

Next generation Sequencing (NGS): A new way to answer old questions

David Rodríguez Lázaro, DVM, MSc, PhD
Microbiology Division
Department of Biotechnology and Food Science
Faculty of Science
University of Burgos
Plaza Misael Bañuelos s/n
Burgos, Spain
drlazaro@ubu.es

The promotion of a high level of Food Safety and Quality is a major policy priority worldwide. The globalization of the food market, the increase of personal and food transportation, and the changes in food consumption habits provide s a lot of opportunities and challenges to minimize incidence of Food Safety in the stakeholders. In recent years, several stogies have been developed to foster repid and accurate answer to the needs poed by the food insutry and consumers, being the Next generation Sequencing a very promising alternative to exploit strategies for rapid identification of foodborne pathogens and source attribution, as well as characterisation of isolates and microbial communities to address risk assessment within the microbial risk management.

In this scenario of activities, NGS can play a relevant role in several activities. Firstly, NGS can instigate a more effective implementation of alternative methods for detection, identification and quantification of foodborne pathogens. Because the classical microbiological methods are time consuming, and laborious, even not always reliable for instance the presence of viable but non-culturable VBNC forms as happens with *Campylobacter*. In parallel, NGS can be a pivotal tool on epidemiological studies in the food chain, which is one of the pillars of the risk assessment. For that, the first step is the determination of prevalence and contamination loads of microorganisms of interest in different food commodities. The results will allow to develop strategies for preparedness for emerging agents transmitted via food to anticipate food alerts. In addition, the prevalence and epidemiological data will also allow to design effective and fit-for-purpose sampling methods and schemes in different food commodities. Importantly mainly for unknown and unexpected agents is inexcusable the use of to identify the source attribution for particular microorganisms. In addition, it is also important to use this cut-of-the-edge tool for source localization along the food chain. A third relevant aspects in where NGS is essential is on the characterisation of foodborne pathogens for specific traits using molecular approaches. For example, once we have obtained specific potentially pathogenic isolates from different food commodities we could characterise them for the presence of specific traits by genomic approach : (multi-)antibiotic resistance properties, or virulence characterisation using NGS for the presence and gene expression of the major molecular determinants of virulence. In addition, exploiting NGS let us deeply characterise the isolates allowing establishing phylogenetical relationships. These findings will support to quality departments in the food industry to determine if the food and food environment related contaminations

are recurrent (i.e. the strains of a given microorganism obtained during several samplings in the same facility is the same isolate) or are new contaminations (i.e. although strains of a given microorganism are obtained during several samplings in the same facility, they are not the same isolate). This is a critical aspect to define cleaning and hygienic measures within the food industries, as the strategies for disinfection can be totally different. When, a recurrent contamination is found, the strains isolated are usually well adapted to the food production facilities (usually very stressing for microorganisms) and usually have the capacity to produce biofilms were are quite resistant to regular cleaning procedures. Therefore, in that given case, the cleaning and disinfection must be more exhaustive even when the original problem (i.e. contamination with a given microorganisms) is the same. Consequently, the application of molecular tools will help dramatically to establish correct hygienic strategies in the Food Industry.

Similarly, NGS can help us on ecological studies of microbiota in food. Through this activity, using NGS, we can characterise the microbiota present in a given food or in a food environment. This is a novel and interesting approach for instant to characterise food spoilage. Usually the spoilage of a given product is produced by a misbalance on the common microbiota, and the ecological development of spoiling microorganisms. Differently to what happen with foodborne pathogens where the development of a specific microorganism produces a cascade of symptoms that finally can be clinically significant for disease attribution, the spoilage of foods is a combination of different factors in which the modifications of the microbiota can play a main role. Consequently, the definition of the microbiota of given products specially those raw or fermented or those with a slight disinfection treatment, i.e. the fingerprint of the microbial commodity, will allow both to characterise the factors influencing the spoilage and the microbial groups implied. The spoilage of foods is a research field not too studied, particularly if it is compared to foodborne pathogens research but important because of its economical important but as well as it can reduce the food shelf life that it is important for the commercialization chain, basically exportation. In addition, the funding programs both at national and international level, highlights that one of the pivotal research and socioeconomical areas of implementation must be the development of strategies to guarantee food security. Food security is not only related to the improvement of production programs, but particularly to delivery of the goods produced in good state, and consequently, the spoilage of foods therefore plays a major role.

In conclusion and summarizing what has been described above, NGS can help us to answer in an innovative, accurate and effective manner the traditional questions that affects the food safety and quality most significantly.