<u>Kytaro, Inc.</u> Detection System for Multiple Antibiotic Resistant Pathogens

Sepsis is a source of major health concerns. 30 million people suffer from sepsis every year worldwide. Sepsis progression is rapid. Mortality rates increase by 7% each hour that antibiotics are delayed in sepsis shock patients. Therefore, patients who acquire blood stream infections (BSIs) need broad-spectrum antibiotic treatment within approximately 3 hours. However, these antibiotics are not effective against all BSI and may cause adverse reactions in some patients. BSIs are generally diagnosed through blood culture that detects bacteremia and fungemia. These cultures are labor intensive and take between 18 hours and 6 days. Existing solutions relying on PCR either lack sensitivity and/or speed. Microbial cells often appear in the blood in very low concentrations of less than 1 CFU/mL. The development of faster and more sensitive pathogen diagnosis tools is therefore critical for the successful treatment of sepsis. Despite considerable advances, detection of very low number of pathogens is an unresolved challenge. We propose a new inexpensive and simple technology to significantly accelerate pathogen detection by concentrating and detecting pathogens within a few hours (<4h) at above detection limits. This method becomes part of the initial incubation and enables extraction of pathogens from the entire volume of sample. Using this simple technique, the bacterial quantity reaches sufficient levels for detection in an early stage of pre-enrichment. Eliminating the need for diluting or aliquoting like other techniques. We have already preliminary proof-of-concept. In order to demonstrate utility in clinical settings, in this effort, we will demonstrate that we are able to detect and identify multiple antibiotic resistant pathogens with starting with a concentration of 1 CFU in 10 ml of blood in less than 4 hours. We will then demonstrate selectivity without false positives in multiplexing to demonstrate scalability of the solution. This novel and innovative in vitro diagnostic test that can detect antibiotic resistant pathogens in inpatient and/or outpatient settings. The solution is easy to use (simple arrangement and lateral flow strip tests), fast (<4 hour, we are aiming for 3 hours), sensitive (1 CFU in 10 mL), with high specificity, and multiplexing ability to process a broad range of specimen types. This collaborative proposal leverages their strengths in developing technologies for rapid pre-concertation and biosensing to significantly shorten the time taken to detect antimicrobial