AGENCE NATIONALE DE LA RECHERCHE

ANR

Presentation of the funded projects in 2010 for the Blanc International SVSE 1 Programme

Blanc International programme

YEAR 2010

Project title	**Achilles** : Triggering selective apoptosis of multidrug resistant cancer cells overexpressing ABC transporters
Abstract	The aim is to identify compounds targeting multidrug-resistant cancer cells overexpressing ABC transporters for triggering their apoptosis. These compounds, by difference to classical inhibitors of drug-efflux pumps, produce collateral effects by targeting critical components of the pathophysiological matabolism set up by cancer cells in connection to the activity of overexpressed ABC transporter. Our goal is to target the "Achilles'heel" of multidrug-resistant cancer cells. The hungarian group of Gergely Szakacs, in collaboration with the NIH of Bethesda in United States, has discovered compounds targeting cancer cells overerexpressing the Pgp (ABCB1/P-glycoprotein) ABC transporter and triggering their apoptosis, by likely altering metal chelation components. In parallel, our group in Lyon has demonstrated that verapamil selectively triggers the apoptosis of other cancer cells, which overexpress MRP1 (ABCC1), by producing a massive efflux of reduced glutathione (GSH) upon binding to the transporter and stimulating its activity. The objective of the present French-Hungarian application is to transversally collaborate for further characterize these two original approaches of eliminating cancer cells, and extend the "Achylles' heel" concept to cancer cells overexpressing ABCG2 (BCRP). This is especially relevant since ABCG2 shares transport substrates with both Pgp, such as camptothecins, and MRP1, such as methotrexate and ... GSH ! Furthermore, ABCG2 is a stem-cell marker, which supports studying its role in cancer stem cells. We propose to study cytotoxic compounds which are selective for cancer cells overexpressing Pgp, MRP1 or ABCG2, and optimize them in vitro i) by screening with cell-survival and GSH-efflux assays, ii) synthesis of derivatives by medicinal chemistry, and iii) establishment of quantitative structure-activity relationships allowing the construction of a molecular model for designing and synthesizing second-generation compounds with more potency and specificity. Targets identification will allow us to elucidate the molecular and cellular mechanism of action of the compounds in connection to apoptotic signaling. Finally, mice models with xenografted human tumors will allow the evaluation of the in vivo activity, towards reduction of tumor growth, of in vitro-selected

compounds, which will hopefully constitute a newclass of anticancer drugs. The project gathers 4 complementary partner teams: 2 French ones from Lyon (F1, F2), and 2 Hungarian ones from Budapest (H1, H2). Partner 1 (F1) is expert in molecular and cellular biology, biochemistry, medicinal chemistry and biocomputing on ABC transporters, Partner 2 (F2) in cellular signaling and animal experimentation, Partner 3 (H1) in cellular biology related to multidrug ABC transporters, and Partner 4 (H2) is a Biotech company dedicated to establish in vitro cell models and in vivo tumor models. The 3-year proposed project is divided into 4 Tasks/Workpackages. Task 1 is aimed at identifying specific compounds of ABC transporter-overexpressing cells by in silico screening of chemical libraries, and in vitro cell-based screening assays. Task 2 concerns compound optimization by computer-assisted drug design and medicinal chemistry. Task 3 concerns target identification, and investigation of their molecular interaction with compounds and cellular signaling leading to apoptosis. Coordination will be managed by A. Di Pietro and G. Szakacs. Potential valorisation concerns short-term use of the newly-identified targets for diagnostic and prognostic biomedical applications, and long-term development of new anticancer drugs of clinical relevance.

Partners	Institut de Biologie et Chimie des Protéines, Université Lyon 1 Centre Léon Bérard de Lyon, INSERM U590 Creative Cell Ltd, Budapest, Hungary Hungarian Academy of Sciences, Budapest, Hungary
Coordinator	Attilio DI PIETRO – Institut de Biologie et Chimie des Protéines, BM2SI, UMR 5086 CNRS-Université Lyon a.dipietro@ibcp.fr
ANR funding	467 446 €
Starting date and duration	36 mois
Reference	ANR-10-INTB-1101
Cluster label	

Project title	**MULTISCALEFUNIM** : Multiscale functional imaging in the central nervous system
A stract	This project on functional imaging in the central nervous system (CNS) touches on fundamental research, health-related issues and technological development. To understand the functioning principles of the CNS, we need experimental access to its elements, at multiple scales, a multi-scale approach is required for a comprehensive experimental investigation, covering the macro (cm) - meso (mm) - and micro (µm) -scopic levels. Large progress has been made concerning the macro (fMRI, EEG, MEG) and meso-scopic (Optical Imaging) scale in functional imaging, but the microscopic approach can still be largely optimized. In particular, 2-photon microscopic imaging is limited to a 2D scanning and no "real" 3D region of interest selections are possible, which severely limits the accessible structures and/or achievable imaging speeds and thus the scientific questions that can be addressed. Femtonics has developed proprietary technology that overcomes this problems. Yet, the technology is operative currently only in in-vitro thin brain slice preparation. Being able to bring full 3D microscopic technique to in vivo imaging, in particular in large-brain mammals such as rodents and non-human primates is a key stage along a long-term track aiming at intra-operative imaging in human subjects. Moreover, being able to image the activity of small networks in vivo in animal models of human disease will open the door to a better understanding of the pathophysiological mechanisms of debilitating neurological diseases such as epilepsy or neuro-vascular affection. Another long-term prospect is the use of such technology in awake non-human primates to unveil small network dynamics underlying cognitive functions. It shall be noticed that first two-photon imaging studies have been published in cat visual cortex only 4 years ago (Ohki et al., 2005). In this highly competitive research track, there is a risk that European brain research will be rapidly outdated. A collaborative network such as the one proposed here offers one possibility to catch-up in the race for imaging living sub-cortical and cortical tissue during functional tasks. Over the three years project, the core objective of the Hungarian-French collaboration is therefore to adapt this technology, first to whole spinal cord preparation imaging, then to in-vivo rodent and finally anesthetized non-human primate brain imaging. In France, we shall then use it to pursue our scientific questions, both at the fundamental and translational levels (see below). On the hungarian side, the goal of consortium is to push the technological developments up to applications to human brain surgery, the long term objective being to reach the technological challenge of performing laser micro-surgery down to the single cell level, selectively. A central

	challenge that will be tackled is to adapt custom-developed cutting edge 2-photon microscope technology for understanding the functioning of the CNS in-vivo, and to integrate it with meso-scopic functional imaging mehods (optical imaging of intrinsic and voltage dye sygnals) already existing on-site. Therefore, using the french team's expertise in in-vivo imaging, the hungarian partners will first realize a new microscope prototype, capable of performing in vivo imaging, starting from the current slice-preparation model. Next, the french ream will use this tool for a detailed functional exploration of the central nervous system Here a set of more physiology-oriented tasks are foreseen, to be carried out (mostly) by the French teams.
Partners	CNRS DR 12 Délégation Provence et Corse/ Institut de Neurosciences Cognitives de la Méditerranée CNRS DR 12 Délégation Provence et Corse/ Plasticité et Physio-Pathologie de la Motricité Inserm UMR_S 751 Epilepsie & Cognition Femtonics Research and Development Ltd., Hungary Institute for Psychology of the Hungarian Academy of Sciences National Institute of Neuroscience, Hungary
Coordinator	Ivo VANZETTA– CNRS DR 12 Délégation Provence et Corse/ Institut de Neurosciences Cognitives de la Méditerranée ivo.vanzetta@incm.cnrs-mrs.fr
ANR funding	750 000 €
Starting date and duration	36 mois
Reference	ANR-10-INTB-1102
Cluster label	