

**Scientific report – phase 1 and 2 (October 2020-December 2021)**

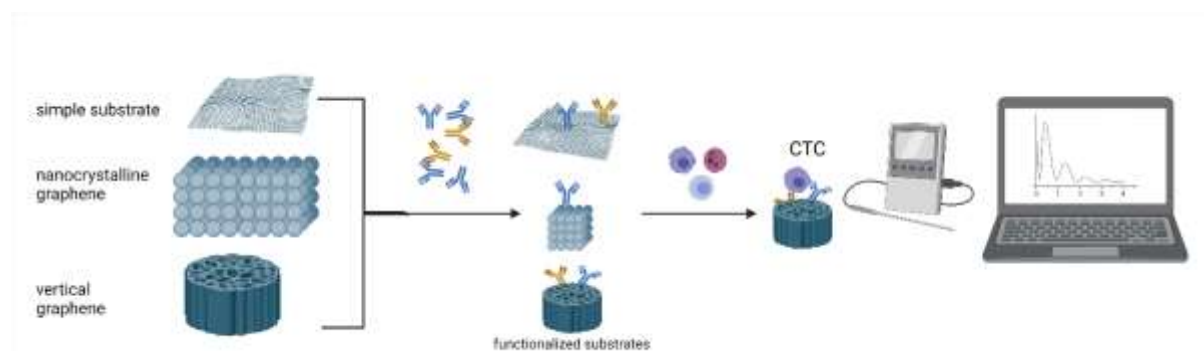
**PED 382/1.10.2020 – CTNANOSCAN**

**Phase 1: Scientific and technical design of functionalized substrates**

**Phase 2: Selection and structural characterization of functionalized nanostructures**

### **Summary**

During the first two stages of the project CO and P1 have collaborated to obtain at least one type of substrate, functionalized with anti EpCAM antibodies and/or CD36 for tumor cell capture. This substrate can be included in a sensor that records and documents the binding of circulating tumor cells (fig.1).



*Figure1. Workflow of the project CTNANOSCAN. Following the documentation of the state-of-the-art, several graphene-based substrates were developed, characterized and functionalized with anti-EpCam and anti-CD36 antibodies. The functionalized substrates were incubated with normal and tumor cells, compared for selectivity and recorded using a mobile sensor, for impedance signals. Image made with BioRender and presented at the 14th International Conference on Pathology, November 4-6, 2021, Bucharest, presentation awarded with 2nd prize in the Young Researchers category.*

Substrates based on nanocrystalline graphite and vertical graphene were selected, as they did not bind with high affinity normal and tumor cells in suspension, and at low cell concentrations (2000 cells/mL), the binding percentage was below 5%. This lack of cell-to-substrate non-specific interaction is a guarantee of the specificity offered by subsequent functionalization. The optimal functionalization depended on the type of substrate: for graphite nanocrystalline cysteamine had optimal results, and for vertical graphene – protein A. To test cell adhesion, a standardized line of normal human monocytes and a standardized line of breast cancer cells were used. To test the impact of the CD36 receptor on the capture of tumor cells and the effectiveness of the functionalized substrate (the aim of the next phase), these lines have been genetically edited to knockout the receptor.

The scientific activities resulted in 1 original article accepted in a BDI journal, 1 ISI review article under evaluation and 3 participations in international conferences, of which 1 ISI-indexed (2 poster presentations and 1 oral presentation, awarded with the 2nd prize).