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Activity

Reverse Phase protein Arrays represent an innovative technology that allows studying protein expression levels and the activation status of cell signalling pathways. It combines high throughput analysis with minimal sample consumption. Using a dedicated arrayer, only 1 ng of proteins extracted from tissues or cell lines are printed onto nitrocellulose covered microscope slides (arrays). Proteins of interest are then quantified using highly specific primary antibodies. Up to one thousand samples can be analysed simultaneously on the same array. We systematically print serial dilutions and replicates for all samples, making the technology highly robust and quantitative.

Our platform has validated more than 640 primary antibodies for RPPA labelling so far, among which almost 150 antibodies against phosphorylated proteins. This panel of antibodies covers the majority of cell signalling pathways. For each project, the project leader selects the antibodies of interest for his/her project among the validated antibodies. New antibodies are continuously being tested in the context of ongoing scientific projects.

Aims

Our overall objective is to provide new biological insight on the protein level, with the ultimate goal to improve the treatment and the outcome of cancer patients. We take part in diverse projects, ranging from fundamental to clinical trials, where we offer a personalized high-throughput proteomic analysis on samples provided by our collaborators, allowing:

- The analysis of protein expression levels

- The analysis of cell signaling pathway activation status and dynamics
- The identification of prognostic or predictive biomarkers
- The identification of potential therapeutic targets
- Classification of samples based on protein profile
- Pharmacodynamics studies

Services

We provide a personalized proteomics analysis on your samples, including the following steps:

- Feedback about the experimental set-up, help with grant applications
- Validation of new primary antibodies
- Sample preparation (lysis, protein concentration measurement)
- Array creation (filling 384 well plates, array printing)
- Array labelling with the antibodies of your choice
- Scanning and quantification of your signal
- Semi-automated quality control
- Data Normalisation
- Exploratory data analysis (PCA and Hierarchical Clustering) and differential data analysis according to your scientific questions
- Assistance during publication writing
- Secured storage of samples and data for at least 3 years

Networking

- Our platform is part of the Global RPPA Society, which comprises the main RPPA laboratories world-wide. This network allows us to remain up-to-date thanks to yearly workshops and to exchange lists of validated antibodies.
- The platform has been involved in 3 European FP7 projects
- Platform supported by the Cancéropôle Ile-de-France
- Service provided to pharmaceutical companies (Roche, Sanofi, Servier, ...)
- Collaboration with the INSERM unit U900 for data management and analysis.

Equipment

We are equipped with:

- an Aushon 2470 arrayer: prints 1ng of each protein extract onto arrays

- a DAKO Autostainer Plus: for automated array labeling
- a Genepix 400B scanner: fluorescent slide scanner
- an Innoscan 710AL (Innopsys): infra-red slide scanner
- a Precellys (Bertin): grinder for tissue lysis
- an EpMotion pipeting robot (Eppendorf) for high-throughput pipeting
- the MicroVigene software (VigeneTech) for signal quantification
- two secured freezers for optimal sample and antibody conservation

Training

The personnel of the platform performs the experiments for all projects. Nevertheless, a training/demonstration can be organized upon request.

Key publications

Year of publication 2017

Biau J., Chautard E., De Koning L., Court F., Pereira B., Verrelle P., Dutreix M. (2017 Jul 1)
Predictive biomarkers of resistance to hypofractionated radiotherapy in high grade glioma
RADIATION ONCOLOGY : 12 : 123 : [DOI : 10.1186/s13014-017-0858-0](https://doi.org/10.1186/s13014-017-0858-0)