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SMARTDIAGNOS: Sample Concentration Integrated Solid-Phase PCR for Next-Generation of Sepsis Diagnostics

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At the Third International Consensus Definitions in 2016 Sepsis has defined as "life-threatening organ dysfunction caused by a dysregulated host response to infection". Sepsis is one of the ten leading causes of mortality in intensive care units (ICU) worldwide. It is too complicated to determine the exact number of patients suffering sepsis, but numerous studies report that the incidence of sepsis is increasing over time, offsetting declining case-fatality rate. It was estimated that at least 19 million people experience sepsis each year worldwide. In the USA, sepsis causes more than 200 000 deaths each year, and costs exceed \$24 billion per year for the direct health care. In Europe, 27% death was estimated from 35% of patient in ICU suffering sepsis and costs 7-21 billion EUR per year for the health sector. To date, early identification of pathogen causing sepsis is a crucial and unique way to reduce mortality, due to every hour of delay in appropriate antimicrobial therapy increases mortality by 5-10%. Current diagnosis methods for sepsis such as conventional blood culture, MALDI-TOF, Septifast and other nucleic acid-based multiplex amplification techniques are time consume (24- 68 hs) and/or low sensitivity. The H2020 EU funded SMARTDIAGNOS project will generate new diagnostic tools for accurate and earlier pathogen detection and identification. In this study, a number of innovative technologies such as pathogen concentration, direct PCR, solid-phase PCR, supercritical angle fluorescence (SAF) microlens array will be developed and combined into a streamlined point-of-care (POC) and a LAB device to archive ultrasensitive, rapid, and unlimited multiplexing pathogen detection in blood samples. So far, the immune-magnetic beads could efficiently concentrate bacterial pathogen at a low concentration of 10^1 to 10^2 CFU/mL from the human pathogen spiked blood sample within 40 min. Integration of a direct solid-phase PCR using a Phusion hot start DNA polymerase and optical SAF microlens array in a single polymeric Lab-on-Chip platform allows for highly sensitive and multiplexed pathogen detection and their antimicrobial resistance genes within 3 hours (including sample preparation and target concentration) with a detection limit of 10 CFU/mL of the blood sample. Fast, sensitive, and accurate detection of the pathogen causing sepsis and their AMR genes will improve patient outcome, shorten intensive care stay and thus reduce mortality and health care costs.