PhD thesis: Nanoporous silicon chemical functionalization for sepsis diagnosis.

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Sepsis, blood bacteremic infection, is one of the main mortality causes in the world. It is essential to treat sepsis as quickly as possible since the chances of survival decline by 7% each hour. As current sepsis diagnosis require several days, broad-range antibiotics are massively used in a first instance, driving the global antibiotic resistance threaten. Recently, 6 classes of blood metabolites have been identified as sepsis biomarkers. Metabolites being low molecular weight species present in trace in blood among abundant larger species, their detection and clinical use as biomarkers is a global failure. There is an urgent need for reliable and rapid metabolites separation strategies, from serum, compatible with classical Mass Spectrometry (MS) analysis for sepsis diagnosis.

In 2014, a 15 minutes isolation technique of metabolites from serum compatible with MS was demonstrated at CEA-LETI (Grenoble, France) [1]. A drop of serum is spotted onto nanoporous silicon (pSi) leading to metabolite sterical trapping into the pores, while larger species are washed from the surface. Then, metabolites are directly analyzed by MALDI-MS. Statistical analysis of mass spectra discriminates highly pathological samples from normal samples. Nevertheless discrimination of the intermediate disease stages is impossible and reflects a lack of sensitivity in disease specific metabolites isolation and detection.

Based on these results, this project aims at developing an innovative platform, based on pSi and MS, for highly sensitive sepsis diagnosis within 15 minutes. Two novel strategies have been identified to solve the sensitivity problem: (i) development of an innovative pSi matrix that, in addition to steric exclusion of larger species, enable isolation of sepsis-specific metabolites, among other metabolites, from serum. To achieve this selectivity toward sepsis metabolites, we propose to monitor the pSi trapping capabilities by tuning its 3D surface chemistry. (ii) Optimization of the metabolites detection limit by using Desorption Ionisation On Silicon (DIOS)-MS instead of MALDI-MS. Herein we propose to optimize the DIOS-MS detection limit by evaluating different pSi elaboration techniques (electrochemical process, Glancing Angle Deposition) and by tuning pore size, depth, porosity and morphology.

Framework and Timetable

1- **Identify surface functionalization that offer the highest binding affinity for each class of purified sepsis-specific metabolites through interface characterizations and modelling.** To this aim, silane molecules will be used to functionalize different pSi matrixes. Metabolite trapping capabilities of the resulting modified surfaces will be evaluated by XPS, infrared spectroscopy, AFM in the spectroscopic mode [2]. Molecular dynamics simulations will be developed to model metabolites / silanes interactions and predict and optimize metabolite trapping event.

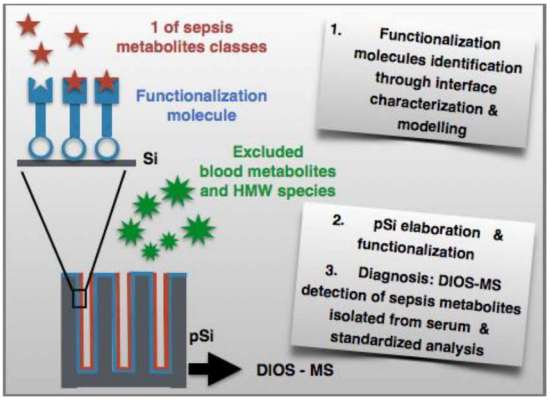
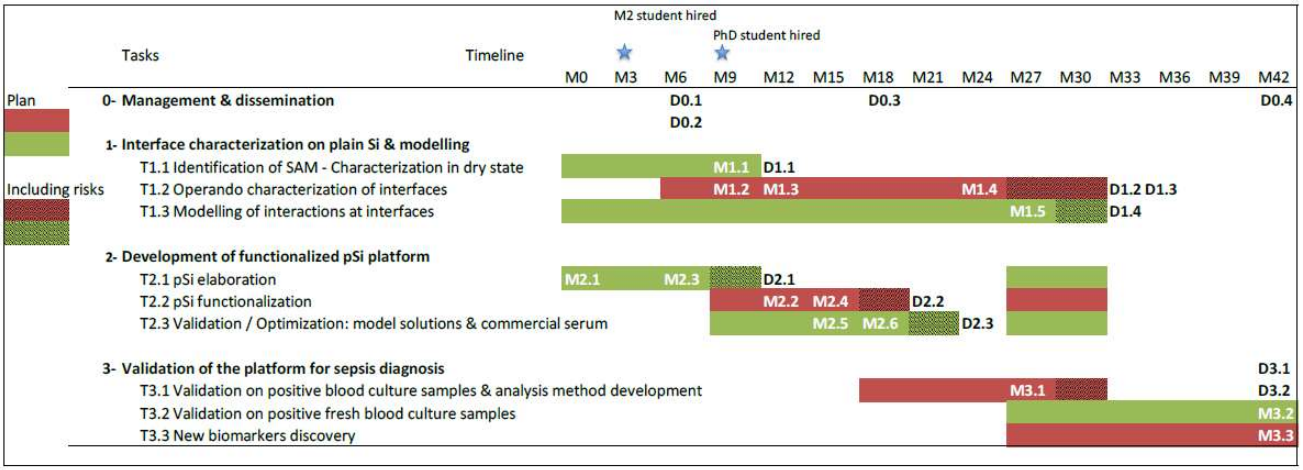


Fig. 1: Framework of the project

2- **Develop 3D functionalized pSi matrixes for sepsis-specific metabolite capture from serum and for optimizing DIOS detection.** To this aim, a process for the 3D functionalization of porous silicon will be developed. DIOS-MS detection limit will be optimized by evaluating different pSi elaboration techniques and by tuning pore structure. The clear advantage of both DIOS-MS analysis and functionalized pSi, compared with unfunctionalized pSi and MALDI-MS, for the detection of sepsis metabolites from model solutions and then from serum, will be demonstrated.

3- **Benchmark 3D functionalized pSi matrixes & DIOS-MS analysis against the gold standard sepsis diagnosis method.** Several cohorts of patients will be evaluated in collaboration with The Grenoble CHU. MS profile analysis is complicated by the inherent variability in each sample. A method combining data pre-treatment and multivariate statistical analysis will be developed and standardized.



Collaborations and expertise of the team:

- **INL** expertise: pSi elaboration, surface functionalization, surface characterization, and simulations,
- **CEA-LETI** (Grenoble) expertise: thin film deposition on silicon, mass spectrometry, metabolomics and metabolite trapping in pSi,
- **Grenoble CHU** expertise (pathological samples)
- **MEDIMPRINT** (Grenoble) expertise for industrial transfer.

References

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