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Opinion

Next Generation Sequencing in Food Microbiota: Biotechnological and Food Safety Benefits

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Since advances in technology have always driven discoveries and changes in microorganism taxonomy, taxonomic identification is an issue of primary importance when approaching the study of food microbiota. In this scenario, genomics now underlies a renaissance in food microbiology therefore accelerating food safety monitoring and food production processes [1,2]. Microbial taxonomy directly influences a number of basic scientific and applied fields where microorganisms are involved [3] including food production, conservation and probiotic activity.

Progresses in sequencing technologies and bioinformatics analysis of data, led nowadays to a more complex scenario of food microbial communities, offering a panel of analytical tools able to screen the whole microbial community of food matrices. The use of universal markers produces several DNA barcode fragments, corresponding to the each bacterial species present in a food sample. With the ultimate goal of characterizing the complete spectrum of microorganisms, several novel approaches, referred to as 'Next Generation Sequencing' (NGS) and, more recently, 'High Throughput Sequencing' (HTS), have been developed [4-7].

Food microbiology deals with the study of microorganisms that have both beneficial and deleterious effects on the quality and safety of food products. The fast and low-cost NGS approaches have revolutionized microbial taxonomy and classification and have changed the landscape of genome sequencing projects for food-associated microbial species. The NGS-driven advances have been exploited mainly to re-sequence strains and individuals for which reference genome sequences are available in order to sample genomic diversity within microbial species. The NGS approaches have greatly increased the ability of researchers to profile food microbial communities, as well as to elucidate the molecular mechanisms of interesting functionalities in food ecosystems. These applications enable the culture-independent sequencing of collective sets of DNA or RNA molecules obtained from mixed microbial communities to determine their content [6].

NGS techniques have promoted the emergence of new, high-throughput technologies, such as genomics, metagenomics, transcriptomics and metatranscriptomics, etc. As compared to previous culture-independent methods, the number of nucleic acid sequences analyzed by NGS techniques is exceedingly higher, allowing a deeper description of the microbial constituents of the ecosystems. These technologies can be used in two substantially different ways: sequencing the total microbial nucleic acids (shotgun sequencing) and gene-specific sequencing (targeted sequencing). For the latter, segments of highly conserved DNA or cDNA sequences are first amplified by PCR using universal or group-specific primers. Targeted techniques provide a snapshot of the diversity and phylogeny of the different elements making up microbial populations. The term phylobiome has been introduced recently to refer to the phylogenetic information gathered using this approach [8].

In addition, shotgun techniques inform on the genetics and functional capabilities of the microbial constituents of food ecosystems, providing insights into the number and potential function of genes within the community [6,9]. Both shotgun and targeted techniques have already been used to study the microbiology of a series of foods and food fermentations, and pertinent reviews have recently been compiled [4,6,10]. However, research in this area is so active that findings must be continually reviewed, and the current and potential applications of these constantly updated.

Interesting review articles on various aspects of the impact of NGS technologies on food microbial genomics were drafted and provided complete information about the most common NGS systems and platforms and then addressed how NGS techniques have been employed in the study of food microbiota and food fermentations, discussing their limits and perspectives. The most important findings are reviewed, including those made in the study of the microbiota of milk, fermented dairy products, and plant-, meat- and fish derived fermented foods [4-6,11,12].

Recently, various studies used NGS approaches to study the microbial ecosystem (in terms of diversity and dynamics) of different fermented foods [10,13-20] and in most cases, the obtained results could be of great impact on the food supply chain to improve industrial biotransformation processes, enhance quality of final products, extend the shelf-life and valuating local productions.

Investigation of the bacterial communities involved in fermented products can be carried out by microbiological and molecular methods. The latter approaches are now being revolutionized by the introduction of NGS, most specifically Illumina platforms which provide million of reads up to 300 x 2 bp length, with further advancements expected in the near future [15]. We are rapidly moving to the postgenomic era, when a complete assessment of the genes present in a certain sample will be obtained, and their expression and activity assessed by metatrascriptomic and metaproteomics. An important step in this direction is represented by the quantitative assessment of bacterial taxa, which can be achieved by sequencing of 16S rRNA amplicons: the identification on the basis of 16S NGS data of the species present in a food sample can greatly support metagenomic analyses to be conducted on the whole microbial DNA.

This highlights the great potential of the NGS application to microbial ecology of fermented meat products to gain a complete and in-depth picture of the bacterial species and identify species that cannot detected with classical microbiological and molecular methods.

Moreover, RNA-based analysis can significantly increase the ability to identify the impact of the microbial population on organoleptic characteristics of typical food products; when RNA is analyzed, the microbial populations that are metabolically active can be potentially detected and identified, and these are the populations that contribute the most to the fermentation process [21].

Conflict of Interest

No any conflict of interest exists.

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