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TECHNICAL BACKGROUND PAPER

Applications of Whole Genome Sequencing in food safety management

Food and Agriculture Organization of the United Nations
in collaboration with the World Health Organization

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FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS
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Acronyms

AMR	Antimicrobial Resistance
CDC	Centers for Disease Control & Prevention (USA)
CGIAR	(originally) Consultative Group on International Agricultural Research
DDBJ	DNA Data Bank of Japan
DNA	Deoxyribonucleic Acid
EBI	European Bioinformatics Institute
EFSA	European Food Safety Authority
EMBL	European Molecular Biology Laboratory
ENA	European Nucleotide Archive
FAO	Food and Agricultural Organization of the United Nations
FDA	Food & Drug Administration (USA)
GMI	Global Microbial Identifier
ICOPHAI	International Congress on Pathogens at the Human Animal Interface
ILRI	International Livestock Research Institute
INSDC	International Nucleotide Sequence Database Collaboration
MoU	Memorandum of Understanding
NCBI	National Center for Biotechnology Information (USA)
NGS	Next Generation Sequencing
NIG	National Institute of Genetics (Japan)
NIH	National Institute of Health (USA)
NLM	National Library of Medicine (USA)
OIE	World Organisation for Animal Health
PCR	Polymerase Chain Reaction
PFGE	Pulsed-Field Gel Electrophoresis
PII	Personal Identifiable Information
PT	Proficiency Testing
RTE	Ready-To-Eat
SOP	Standard Operational Procedure
SRA	Sequence Read Archive
USDA	United States Department of Agriculture
WGS	Whole Genome Sequencing
WHO	World Health Organization

Executive summary

Recent advances in Whole Genome Sequencing (WGS) technology have the potential to play a significant role in the area of food safety. WGS provides rapid identification and characterization of micro-organisms, including antimicrobial resistance (AMR), with a level of precision not previously possible. With the rapidly declining cost of this technology, WGS applications in food safety management, including the opportunities it provides for enhanced integration of information from other sectors such as human and animal health, could contribute to greater consumer protection, trade facilitation, and food/nutrition security. However, the level of understanding of the concepts and potential use of WGS in food safety management vary among countries. This document aims to take the first steps in addressing these gaps, and providing answers to some of the questions which food safety officials, particularly those in developing countries, need to ask and consider if they are to make informed decisions about the potential value of WGS in food safety management in their particular context. Four real-life case studies are presented to highlight key benefits and potential drawbacks of WGS in food safety management. Benefits and drawbacks are explained in detail, including the issues related to global data sharing. The document discusses challenges in employing WGS within the regulatory framework in both developed and developing countries, and highlights considerations for countries with limited capacity and resources. A simple exercise to enable developing countries to assess the feasibility of incorporating WGS into national food control systems through a step-by-step approach is described. While WGS can significantly contribute to improving food safety management, it still relies on the appropriate interpretation of laboratory data in the context of epidemiological evidence; WGS alone will not suffice. Despite the challenges, WGS is poised to become standard methodology in some places for the identification and characterization of foodborne pathogens. Finding appropriate mechanisms for data sharing will be an important element of its application. WGS is not effective if it is used in a single sector alone or multiple sectors in silo. It is important for food safety competent authorities to consider taking a One Health approach, collaborating closely with their relevant national partners in different sectors and involving relevant stakeholders. In this way, effective management of various multidisciplinary and cross-cutting food safety issues can be achieved. In all of this, there is a strong need at the global level to ensure that situations in developing countries are fully taken into account, and that the technology advances in an appropriate direction, in order for WGS to become an effective tool for all.

Keywords

Food safety, microbiology, whole genome sequencing, WGS, antimicrobial resistance, AMR, data sharing, food security, food recall, public health, One Health, FAO, WHO

1 Introduction

1.1 Background

Food safety is a global concern, and consumers have the right to safe and nutritious food (FAO, 1996). Considerable efforts to strengthen food control systems have been made in many countries. However, while progress has been made, the estimated global burden of foodborne diseases: 600 million foodborne illnesses and 420 000 deaths from 31 major food safety hazards in 2010 (WHO, 2015)[1] and the related social and economic costs (loss of income, employment and market access) remain unacceptably high. In addition, food safety is an important element in achieving food security, which is negatively impacted by both the health and economic aspects of unsafe food. Within this context, there are ongoing efforts in the way in which the building blocks of food control systems are developed and implemented, from legislative aspects to surveillance and monitoring programmes, and the tools used by laboratories.

Whole Genome Sequencing (WGS) has recently emerged as a new tool, and offers great potential in the way we investigate, assess and manage microbiological food safety issues and illnesses. It allows the identification and characterization of micro-organisms with a level of precision not previously possible, therefore potentially minimizing much of the uncertainty which impacts our ability to respond and manage microbiological food safety issues effectively and efficiently. This, in addition to its rapidly declining costs, increases the attractiveness of incorporating such a tool in food safety management.

While several industrialized countries have been moving forward with the technology, its application is limited, particularly in developing and transitional countries. Information describing the potential benefits, possible drawbacks, infrastructure requirements and technical challenges is urgently needed if countries are to make appropriate decisions about when and where to use this technology to strengthen national food control systems.

1.2 Purpose of the document

The purpose of this document is to assist countries in gaining greater understanding of using WGS in food safety management. In particular, it aims to provide useful key elements relevant to the applications of WGS, to enable food safety officials in developing countries to make informed decisions about its usefulness in their particular setting.

1.3 Scope of the document and target audience

This document addresses the application of WGS within the national regulatory frameworks for food safety. In this context, the primary target audience of the document is competent authorities or other authorities with responsibilities for food safety at national level. It is particularly relevant to those working in the areas of microbiological food safety. While not the primary audience, this document is also relevant to the food industry, and research and academic institutions, who are likely to have an important role to play in the possible uptake and implementation of this technology.

[1] It should be noted that these estimates have been characterized by the authors as underestimates, especially as only three chemical hazards have been included.

2 Food safety and WGS

Whole Genome Sequencing (WGS) is poised to provide considerable improvements in the management of food safety worldwide. It represents a significant new tool in the area of food safety, including foodborne disease surveillance, food inspection (testing) and monitoring, outbreak detection and investigation (including food attribution studies), and food technology developments. In comparison to the plethora of molecular identification and characterization technologies available to date, WGS is, conceptually speaking, quite simple, regardless of the platform used. Deoxyribonucleic acid (DNA) is purified, labelled and sequenced, and the results are analysed and visualized using bioinformatics tools. This simplicity provides advantages over the array of current molecular methods that are based on different biological features. WGS is universal for all organisms, and so can embrace human and animal health surveillance, food monitoring and production environment testing. Its simplicity also facilitates and promotes knowledge transfer and training. Additionally, the cost of WGS is rapidly declining. The technology, which until recently was only feasible in large institutions for research purposes, will soon be able to be used for routine food safety management at national and international levels.

The data generated by WGS is, in theory, unambiguous[2]. WGS provides virtually the entire genome, which facilitates targeted exchange and comparison of its data. The results are not only useful for food monitoring, disease surveillance, outbreak investigation and response, but also in addressing broader questions that are critical for food safety improvements and preventive measures, through source tracking, source attribution and the identification of transmission pathways. Since the data comprise the genetic code, WGS results can be used for more than one purpose simultaneously — such as identification, subtyping, virulence marker detection, AMR predictions, and genome-wide association studies. The data can also be mined or reanalysed at any time, which may be useful for the response to, and management of, emerging pathogens over time.

Significant benefits may also arise from WGS application both in disease surveillance and in food monitoring. In human disease surveillance and outbreak response, the vastly increased amount of information provided by WGS makes it possible to precisely refine case definitions, which in turn makes it possible to detect and solve outbreak clusters more quickly, thus preventing additional cases of illness sooner. Because of this high-resolution sequence data, matches between human clinical isolates and food or production environments often provide stronger hypotheses than those provided by matches using older methods. In food monitoring, WGS is used as forensic evidence for source tracking and to inform regulatory action. Since food is a global commodity, global use of this common technology facilitates sharing and collaboration across sectors, and greatly increases the availability of contextual data when interpreting results and recommending regulatory actions with scientific basis.

As mentioned in Section 1.1, the potential for WGS to enable the identification, mitigation and prevention of food safety problems more quickly and accurately than current laboratory methods also translates into reduced economic losses and food waste from incorrect or imprecise implications of wrong products and commodities, both of which are important contributors to food security.

It is, however, important to emphasize that WGS cannot stand alone. It represents one source of information in the complex systems that comprise the whole food supply chain. The technology requires that clinical, food and environmental isolates/samples from routine testing, inspection and

[2] It should be noted that sequencing results can contain errors, including sequence-specific errors. There are ways to reduce the frequency of such errors, and further optimization of the technology is expected to limit this occurrence.

surveillance, and associated data are made available, and that infrastructure is in place to utilize the data for regulatory food safety and public health action. Thus, the implementation of WGS should be accompanied by the establishment of an integrated national food control system and relevant food safety programmes that assimilate information from different sources.

3 Current applications of WGS in food safety management at the national level

3.1 Introduction to the case studies

Whole Genome Sequencing (WGS) has already been used in some countries to subtype common foodborne pathogens in routine regulatory management. In these countries, high-resolution WGS subtyping data has provided more robust outbreak detection, and facilitated subsequent epidemiological investigations. It has also been used in the identification of international outbreaks, based on examination of isolates from clinical samples and from implicated food products in collaborative efforts between countries. In this chapter, four case studies are presented to highlight some benefits of WGS in food safety management. These include: 1) how the high specificity of WGS data allows improved case definition to enhance outbreak management; 2) how the high sensitivity of WGS data allows linkage of seemingly sporadic diseases occurring under the "outbreak surveillance radar"; 3) how subtyping by WGS can aid determining the root cause(s) of complex outbreaks; and 4) how some scientific benefits of WGS are drawing the attention of policy makers in a developing country.

Traditional typing for enteric pathogens requires organism-specific reagents, protocols and methods. WGS, however, is not organism-dependent[3], allowing several pathogens to be sequenced at the same time, enabling simpler, faster and less expensive laboratory operations. The following cases are underpinned by several prerequisites and assumptions:

1. a functioning national food control system with effective food monitoring/testing, surveillance and response;
2. a functioning intra-sectoral and inter-sectoral laboratory infrastructure and networks, including the capacity for sample management collection (storage, transport etc.);[4]
3. an adequate throughput of isolates to sequence, for full cost efficiency;[5]
4. the ability to isolate pathogens from clinical, food, or environmental samples;
5. available technical support in all aspects of the bioinformatics and laboratory pipelines, including computational support;
6. sufficient data storage and connectivity capacity for data transfer; and
7. sufficient informatics capacity and infrastructure.

3.2 Case 1 - Real-time WGS based health surveillance and food monitoring for *Listeria monocytogenes* provided higher resolution than current methods for rapid cluster identification

In 2013, public health and food regulatory partners in the United States began real-time surveillance for *Listeria monocytogenes* in clinical, food and environmental samples. WGS was used in parallel

[3] Organism-specific interpretation of the results of WGS is still essential.
[4] This could start with strengthening and designating a few national reference laboratories to act as WGS hubs/centres.
[5] To further reduce costs in developing countries, WGS could initially be used only in National Reference Labs in the key sectors of Health, Agriculture, Environment and Trade.

with the current subtyping method called Pulsed-Field Gel Electrophoresis (PFGE). In the second half of 2014, WGS and PFGE identified several disease clusters believed to be associated with a specific seasonal food product. PFGE indicated three clusters, two clearly associated with the seasonal food product based on available epidemiological information, and one with case-patient food histories that implicated both the seasonal food product and another Ready-To-Eat (RTE) food product more commonly associated with *L. monocytogenes*.

Whole Genome Sequencing (WGS), however, enabled the investigators to exclude case-patients who had not been exposed to the seasonal food product because the WGS of *L. monocytogenes* from these cases revealed the isolates were unrelated. The WGS data also showed that one of the cases with a different PFGE pattern was part of the disease cluster associated with the seasonal food product. By redefining the clusters with WGS data, public health and food regulatory officials were able to confirm the seasonal product as an implicated food vehicle early in the investigation. The evidence for the identified food vehicle was stronger because it was supported by both WGS and epidemiological information.

The regulatory response to this contamination event was accelerated, and additional listeriosis cases likely prevented, partly due to the agreements in place between clinical, food and environmental agencies to share their preliminary genomic data at a publicly-available central facility[6]. Food, clinical and environmental sample WGS data were uploaded in real-time, and phylogenetic trees were rapidly constructed. The data were also publicly posted and updated almost daily, allowing all agencies to view and exchange critical information, resulting in timely action. These pathogen data portals remain open to any country, industry or academic institution wishing to upload their own WGS isolate data for rapid comparison and clustering; or to download data to use for their own analyses. While this example concerns *Listeria*, a similar approach can be applied to other foodborne pathogens including *Salmonella*, *Escherichia coli* and *Campylobacter*. In the United States, the Department of Agriculture (USDA), United States Food and Drug Administration (FDA) and Centers for Disease Control and Prevention (CDC) routinely investigate multiple foodborne illness clusters on a weekly basis, and these investigations are prioritized and considered for follow-up action based on the strength of available information.

The *Listeria* case, like many others, was followed up with academic research, which identified ways in which similar outbreaks could be prevented in the future. Understanding the true source of an outbreak helps determine preventative controls and mitigation strategies that can be put in place for future protection of the food supply chain.

3.3 Case 2 – WGS identified apparently sporadic cases over a long time period as epidemiologically related, preventing more cases

In Denmark, real-time *L. monocytogenes* surveillance of human infections using WGS was implemented in 2013. Food isolates from samples collected through official food inspections have also been sequenced since 2014. WGS has replaced PFGE for high discriminatory typing. Since WGS is a species-independent method, it was found that real-time surveillance was easier to fit into the routine laboratory procedures by using WGS for subtyping. For example, *L. monocytogenes* isolates can be sequenced at the same time as other organisms, such as *Salmonella*, which was not the case when performing PFGE.

Using WGS techniques, two independent and genetically distinct genetic clusters of *L. monocytogenes* isolates from clinical infections were identified between 2014 and 2015. The genetic

[6] NCBI Pathogen Detection, available at http://www.ncbi.nlm.nih.gov/pathogens.

clusters included ten cases each over a period of 24 months, with patients geographically dispersed across the country. For the first cluster, epidemiological investigations were initiated when a cluster of four cases over a period of eight months was identified by WGS. Although a few more cases with the same WGS subtype were found in the following months, the investigation was closed with no source identified. Almost two years after the first case, one more case was detected. Shortly afterwards, environmental samples taken at Company X were found by WGS to match the cases. Review of case interviews supported a hypothesis that the outbreak source was RTE products from Company X sold in a supermarket chain. The food authorities investigated the production facility, and found several production lines implicated with different RTE products produced by Company X likely to be causing the outbreak. No new cases appeared in the following months. The second cluster developed in a similar manner, implicating a different Company Y producing the same kind of RTE food. Similar to the first cluster, the epidemiological investigations were inconclusive until a matching isolate from a food sample produced by Company Y was found through WGS typing.

These outbreaks show that apparently sporadic cases, probably resulting from low levels of contamination of food products, can be identified and linked by WGS-enhanced epidemiological investigations. They also show that sampling and WGS-typing of food and environmental isolates are often useful for source identification to guide previously inconclusive epidemiological investigations. WGS was also able to precisely distinguish the first cluster caused by RTE products from Company X and the second cluster caused by the same kind of RTE products from Company Y. WGS significantly improves specificity and sensitivity in case definition when diseases occur over long time periods, and increases confidence in hypotheses derived from product-case matches.

3.4 Case 3 – WGS-based investigation identified the root cause of a *Salmonella* outbreak and preventing future outbreaks

Public Health England began real-time sequencing of all presumptive *Salmonella* spp. received from April 2014. In June 2014, a large multi-national outbreak of *Salmonella enterica* ser. *Enteritidis* was linked to the consumption of eggs. Over 350 cases were reported in several European countries. A clear statistical correlation between the UK egg distribution network and distribution of the outbreak related patients was revealed by WGS. This indicated that the outbreak of *Salmonella* related to the source of the eggs.

Whole Genome Sequnecing (WGS) showed that five restaurants in England associated with point-source outbreaks were distinct but linked. Clinical, food and environmental samples in several European countries showed that separate introductions of contamination had occurred from at least two premises owned by a single European egg producer with broad product distribution.

This case shows the power of WGS in revealing the epidemiology behind an outbreak, which allowed the definitive source of the outbreak — a single egg producer — to be identified and targeted for intervention, rather than just the restaurants where the contamination reached the population. Targeted intervention further up the food production chain can be additionally effective in reducing further risks. This case also highlights the importance of the availability of genome sequencing data from multiple countries, demonstrating how global sharing of WGS data could enhance the response to a foodborne outbreak, to further protect public health and identify a particular source of contamination.

3.5 Case 4 - Impact of WGS on food safety and public health policy: Kenyan experience

The Kenya Medical Research Institute at the Centre for Microbiology Research, in collaboration with the Welcome Trust Sanger Institute and the International Livestock Research Institute (ILRI), has been conducting PFGE standard sequencing as well as WGS on *Vibrio* spp., *Salmonella* spp., *Campylobacter* spp. and *E. coli* strains for 15 years. The data emanating from these activities have assisted the government to understand the potential benefit of WGS techniques in mapping disease hotspots, revising existing empiric treatment regimens, and identifying high-risk foods during outbreaks. The government is now more willing to consider investing in food safety and outbreak investigations. Subsequently, several working groups have been established, or are in the process of being established. There is a renewed interest by government ministries to analyse microbial food contamination and antibiotic drug resistance in priority pathogens along the food-chain continuum, with a view to providing regulatory interventions on AMR, surveillance, food testing/monitoring and other priority issues associated with foodborne diseases. A number of factors will contribute to this effort, of which WGS data will be one, but data in itself is not enough. There needs to be a significant amount of advocacy work to ensure that the benefit of WGS comes to the attention of the relevant people — not only policy makers, but also regulatory officials in animal health, human health and food safety sectors, the food industry and consumers.

4 Benefits and drawbacks of using WGS in food safety management

4.1 Key benefits

As the case studies in Chapter 3 highlight, there are clear benefits in adopting WGS technology in food safety management. Such benefits include:

1. **Performance (specificity/sensitivity):** WGS provides more precise information on pathogens than conventional methodologies, by providing virtually the entire genomic sequence. This allows for a much more specific linkage of isolates among human cases and food or environmental isolates, to provide strong hypotheses concerning the source of illness. This enables regulatory authorities to respond to outbreak investigations with a more targeted approach, limiting the number of implicated food products (and therefore those affected by recall) as well as the size of outbreaks (and therefore the number of human cases).
2. **Cost**: WGS analyses are less costly than the myriad of standard subtyping methods necessary to characterize a single pathogen, and which vary according to the pathogen of interest. After conducting just one experimental procedure, WGS provides virtually the entire genetic code of the pathogens investigated. From this genetic code, the lab analyst can identify serotype and virulence factors, and predict AMR of an isolate, without performing additional laboratory analyses. In evaluations performed in selected countries, WGS has been found to be less expensive than existing systems that require numerous sets of tests and techniques (Joensen et al., 2014). A cost and time comparison with current typing techniques was performed in 2015 for selected European public health institutes. This review concluded that "from a cost perspective, on a per isolate basis, WGS can be less expensive than current typing methods for pathogenic *E. coli* and *Campylobacter* spp. For *Listeria*, the cost is more or less the same, and for *Salmonella*, depending on the throughput, the cost can still be somewhat higher. The total time required for WGS is already comparable to that of current typing methods" (ECDC FWD-Next report, 2015).
3. **Speed**: In an optimal set-up, WGS results can be provided within a few days. This is faster than current typing approaches (typically step-wise). Additionally, the detailed analysis enabled by WGS provides more information, fully characterizes the pathogen, and allows for

better source-tracking and root cause determination. WGS can also more quickly provide specific links between isolates, thus picking up putative outbreaks earlier, with fewer cases. The faster and real-time potential compared to traditional methods is a key benefit of WGS in food safety management. Both precision and speed can help to ensure that appropriate action is taken rapidly to protect public health. This can also reduce food waste and damage to industry by avoiding unnecessary non-specific wide-sweeping food recall.

4. **Universality**: An important aspect of WGS methodology is that it is universal across all pathogens[3]. As the traditional methodologies often require laboratories to accredit species-specific identification and typing methodologies, the universality of WGS has a benefit in cost and time efficiency. This benefit could be especially important in developing countries in their process of building effective national food control systems.
5. **Ease of learning and use**: WGS techniques are considered to be relatively easy to learn and apply compared to conventional methods such as serotyping or PFGE. In addition, the universality described above may reduce training requirements, as a large number of pathogens can be analysed in a single laboratory environment.
6. **Ease of sharing**: WGS provides a basic common language that can be easily exchanged electronically around the globe. The relevant data can be stored in repositories and analysed and reanalysed locally at any time. This is an added value over current methods of global data sharing, because it provides the context for local investigations.
7. **Flexible and amenable to reanalysis**: The genetic code of an organism is the most basic information one can get from the organism. When using WGS, there are a number of different sequencing and bioinformatics analytical platforms that can be used at the same time. As new sequencing technologies emerge, or newer analytical methods are developed, historical sequence data can be compared to data generated with new technologies, or can be reanalysed using new bioinformatics platforms. For instance, if new resistance or virulence genes are identified, existing WGS data can be rapidly screened, providing a fast global overview for risk assessment and management (Hasman et al, 2015).
8. **Greater confidence in decision-making:** As WGS has high specificity and sensitivity, it provides greater confidence in regulatory decisions made by competent authorities in food safety, public health and agricultural sectors, as well as those decisions made by food producers and providers.
9. **Easier access to trade and markets**: Using WGS is likely to help competent authorities ensure that they are in compliance with relevant international trade agreements and practices. This will result in trade partners having increased confidence in a nation's food control system.

4.2 Potential drawbacks

As with all new technologies and innovations, there are potential drawbacks as well as benefits in using, or transitioning to the use of WGS in food safety management. Such drawbacks include:

1. **Cost**: The cost of WGS has been declining for a number of years, and that trend is likely to continue. However, the real cost of equipment and consumables for sequencing may still be prohibitive for some developing countries, whose laboratories do not currently perform typing. Additionally, those without established surveillance systems to supply isolates for sequencing may not see the cost benefit of adding WGS capability. Particularly in developing countries, implementation of WGS may divert essential resources from more pressing priorities, such as improved food security situations, development of basic monitoring and surveillance systems or improved water quality. Some countries may choose to support current subtyping methods for a period of time whilst also implementing WGS, which will add an additional burden to both human and financial resources.
2. **Perception of cost**: As WGS is relatively new, many people, especially those who recently introduced to it may consider that as the technology is extremely novel, it must be

expensive. This perception can be a real barrier in considering adoption of the technology into day-to-day food safety management.

3. **Data storage**: WGS generates large amounts of data (in the range of tera- and peta-bytes). It requires both physical space and virtual space thus can also be costly to store in local data repositories. One possible solution could be to submit such data to the global data repositories, which can then make the sequence data publicly available. However, this requires a well-controlled global data sharing mechanism. Possible benefits and drawbacks of such a global data sharing mechanism is further discussed in Section 4.3.
4. **Infrastructure – internet connection/speed**: The large amounts of data generated by WGS need to be transferred through the internet to be available and of benefit to the global community. With limited internet connectivity and bandwidth, and frequent interruptions of power, this can be a major issue for many developing countries.
5. **Data handling**: Most laboratories, especially those in developing countries, do not have access to well-trained bioinformaticians locally, and thus cannot fully take advantage of WGS with their own data analyses. One solution may be access to knowledge networks and software/online platforms (perhaps through the global data repository community) or through partnership with experienced groups that could instruct relevant professionals in-country and help with initial genome studies. Once again, these solutions assume that data are publicly available and that sufficient connectivity exists to access such platforms.
6. **Interpretation of WGS data**: Even with access to basic bioinformatics/genomics software/online platforms, the interpretation of the data, especially in combination with epidemiological information, may not be easy. Training the microbiologists performing the sequencing, as well as the end users of the data, is a critical part of implementing this technology.
7. **Sustainability**: If local and socioeconomic benefits are not well demonstrated and communicated, WGS may not be sustainable.
8. **Possible imbalanced trade opportunity**: If some countries implement WGS for food safety management, these countries are likely to apply the same system for imported food products. However, it is possible that some developing countries with limited resources and capacity may not be able to provide the same level of WGS-based data on food products they export. Thus, they may not be able to enter into mutual collaboration on an equal basis with trade partners who have more resources. This will occur especially when developing countries are not well informed of the benefits/implications of using WGS and how it is used in other countries. A commitment at the global level is needed for countries with established WGS capability to assist countries with limited resources and capacity to benefit from the technology.
9. **Trust**: Not only is there an issue around the legal ownership of publicly available WGS data and applicable privacy laws, there are concerns on the part of data producers, generators and collectors about the ultimate use of their data, possibly due to lack of trust. This can lead to those who hold data to be over-protective of it, and means data sharing may be problematic — even within the same country. Additionally, lack of trust between data providers and data users can lead to data users being sceptical of the data itself. This happens especially when the data imply undesirable results for data users. This issue around trust is common within any other data sharing situation.
10. **Need for basic epidemiology, surveillance and food monitoring/testing infrastructure**: If there are no isolates to analyse, or no authority exists to act on the data, then implementation of WGS technology has limited usefulness and is not a cost-effective investment. In such countries or regions where this is the case, establishing food control systems that include routine collection and analysis of clinical, food, and environmental samples is a pre-requisite to implementing WGS. If basic routine and regulatory capabilities exist, then there may be good justification for focusing on the implementation of WGS, and

to consider housing it in a central public health facility, food testing laboratories or research units.

4.3 Benefits and challenges of global data sharing

Within the international community, there is increasing recognition of the importance of sharing WGS data in food, public health, agriculture and other relevant sectors. As WGS data are universal, there is no point in sequencing exactly the same genomes multiple times and storing the relevant large quantity of data in multiple places. Storing data in global repositories and making such data publicly available help prevent duplication of efforts when sequencing strain collections. Global data sharing contributes to better use of financial resources, scientific progress for higher-quality analysis and building greater accountability with more reliable outputs.

Foodborne pathogens are living organisms, so it is important to note that they keep evolving. With the globalization of trade in various food commodities, as well as the millions of international travellers, these pathogens can spread around the world more quickly than ever before. Trend analysis of pathogen evolution, and movement for rapid response to international food safety incidents, are only possible with the analysis of globally shared data stored in a harmonized way.

It is also important to note that developing countries could gain the most from globally shared WGS data. If a food safety problem linked to internationally traded food products is identified at the global level, real-time information can be used locally to identify similar problems, and the competent authority, together with the food industry, can quickly conduct appropriate interventions to minimize negative health, social and economic impacts. Without globally shared WGS data, it is impossible for a single country to undertake this process quickly, especially if that country has limited financial and human resources.

However, there may still be hesitation about sharing local data globally. This is because ethical and social challenges exist, in particular, within the context of inequities and different levels of capacity. Concerns about data sharing mainly centre around acknowledgement of data owner, producer, generator or collector; and the process of data sharing, including transparency, confidentiality and accountability. Additionally, challenges remain in the validation of reference materials and reference datasets for a global database. There is a strong need for global efforts in harmonization, validation of protocols and quality assurance of WGS methods.[7]

Another challenge is the concern over the concept of "scientific parasites" - scientists may choose to hold onto their WGS data to avoid other scientists using their data for publication. A functioning and effective global WGS data sharing mechanism will only be possible if there is a system that gives all data submitters the assurance that sharing their data will not work against them. Such a system would also require an understanding of what data will be publicly available, and how the submitter can control how much metadata[8] is shared with the WGS data.

Real-time global data sharing may also concern some countries with regards to unforeseen consequences in international outbreaks of foodborne pathogens. For example, providing data showing that a given pathogen is present in food products in one country may result in technical

[7] Many countries are already proposing projects for this. In the United Kingdom, for example, controlled samples with known genomes and metagenomes are being developed and made available to anyone who requests them.

[8] Metadata refers to data providing information about one or more aspects of the data. In the case of genome sequencing, it could refer to where the organism was isolated, when it was isolated etc.

barriers to trade from other countries. Another fear may be that Country A may identify a pathogen linked to specific food sources in Country B and act publicly on this, without engaging with Country B.

In addition, protection of personal identifiable information (PII) is also an issue to consider when discussing global data sharing. While the intention of PII laws is valid, some use this as an excuse not to share raw WGS data. PII laws and the way they are interpreted can vary among countries and, in some cases, create a barrier to sharing key data/information globally, potentially reducing the value of the data itself.

In the long term, it is anticipated that increasing recognition of the benefits of global data sharing will allay such concerns and fears, although economic and capacity requirements may still challenge its realization.

Identifying and developing a mechanism whereby countries have the potential to share data, possibly through anonymous hubs, may be a solution. This could apply not only to national competent authorities but also to individual food companies and academia. National and international guidance and regulation will be invaluable in promoting access to, and sharing of, data in global databases. Trust-building activities, awareness raising and dialogue exchange are essential to promote global data sharing.

There are several internationally recognized, voluntary databases that host relevant WGS data. These include, but are not limited to:

1. **National Center for Biotechnology Information (NCBI)**: NCBI is part of the United States National Library of Medicine (NLM), a branch of the National Institute of Health (NIH). It advances science and health by providing access to biomedical and genomic information.
2. **European Nucleotide Archive (ENA)**: Europe's primary nucleotide-sequence repository, the ENA consists of three main databases: the Sequence Read Archive (SRA), the Trace Archive and European Molecular Biology Laboratory (EMBL) Bank. It provides a comprehensive record of the world's nucleotide sequencing information, covering raw sequencing data, sequence assembly information and functional annotation.
3. **DNA Data Bank of Japan (DDBJ)**: DDBJ is a biological database that collects DNA sequences, developed by Japan’s National Institute of Genetics (NIG). It is also a member of the International Nucleotide Sequence Database Collaboration (INSDC). It exchanges its data with EMBL at the European Bioinformatics Institute (EBI) and with GenBank at the NCBI.

5 Challenges of employing WGS within a regulatory framework

5.1 Challenges of employing WGS in the regulatory framework of developed countries

It is evident that considerable progress has been made in WGS methodology and that it will eventually replace most existing strain characterization and subtyping tools. However, it remains challenging for some to use WGS-generated data for decision making in a regulatory framework. Therefore, various factors need to be considered when making informed decisions with respect to applying WGS in food safety management. These factors include, but are not limited to:

1. development of harmonized guidelines on good practices for WGS data collection, sequencing quality and validated analysis;
2. validation of methodologies used for data mining and analysis before critical decisions are made based on WGS data (WGS data on their own are not sufficient to determine a course of action unless supported by other information, such as epidemiological data and product

distribution data (food chain information), as well as the support of other current methodologies); and
3. ensuring access to global WGS data (this is an important element in the decision making process, but appropriate mechanisms need to be in place to protect the data and establish a trusting relationship).

In order to incorporate WGS within a regulatory framework, several key peripheral issues need to be addressed:

1. **Legal issues**: There may be issues around liability and accountability that are legally binding in respect of WGS data use in a food safety regulatory framework. Legal aspects in relation to methodologies used, as well as the need for harmonized and accredited typing methods, may also arise. However, augmenting good practices in food safety management with the use of WGS data is likely to provide strong justification for regulatory actions. The more precise WGS-based evidence will boost confidence in management actions, thus it is also possible that the implementation of WGS within a regulatory framework will have a positive effect on legal issues.
2. **Trade issues**: If there are no harmonized techniques and interpretation standards, implementation of WGS in a regulatory framework may increase the risk of the methods being abused to create trade issues. It may eventually be useful to establish Memoranda of Understanding (MoUs) across different international jurisdictions. It is also important to incorporate best practices, interpretation and weight of evidence guidelines, as well as set minimum data quality standards.
3. **Proficiency testing (PT)**: Transparent validation and certification are common needs for all new methods including WGS.
4. **Training/education**: Regulators need to be trained to increase their skills and capacities in WGS technologies and management of WGS data, in order to use them in the decision making process.
5. **Communication issues**: Communication constitutes an important element in sensitizing political authorities, trade partners and the general public to the value of WGS technology in the regulatory process.
6. **Global guidance**: There is a need for globally harmonized, clear guidance documents for national and regional competent authorities, as well as food industry stakeholders, to properly apply WGS technology to food safety management at the national level.
7. **Sustainability**: While it is likely that WGS will play an increasingly important role in food issues, including outbreaks and trade, there should be plans to ensure that sufficient resources are allocated to food safety programmes to allow good laboratory practice in the context of performance.
8. **Continuous improvement**: Newer ways of enhancing WGS may improve the depth of investigations into pathogen behaviour and its correlation with food safety issues. Food safety programmes should therefore be regularly reviewed to ensure essential improvements can be incorporated into their systems.

5.2 Challenges of employing WGS in the regulatory framework of developing countries

It is important to take into account that many developing countries have not been able to fully update their national food control systems with newly available methodologies, despite the great worldwide advances before the onset of WGS. Therefore, the likely scenario for the implementation of WGS is that some countries may not have the minimum facilities required for the implementation of WGS in day-to-day food safety management. In addition, each country may have unique challenges in implementing this technology. It is important for such developing countries to consider adopting a step-by-step approach for the implementation of WGS, so that these steps can be tailor-made to fit each country's needs and preparedness.

The following are key actions that can help in four phases of considering and applying the use of WGS technology for national food safety management.

1) Before considering WGS
    1. **Sensitize** policy makers on the global trend of applications of WGS in food safety management, along with benefits and potential drawbacks. This can be achieved through delivering short presentations at various high-level meetings.
    2. **Assess** the suitability of existing and required infrastructure for WGS adoption and implementation including laboratory space, electricity availability and reliability, as well as availability of equipment and internet access. The ability to manage national WGS data also needs to be evaluated. Chapter 6 further discusses such assessment processes.

2) When suitability of infrastructure for WGS adoption has not been confirmed
    1. **Strengthen** the national food control system including food monitoring/testing, health surveillance system on priority pathogens and food safety laboratories.

3) When suitability of infrastructure for WGS adoption has been confirmed
    1. **Develop** a guidance document for decision makers whose support is critical in adopting WGS techniques at the national level. Such a document should include a clear rationale as to why WGS is beneficial as a day-to-day technique in food safety management, as well as its cost-benefit issues. The document will be of greater use if the minimum requirements for each step are described to adopt this technique at the national level. It will be useful to include examples of successful WGS implementation in developing countries and the roadmap they used, as well as the importance of storing WGS data in a public database. Arranging high-level meetings for decision makers to discuss the document can be the next step forward.
    2. **Train** relevant officials and technicians through on-site or online courses, or through attachment to organizations proficient in the use of WGS in food safety, in order that they gain a greater comprehension of the theoretical and practical aspects of WGS.

4) Transitioning to WGS
    1. **Establish** a mechanism to involve key experts and stakeholders from different backgrounds, such as academia, research agencies, government agencies or ministries, international organizations and international experts in food safety, epidemiology and public health.
    2. **Identify/develop** a mechanism to coordinate WGS activities at the national level. This mechanism should function in:
        a. coordination of food safety monitoring, including food testing and inspection, veterinary healthy, traceability and food/product recall based on isolate traceback;
        b. communication regarding trends in food safety incidents and identification of outbreaks;
        c. archiving, sharing and dissemination of data emanating from WGS, as such data impact on both public health and food safety, as well as on international food trade;
        d. coordination of training on WGS techniques, as it improves self-reliance as far as WGS is concerned, which will in turn ensure sustainability of WGS-based systems;
        e. harmonization of Standard Operational Procedures (SOPs) for WGS techniques, including setting up reference standards and evaluating strategies to lower costs associated with WGS;

   f. sensitization of governmental and non-governmental organizations with a view to improving understanding of the value and benefits of WGS as an effective tool in day-to-day national food safety management; and
   g. enhancement of stakeholder involvement/coordination through existing food safety related committees or commissions.
3. **Ensure** sustainable financial support for the implementation of WGS.

6 Considerations for countries with limited capacity and resources

Any new technology can be perceived as too advanced, or too difficult to implement. Alternatively, it may wrongly be thought of as the perfect solution for all problems. It is important for all countries to look beyond such perceptions and consider the potential value and concrete advantages and disadvantages of implementing WGS at the national level. Collaboration of multi-disciplinary structures of all stakeholders, including the academia/research sector and the private sector, is useful in considering the use of WGS in national food safety management.

In countries with limited capacity and resources, a feasibility assessment conducted jointly with potential national partners in the regulatory framework will be a vital starting point for competent food safety authorities. This can include: 1) priority assessment; 2) prerequisite activities and infrastructure assessment; and 3) preliminary readiness assessment.

1. **Priority assessment**: Before considering the implementation of WGS for food safety management, assess the level of priority for food safety and relevant issues. The following are examples of questions to be considered:
   a. Does the topic of food safety rank very high in the national health and food production/security priorities? Are many people in the country affected by contaminated food or water? Do food safety problems significantly affect the country's industry, trade and economy? It may be useful for technical officers to prepare relevant data and evidence to discuss such priorities in sensitizing policy makers.
   b. If food safety represents a high priority, is WGS likely to be useful for priority pathogens in the country? For example, if cholera is the highest local priority, simple detection may be a higher priority than high-level strain characterization. If this is the case, it may be effective to consider a step-by-step approach, implementing a conventional detecting system while an effective health surveillance system and food/water monitoring system are strengthened, then after all priorities are addressed, WGS can be considered for the next step.

2. **Prerequisite activities and infrastructure assessment**: If the priority of food safety is confirmed as high and WGS seems to be one of the options for food safety management, assess various requirements for prerequisite activities and infrastructure. Asking the following questions may be useful in determining the exact steps to follow in the step-by-step approach:
   a. Is there a national food safety policy and an integrated national food control system?
   b. Is there a functional national health surveillance system for priority foodborne pathogens? Is a national clinical reporting system in place? Is there a functional database for foodborne diseases?
   c. Is there a functional national food monitoring/inspection system? Is there the possibility of developing an epidemiological data repository/database at the national level?

   d. Is there good laboratory practice? Is there suitable laboratory space, including a consistent source of electrical power?
   e. Is there a sustainable supply of equipment? Is there a mechanism for maintaining that equipment?
   f. Is there a reliable internet connection and sufficient bandwidth to support the transfer of data to and from international databases?
   g. Is there a pool of qualified individuals who are trained or can be trained to perform WGS?
   h. Is there a pool of individuals who can be trained to interpret and react to data provided by WGS, combined with epidemiological information?
   i. Can national and local authorities be convinced of the value and utility of WGS?

3. **Preliminary readiness assessment**: Having identified the steps and activities needed before implementing WGS, assess the readiness in implementing WGS at the national level by asking the following questions:
   a. Can the appropriate mechanism/setting for WGS activities be defined? Where is the most appropriate placement for WGS at the national level, such as research institutions, universities, hospitals, governmental/regulatory agencies? Where does the required capacity exist? How will information be disseminated to all parties involved, both nationally and internationally?
   b. If the country does not have appropriate capacity or infrastructure, can WGS activities be accomplished through collaboration with other countries, databases, entities, international organizations, or international networks and consortiums? For example, in addition to the United Nations technical agencies such as FAO and WHO, several international/regional networks, consortiums and entities, including regional political/economical bodies in developing countries, can be useful for developing countries to consider partnering with. They include, but are not limited to:
      - PulseNet International: a network of national and regional laboratory networks dedicated to tracking enteric foodborne infections worldwide;
      - International Congress on Pathogens at the Human Animal Interface (ICOPHAI): a congress designed to bring together world experts in One Health from Brazil, the United States, Ethiopia, Kenya, Thailand, and many other countries in South America, Asia, Africa, and Europe;
      - Global Microbial Identifier (GMI): a non-profit technical consortium on WGS for all micro-organisms, including infectious diseases and food safety matters; and
      - CGIAR (originally established as the Consultative Group on International Agricultural Research but no longer stands for this): a global partnership mechanism addressing agricultural research for development.

After conducting the feasibility assessment, developing countries may benefit from communicating its results with international organizations in order to address gaps and to request assistance in capacity development. A tailor-made capacity development programme may be useful in eventually adopting WGS for food safety through a step-by-step approach, and in fully benefiting from the technology, as well as from the relevant globally shared data.

7 Discussion

7.1 How WGS crosscuts and is relevant to a One Health approach

Prior to the emergence of the practical applications of WGS to food safety, scientists used different DNA profiling techniques, such as those based on polymerase chain reaction (PCR), PFGE and other methodologies to genotype micro-organisms for food monitoring and disease surveillance programmes. In many cases these techniques have major challenges, such as difficulties in portability, and difficulties in standardization between pathogens, countries and even between laboratories and technicians within the same country. They are often time-consuming, require highly trained personnel and expensive equipment, and are relatively costly. WGS has a high potential to overcome such challenges as the technology is implemented and evolves.

However, while WGS supports epidemiological investigations with a high-level of precision, it is not effective if used in a single sector alone or multiple sectors in silo. It is important that food safety authorities collaborate closely with their relevant national partners in agriculture, health, environment, trade, economy, legal, research and other sectors. Involvement of relevant stakeholders is also essential to manage multidisciplinary and cross-cutting food safety issues. Development of a systematic way to involve all stakeholders including 1) private food companies and associations, 2) the IT industry, 3) scientific and analytical companies and industry, 4) academia and research institutes, 5) civil society and consumer groups and 6) farmers and cooperatives is extremely useful in considering the adoption of WGS for food safety management.

FAO, WHO and the World Organisation for Animal Health (OIE) support and promote the approach to address relevant cross-cutting issues in a One Health framework. WGS is a potentially powerful tool in all relevant sectors in food, public health and animal/plant health, thus the topic has a strong relevance to a One Health approach.

7.2 WGS for food industry

While this document mainly focuses on the use of WGS in food safety management by regulatory and public health agencies, it is important to keep in mind that one of the key players in the national food control system is the food industry. Depending on the country situation, adoption of WGS by the food industry may either precede or follow its adoption by regulatory agencies, and may incur additional challenges and concerns including legal/liability issues. It is important that governments consider providing an enabling environment for the food industry to generate, use and share relevant WGS data. In this way, public-private collaboration would promote applications of WGS in the most desirable and appropriate way for improved national food safety management.

7.3 Key needs for global actions and potential roles of international organizations

Several key needs for global actions in the area of WGS for food safety management include but are not limited to:

1. provision of forums to discuss relevant issues, including data governance issues and legal matters around harmonization and standardization at the global level;
2. coordination of relevant activities at the global level to ensure a One Health approach;
3. provision of assistance to developing countries in assessing capacity development needs in strengthening national food control systems (laboratory, epidemiology, food

monitoring/testing, health surveillance, policy and regulation) and infrastructure, defining food safety priorities and the utility of WGS for priority pathogens;
4. facilitation in linking developing countries with relevant networks, consortiums or entities where partnerships can be beneficial for such countries;
5. global facilitation and engagement in international outbreak investigations;
6. provision of harmonized guidance in overcoming technical barriers to trade that may result from WGS-triggered investigations;
7. provision of technical guidance with good practices in application of WGS in food safety, as well as in global data sharing; and
8. Facilitation of public-private collaborations.

8 Concluding remarks

Countries with an existing effective food control system, combined with functioning foodborne disease surveillance and food monitoring/testing systems, are well positioned for adopting WGS technology to augment microbiological food safety management within a risk analysis framework, and are likely to benefit greatly from the technology by sharing data internationally and publicly. Other countries may first need to develop or strengthen basic national food control systems with competent authorities, laboratories, food monitoring/inspection systems and health/epidemiological surveillance capacity in order to fully benefit from future technology upgrades, possibly with WGS. Opportunities to leverage existing WGS and relevant initiatives in other health and agriculture areas to strengthen food safety infrastructure may be useful in promoting the use of WGS in the regulatory framework.

Whole Genome Sequencing (WGS) is poised to become standard methodology in food safety for the identification and characterization of foodborne pathogens, including AMR organisms. Implementation of WGS in support of food safety management is likely to be beneficial for all countries, and countries that do not embrace the technology may be disadvantaged regarding food export and tourism. Nevertheless, there is a need for a global leadership in steering this initiative towards a wider application. Currently there are various voluntary global, regional and national initiatives promoting and taking a coordination role for WGS in food safety. However, as there will be an increasing number of relevant activities, there may be a greater need in the future for international organizations to play a role as neutral and transparent brokers that can fully consider the unique situations of developing and transitional countries. Specifically, proper linking of national and regional WGS data, or facilitation of global data sharing is needed, along with the provision of best practices and guidelines that will facilitate appropriate harmonization and interpretation of WGS data in a manner that promotes public health, enhances food safety, and improves food security. This would also support effective coordination on outbreak detection and investigation at the global level.

While WGS can significantly contribute to improving food safety management, it remains true that laboratory data should be interpreted in the context of epidemiological evidence; WGS alone will not suffice. WGS is another new technology in the laboratory toolbox. It can be a powerful supplement to, but not a substitute of, basic good practices — such as strong public health surveillance systems, multidisciplinary investigations, good agriculture practices, good manufacturing practices and good hygiene practices in the farm to table continuum. These have to be strengthened no matter how widely WGS is adopted in national food control systems.

Sustained commitment and close collaboration at the global level are essential to ensure the implementation of WGS such that, in addition to realizing the food safety, public health and food security benefits, publicly shared data are not used for the purpose of imposing barriers to trade. In fact, integrated and appropriate implementation of WGS should be beneficial to trade by providing developing countries in particular with improved abilities to detect and mitigate potential problems in export commodities, while demonstrating a higher level of control for equivalence purposes, such as traceability. There is also a strong need at the global level to ensure situations in developing countries are well considered, and that the technology advances in an appropriate direction, in order for WGS to become an effective tool for all, and to provide guidance to support all countries in considering the implementation of WGS in food safety management.

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