A protein microarray immunoassay for the assessment of humoral immune responses against human pathogens.

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A microarray consists in the immobilization onto a solid surface (normally a microscope slide) of ordered arrays of tens to thousands of biomolecules of different nature (nucleic acids, proteins, antibodies, carbohydrates, and so on) which retain their specific binding properties: nucleic acids with nucleic acids, proteins with other proteins or antibodies, etc. For this reason, microarray technology enables the analysis of a multitude of biologically relevant parameters in high throughput and with true parallelism. In particular, protein microarrays have been employed for multiple approaches: identification of protein-protein or protein-small molecules interactions, cancer profiling, search for specific antibodies against allergens, autoantigens, microbial and viral pathogens, detection of microorganisms, drugs of abuse and toxins in serum and other biological fluids.

“ToRCH” is an acronym which indicates a group of pathogens responsible of mild or symptomatic infections in the mother, but of very serious diseases in the fetus. A primary infection contracted in the first or last trimester of pregnancy may lead to congenital abnormalities, miscarriage or natural abortion. For these reasons, an early and precise serological evaluation of the antibody titers against all the pathogens potentially dangerous in pregnancy is of primary importance, but often not performed because of high costs and long times required to carry out the tests.

In my Biotechnology Degree Thesis I described the setting up and optimization of a microarray test for the evaluation of IgG and IgM responses against a group of 7 vertically transmissible pathogens in one test: the classical ToRCH [Toxoplasma gondii, Cytomegalovirus (CMV), Rubella virus, Herpes Simplex virus types 1 and 2 (HSV1, HSV2)], Varicella Zoster Virus (VZV) and the bacterium Chlamydia trachomatis. In particular, I dealt with the optimization of the spotting conditions, with the definition of a correct processing protocol, and with the analytical and clinical validation of the test.
During my Ph.D. course, I will keep on dealing with the project that I have already started during my Thesis apprenticeship, by addressing the following issues:

Completion of the setting up of the immunoassay in terms of analytical parameters (precision, accuracy, stability) and clinical parameters (positive and negative predictive values, efficiency).

Once completed the validation, the test will be used for the serological screening of a wide group of pregnant women.

Employment of this same test for the screening of patients at risk of ToRCH-related diseases because of immunodeficiency due to natural causes (HIV infection, hematological malignances) or artificially induced (transplant recipients).

Design of arrays including different antigens related to other groups of pathogens, for which serology may be an important diagnostic tool.