 isolates, sequenced at the Wellcome Sanger Institute and establishing collaborations with 38 countries. In addition to encouraging dialogue between African sites, this project trained bioinformaticians in centres in The Gambia, Malawi and South Africa. This enabled the analysis of data at a local level, and revealed that serotype 1 in West Africa is distinct from serotype 1 across the rest of the continent.[67–71] Furthering epidemiological understanding of this serotype, one of the major causes of life threatening IPD in sub-Saharan Africa, marks an important contribution towards vaccine target discovery and development.

Molecular Epidemiology for Vaccination Policy

Molecular Epidemiology for Vaccination Policy (MEVacP) is another example of a project-based approach that aims to improve global public health by enhancing the diagnosis and surveillance of bacterial meningitis caused by meningococcus, pneumococcus, *H. influenzae* and GBS, through building networks in low-income countries, with an initial emphasis on Africa. The aim of this project, funded by National Institute for Health Research (NIHR) and led by the University of Oxford, is to improve the characterisation and visualisation of outbreaks across the meningitis belt and inform public health vaccination policies, through development

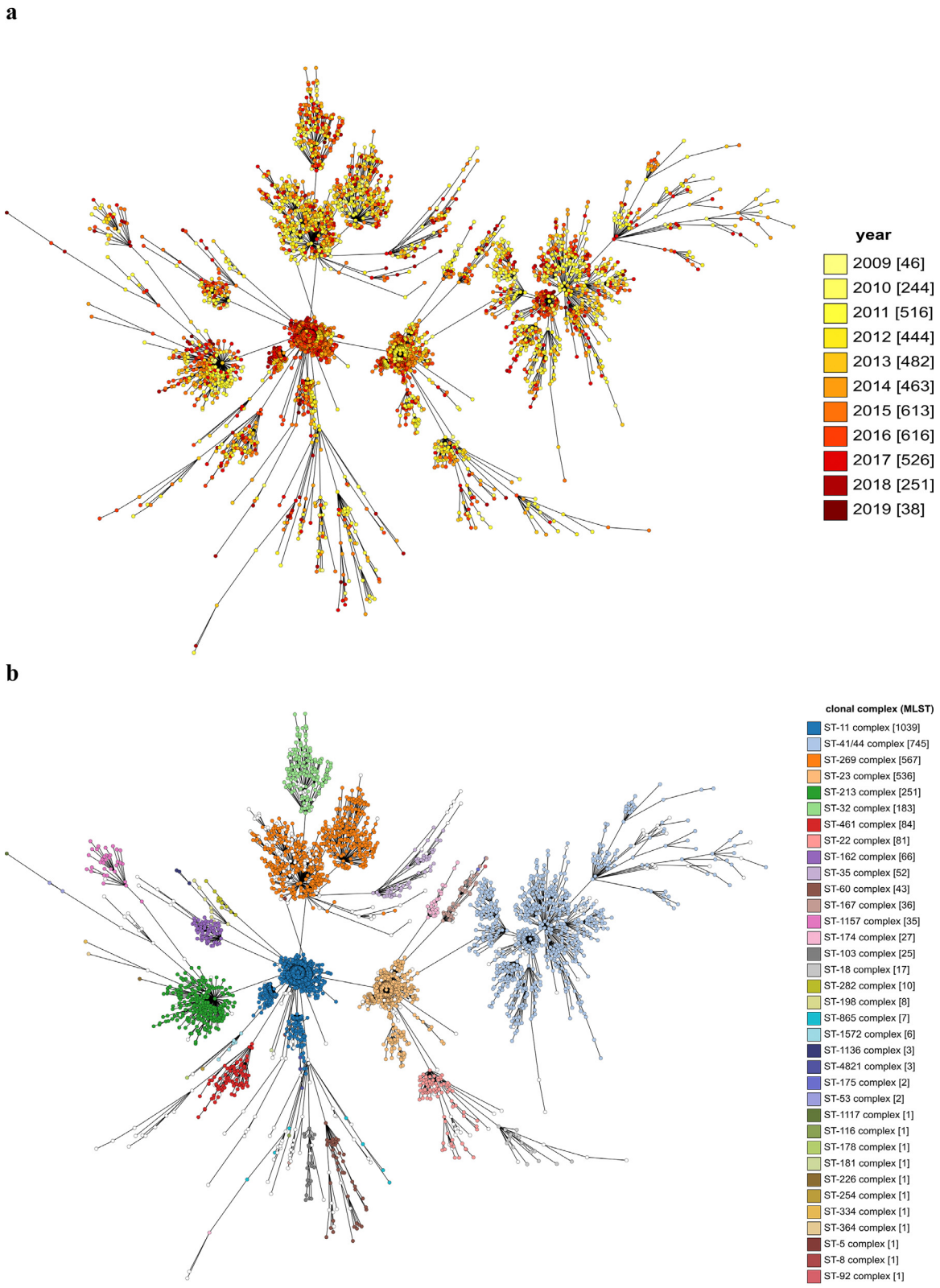

Fig. 3. Meningococcal genomes submitted to the PubMLST Neisseria database, as part of the U.K. Meningococcal Genome Library characterised by core genome MLST (cgMLST), 1605 loci, from 2009 to late 2019 ($n = 4242$); (a) coloured by year; (b) coloured by clonal complex.

Table 1
Meningococcal meningitis in 10 African countries, 2018 [Source: WHO weekly bulletin][76].

Countries	# Meningitis suspected cases	# CSF performed	# Pos	# Nm pos	%CSF performed	% Nm pos	% pos
Benin	322	320	7	1	99%	0.3%	2%
Burkina Faso	2421	1590	211	41	66%	3%	13%
Cameroon	1060	111	5	2	10%	2%	5%
CAR	467	699	37	3	150%	0%	5%
Chad	401	304	102	34	76%	11%	34%
Ghana	987	910	89	39	92%	4%	10%
Mali	755	707	126	13	94%	2%	18%
Niger	1496	1151	543	447	77%	39%	47%
Nigeria	4516	804	310	257	18%	32%	39%
Togo	683	1679	38	3	246%	0.2%	2%
All countries (25)	**20,843**	**8650**	**1531**	**850**	**42%**	**10%**	**18%**

and implementation of a PubMLST-associated web-based platform 'African Meningitis Epidemiology in Real Time' (AMERT). AMERT will operate a peer-to-peer private website similar to the European counterpart, EMERT[72], whereby data in the system are only available to submitters. With restricted access, reference laboratories are reassured that data will remain private, which encourages submissions to be made. Once published in journals, the data can be made publicly available.

Strengthening country laboratory capacity

Currently, molecular surveillance in LMICs is largely driven by expert international centres based in richer countries. In general, LMICs frequently lack laboratory capacity, and have limited access to bioinformatic tools and support, thus limiting opportunities for using sequence-based approaches and the interpretation of sequencing data to identify the causative agents of disease.[60] Existing capacity needs to develop from a reliance on external support, towards surveillance that is directed and delivered by the region in the short term and by individual countries in the long term. To facilitate this, national and regional laboratories with capacity to perform molecular surveillance need to first be identified, and connected, before progress can be made at a country level. However, to decentralise capacity to the country level, greater resource and technical support is required at all levels to achieve sustainable molecular surveillance based on WGS. The rapid development of the technology and the supporting software may assist in this process.

The WHO Collaborating Centers (CCs), the Norwegian Institute for Public Health, the U.S. CDC, the Institut Pasteur Paris, the Medical Research Council Unit The Gambia at the London School of Hygiene and Tropical Medicine (MRCG at LSHTM), and the Global Reference Laboratory for the IB VPD Surveillance Network at the U.S. CDC have laid the foundation for global meningitis genome surveillance by strengthening country capacity for surveillance, laboratory diagnostics and outbreak investigation, particularly in Africa. However, the recovery of isolates from epidemic areas of the African meningitis belt remains low.[73,74] Data from the WHO weekly bulletins show a low but rising proportion of reported cases with cerebrospinal fluid (CSF) samples obtained and analysed, from ~10% to nearly 40% over the past 13 years. While there has been some improvement, still in very few of the analysed CSF samples is the pathogen identified and isolate preserved (Fig. 4).[75] In 2018, 7.4% of the cases had an identified aetiology compared to 2.4% in 2007. Substantial variation among countries is also seen regarding the proportion of cases that have lumbar punctures performed and pathogens isolated (Table 1).[76]

The transfer of clinical specimens to national and international laboratories where sequencing can be performed is also challenging. Typically, the WHO CCs receive isolates from < 10% of reported cases (Table 2).[75] Due to the challenges in performing culture, many laboratories are introducing real-time, or quantitative, PCR (qPCR), bypassing the need for culture, which at present is a prerequisite for routine WGS. WGS requires viable isolates to be obtained which can be problematic, particularly if the patient has received antimicrobial therapy.

MenAfriNet,[77] an international consortium that supports the strategic implementation of case-based meningitis surveillance in Burkina Faso[78,79] Mali, Niger[80], Togo, and Chad, also plays an essential role in strengthening global meningitis surveillance. By 2017, MenAfriNet had enroled 33 million people in case-based surveillance[81], and 92% of suspected meningitis cases had a CSF specimen collected, of which 26% were laboratory confirmed as *N. meningitidis* (56%); *S. pneumoniae* (40%); or *H. influenzae* (4%).[82] This does, however, demonstrate that despite most cases having CSF collection, three quarters of cases still had an unidentified aetiology.

Initial priorities for capacity building include improving laboratory capacity for and standardisation of culture and isolation of pathogens, as currently few laboratories in sub-Saharan Africa can perform culture-based diagnostic assays. Only after this, should efforts be focused on enabling DNA extraction and sequencing. In addition to resource requirements, increased laboratory capacity relies on technical support from WHO CCs and other international partners, who can provide training, quality control and molecular characterisation when needed. However, currently WHO CCs lack permanent funding and as a result several in-country laboratories have large and potentially valuable collections of isolates which have not been sequenced. At a minimum, regional sequencing capacity would be beneficial since sequencing is fundamental to representative genomic surveillance.

Challenges to achieving representative data collections

Ensuring that WGS repositories are up-to-date and representative is a priority, but likely to be challenging to achieve despite global partners' support. During outbreaks, rapid sharing of genomic sequence data is crucial for tracking disease transmission and responding to developing health emergencies. In particular, public health officials and clinicians need to know whether cases are likely to be covered by available vaccines as they occur. To achieve this, a much higher proportion of isolates would need to be sequenced to WGS level than is currently possible.

Current genome repositories include sequences from collections undertaken for particular public health or research initiatives, unsystematic collections from laboratories or hospitals, as well as epidemiologically representative collections. All collections are valuable for surveillance of important disease-causing organisms; however, systematic and comprehensive collection and sequencing from as many countries as possible is the aspiration.

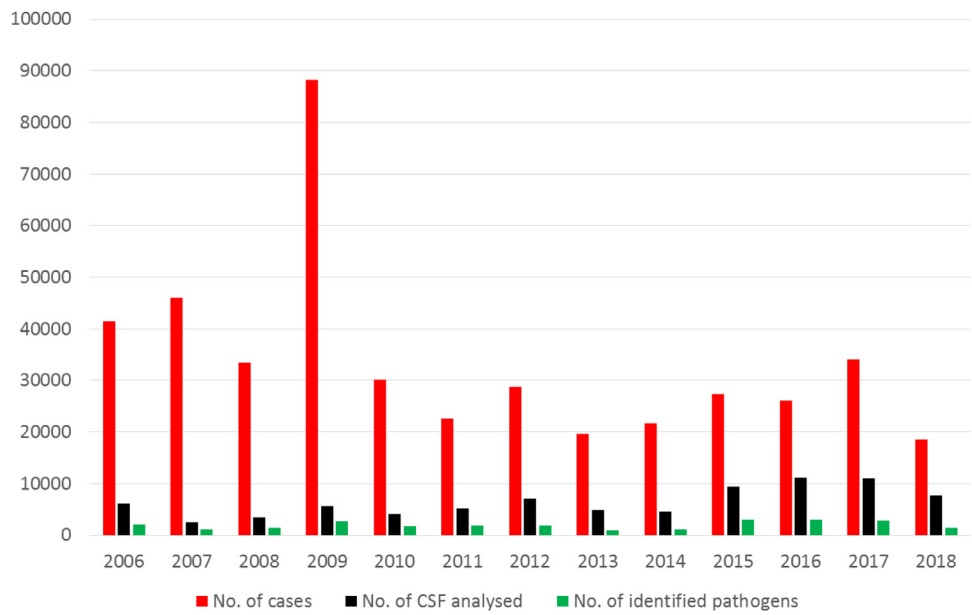

Fig. 4. Meningitis surveillance, 2006–2018: data on suspected meningitis cases from 18 countries south of the Sahara [Source: WHO weekly bulletin][75].

Table 2
Meningococcal isolates sent to WHO-CC's from the laboratory-confirmed cases reported to WHO [Source: WHO weekly bulletin][75].

Countries	2011 Cases	Isolates	2012 Cases	Isolates	2013 Cases	Isolates	2014 Cases	Isolates	2015 Cases	Isolates	2016 Cases	Isolates
Mali	29	6	94	30	6	0	12	0	23	16	44	0
Burkina Faso	257	41	843	167	180	20	210	4	258	11	176	5
Niger	373	17	22	0	11	0	24	0	1390	102	352	0
Nigeria	4	0	4	0	10	7	38	5	20	9	22	14
Chad	104	47	47	23	3	24	0	1	1	1	1	0
Cameroon	92	0	19	4	2	0	0	0	6	0	2	0
Benin	0	0	6	41	5	0	4	0	4	0	13	0
Togo	2	0	9	0	4	0	1	16	36	12	307	42
Ivory Coast	0	0	89	7	0	0	0	0	2	1	3	0
Guinea	0	4	0	0	15	9	13	0	74	0	13	0
CAR	0	0	4	0	0	0	0	0	0	7	56	23
Total	**861**	**115**	**1137**	**272**	**236**	**60**	**302**	**26**	**1814**	**159**	**989**	**84**
% isolates	**13%**		**24%**		**25%**		**9%**		**9%**		**8%**	

Choice of clinical specimen and associated challenges

Resource-poor settings face multiple challenges in contributing isolates to sequencing repositories. One such challenge is the difficulty in obtaining viable culture from CSF, partly due to restrictions or reluctance in performing lumbar punctures.

Blood samples offer an alternative solution to increase the proportion of cases for which specimens are available; however, bacterial load can limit successful culture recovery due to: (i) early infection when the organism may not be sufficiently abundant in blood; and (ii) prior antibiotic use which clears the organism more rapidly from the bloodstream than from the CSF. In children, blood PCR may be positive for pneumococcus as a result of heavy asymptomatic nasopharyngeal colonisation alone.[83,84] Additionally, few laboratories in low-income countries are currently able to perform blood culture. Due to such limitations, blood sampling should be considered as an additional means of confirmation, rather than a replacement for CSF testing.

Another solution would be to sequence bacteria from CSF samples directly. Metagenomic approaches were shown to be effective in a recent outbreak in Liberia, where no isolates were available.[11,85] While contamination with non-meningitis causing organisms or human DNA could reduce sensitivity and raise patient privacy issues, targeted sequencing approaches or removal of human sequence data after sequencing would mitigate this risk.

Vision for a global meningitis genome partnership

Public sharing of WGS data

A global representation of sequence-based typing including WGS is crucial; however, unique isolate identifiers should be defined to allow isolates to be tracked internationally within genome repositories. This is essential for the effective control of epidemics, vaccine evaluation and development. A global overview also requires genome sequence data to be open access, assembled in a readily interpretable and analysable manner, and associated with appropriate metadata.[1] Data holders are also expected to upload short read data to the ENA so that data are open to scrutiny. While the data privacy issue is complex, the requirement for metadata has to balance privacy concerns, with the objective of enabling the interpretation of genomic data for public health benefit. An agree-

[1] Metadata: provide an in-depth controlled description of the sample that your sequence was taken from. Essentially the 'what, where, how and when' of your study from collection to sequence generation, plus contextual data such as environmental conditions or clinical observations. (*Definition from the European Bioinformatics Institute, Available at:* https://www.ebi.ac.uk/training/online/course/ebi-metagenomics-portal-submitting-metagenomics-da/what-are-metadata-and-why-are-they-so-im-0*)*

ment on the minimum core set of associated metadata that should be deposited with WGS data is needed.

Clear codes of practice (COP) should be established for users of repositories within the partnership and for data publication. The consensus COP guidelines at their basic level, serve to secure public access by defining the permitted uses of repositories, thus reassuring submitters who may have legitimate concerns about the use of their data.

Curation and stratification to ensure data quality

The active curation of genome libraries is essential to ensure data quality and enable users to interpret sequence data meaningfully. While automation can reduce the burden of curation, manual curation will be required for a small percentage of data for the foreseeable future. Stratification, through clear labelling of genome libraries is essential to ensure that users understand what the libraries contain, how they are arranged and the origins of the isolates they are studying, *i.e.* carriage or disease isolates, systematically collected or not.

Overlap among different genomic platforms and databases: the importance of data linkage

Existing genomic databases overlap in content and have different but partly-overlapping functions. The interpretation of these databases does, however, rely on the ongoing generation and maintenance of a unified nomenclature. For example, a fundamental function of PubMLST is the ability to allow comparisons to be made among isolates within a species and from multiple datasets.

Data linkage among existing databases, facilitated by APIs, is essential to achieve data harmonisation, avoid fragmentation of information, and enable interpretation. However, while APIs have many advantages, a caveat to their use is that sporadic download of data may mean that users are unaware of the data source, and unknowingly conduct analyses on un-representative samples, risking misleading results and unwarranted assumptions.

Aspiration for a single interface

For a GMGP to attract submissions on a global scale, there is an aspiration for a single interface that successfully draws from publicly-available genomic and provenance data; however, portals to facilitate this are needed. Combined with visualisations, such a system would also make the valuable information gained from WGS data more widely accessible, *e.g.* to public health officials, ministries of health, global health experts, science/health journalists and the broader public. Multiple levels of complexity are, however, required for different users, such that experts can continue to undertake advanced data analysis, while non-expert groups can engage with relevant public health information.

Conclusions and next steps in the development of a global meningitis genome partnership

While there has been a rise in the use of WGS, this has mostly been driven by a small group of countries with adequate capacity and resources to participate and benefit from performing WGS. As a result, current WGS collections are not representative of the global meningitis picture. The advent of the Global Roadmap to Defeat Meningitis provides an excellent opportunity to create a new vision for the role that sequence-based approaches, including WGS, can play in helping to defeat meningitis by 2030, particularly through improving the global surveillance of meningitis. A steering group is being established to coordinate the initiative and encourage high-quality data curation. Next steps in the development of a partnership will include: (i) developing guidelines on open-access sharing of genomic data, (ii) defining a core set of metadata to facilitate open data sharing while protecting confidentiality (iii) engaging with countries not currently involved in existing repositories; (iv) improving countries' capacity to culture and isolate pathogens; (v) consideration of the need for additional WHO CCs in different regions; (vi) improvements in visualisations to provide data in a way people can understand while engaging public health bodies, governments, epidemiologists, scientists from other fields, journalists, and the public; and (vii) establishment of a pooled fund to enable capacity building and country-driven engagement with the partnership.

Declaration of Competing Interest

AM is an employee of the GSK group of companies.

AvdE has received grants from Pfizer, consultancy fees paid directly to the institution from GSK and participated in Science Advisory Boards for Pfizer, GSK and Sanofi Pasteur.

ER, LG & VS represent Meningitis Research Foundation, which receives grants from Sanofi Pasteur, GSK and Pfizer.

JF is an employee of Pfizer Inc and may hold stock/stock options.

JL & RB perform contract research on behalf of Public Health England for GSK, Pfizer and Sanofi Pasteur.

JV acts as temporal advisor and receives grants for research from Sanofi-Pasteur, Novartis Vaccines, GlaxoSmithKline and Pfizer, payed to his institution.

LHH has served as a consultant to GSK, Merck, Pfizer, and Sanofi Pasteur.

LS is currently employed by the GSK group of companies and may hold GSK shares as part of her employee remuneration.

PO is an employee of Sanofi Pasteur.

SDB, HBB, SB, ABB, DAC, LF, OBH, RSH, MJvR, KAJ, BKA, SL, MLF, ML, NM, JM, MCJM, LMS, CMCR, OR, XW, SY and JMS have no conflicts of interest.

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