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DEVELOPMENT OF A RANGE OF SPIKES FOR NGS IN THE CONTEXT OF BIOSAFETY

Tarfa Mustafa[1], Camille Marmai[1], Maïlys Daniau[1], Magali Roche[1], Thibault Guinoiseau[1] et and Catherine Legras-Lachuer[1,2]

[1]Viroscan3D, France
[2]Dynamique Microbienne et Transmission Virale, France

NGS has grown considerably over the past years for various applications, including infectious diseases (virology, microbiology, metagenomics), clinical research and R&D. NGS is also increasingly used for quality control (biosafety) to identify all nucleic acids that can potentially contaminate a product. A disadvantage of NGS however, is that it is not possible to quantify the viral or bacterial genome from the number of reads, without adding already quantified external control sequences. The aim of this work was to develop a range of positive controls (spikes) which will allow the determination of the detection threshold of a contaminant and to quantify a pathogens genome in samples, according to coverage and depth. From *Xenopus Laevis*, we have selected unique sequences between 500 and 2000 bp showing no significant homology to viral, bacterial, fungal or human (and other eukaryote species) genomes (>1.5%). The DNA fragments which were homologous with ribosomal, repeat or overlapping sequences have been also eliminated. A total of 36 spikes distributed in six pools of six spikes were produced (ssDNA, dsDNA and RNA between 500 and 2000 bp). Every bank of spikes was quantified by qPCR and verified by Sanger sequencing. The 6 spikes were pooled by quantity, ranging from 1 to 1,000,000 copies. The detection of these spikes was then validated by NGS in order to identify the depth and the coverage according to spike numbers. This work will enable the development of NGS for biosafety, which could replace or complement classical methods (PCR/*in vitro* tests).

Biography

Tarfa Mustafa has been graduated from Grenoble-Alpes University in 2012 as Molecular Biologist, specialisations include Transcriptomics and Metatranscriptomics approaches. Later he has completed his Post-graduation from National Center of Scientific Research (CNRS), Grenoble with subject on Molecular Approaches To Study The Interaction Between Microbial Communities With Alpine Ecosystems. Since 2016, he works at Viroscan3d, Lyon within the Department of Research and Development

Thibault.guinoiseau@viroscan3d.com